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Genetic effects of radiofrequency radiation (RFR)

Analysis of these publications shows that there are more papers reporting effects than no effect. With E representing a biological effect, and NE representing no biological effects, the recent literature finds **RFR-genetic effects at: E=306 publications (70%); NE=134 publications (30%). Gene expression studies: 'effect' =99 (79%); 'no effect' =27 (21%)**

Below is a key to abbreviations used throughout the following list of abstracts and serve as my comments to help the reader quickly identify the significance of each experiment. The summary sentences by each author are underlined.

(E- Effect observed; NE- no effect observed) (VT-in vitro study; VO- in vivo study; HU- human study; LE- long term/repeated exposure; AE- acute exposure; LI- low intensity; GT- genotoxic effect, e.g., DNA damage, micronucleus formation, chromosome alterations; GE- gene expression; OX- oxidative effects, i.e., involvement of free radicals and oxidative enzymes; IX- interaction with other factors to cause genetic effects; DE- effects on developing animals; RP- reproduction, e.g., sperm damage; WS- waveform specific effect, e.g., modulation and frequency; CS- cell type specific effect; EP-epigenetic effect).

Research on genetic effects of radiofrequency radiation

(NE) Agarwal A, Desai NR, Makker K, Varghese A, Mouradi R, Sabanegh E, Sharma R. Effects of radiofrequency electromagnetic waves (RF-EMW) from cellular phones on human ejaculated semen: an in vitro pilot study. Fertil Steril 92: 1318-1325, 2009. (VT, HU, AE, GT, RP, OX)

OBJECTIVE: To evaluate effects of cellular phone radiofrequency electromagnetic waves (RF-EMW) during talk mode on unprocessed (neat) ejaculated human semen. DESIGN: Prospective pilot study. SETTING: Center for reproductive medicine laboratory in tertiary hospital setting. SAMPLES: Neat semen samples from normal healthy donors (n = 23) and infertile patients (n = 9). INTERVENTION(S): After liquefaction, neat semen samples were divided into two aliquots. One aliquot (experimental) from each patient was exposed to cellular phone radiation (in talk mode) for 1 h, and the second aliquot (unexposed) served as the control sample under identical conditions. MAIN OUTCOME MEASURE(S): Evaluation of sperm parameters (motility, viability), reactive oxygen species (ROS), total antioxidant capacity (TAC) of semen, ROS-TAC score, and sperm DNA damage. RESULT(S): Samples exposed to RF-EMW showed a significant decrease in sperm motility and viability, increase in ROS level, and decrease in ROS-TAC score. Levels of TAC and DNA damage showed no significant differences from the unexposed group. CONCLUSION(S): Radiofrequency electromagnetic waves emitted from cell phones may lead to oxidative stress in human semen. We speculate that keeping the cell phone in a trouser pocket in talk mode may negatively affect spermatozoa and impair male fertility.

(E)Aitken RJ, Bennetts LE, Sawyer D, Wiklendt AM, King BV. Impact of radio frequency electromagnetic radiation on DNA integrity in the male germline. Inter J Androl 28:171-179, 2005. (VO, LE, LI, GT, RP)

Concern has arisen over human exposures to radio frequency electromagnetic radiation (RFEMR), including a recent report indicating that regular mobile phone use can negatively impact upon human semen quality. These effects would be particularly serious if the biological effects of RFEMR included the induction of DNA damage in male germ cells. In this study, mice were exposed to 900 MHz RFEMR at a specific absorption rate of approximately 90 mW/kg inside a waveguide for 7 days at 12 h per day. Following exposure, DNA damage to caudal epididymal spermatozoa was assessed by quantitative PCR (QPCR) as well as alkaline and pulsed-field gel electrophoresis. The treated mice were overtly normal and all assessment criteria, including sperm number, morphology and vitality were not significantly affected. Gel electrophoresis revealed no gross evidence of increased single- or double-DNA strand breakage in spermatozoa taken from treated animals. However, a detailed analysis of DNA integrity using QPCR revealed statistically significant damage to both the mitochondrial genome ($p < 0.05$) and the nuclear β-globin locus ($p < 0.01$). This study suggests that while RFEMR does not have a dramatic impact on male germ cell development, a significant genotoxic effect on epididymal spermatozoa is evident and deserves further investigation.

(E) Akdag MZ, Dasdag S, Canturk F, Karabulut D, Caner Y, Adalier N. Does prolonged radiofrequency radiation emitted from Wi-Fi devices induce DNA damage in various tissues of rats? J Chem Neuroanat. 75(Pt B):116-122, 2016. (VO, LE, GT, RP)

Wireless internet (Wi-Fi) providers have become essential in our daily lives, as wireless technology is evolving at a dizzying pace. Although there are different frequency generators, one of the most commonly used Wi-Fi devices are 2.4GHz frequency generators. These devices are heavily used in all areas of life but the effect of radiofrequency (RF) radiation emission on users is generally ignored. Yet, an increasing share of the public expresses concern on this issue. Therefore, this study intends to respond to the growing public concern. The purpose of this study is to reveal whether long term exposure of 2.4GHz frequency RF radiation will cause DNA damage of different tissues such as brain, kidney, liver, and skin tissue and testicular tissues of rats. The study was conducted on 16 adult male Wistar-Albino rats. The rats in the experimental group (n=8) were exposed to 2.4GHz frequency radiation for over a year. The rats in the sham control group (n=8) were subjected to the same experimental conditions except the Wi-Fi generator was turned off. After the exposure period was complete the possible DNA damage on the rat's brain, liver, kidney, skin, and testicular tissues was detected through the single cell gel electrophoresis assay (comet) method. The amount of DNA damage was measured as percentage tail DNA value. Based on the DNA damage results determined by the single cell gel electrophoresis (Comet) method, it was found that the% tail DNA values of the brain, kidney, liver, and skin tissues of the rats in the experimental group increased more than those in the control group. The increase of the DNA damage in all tissues was not significant ($p>0.05$).

However the increase of the DNA damage in rat testes tissue was significant ($p<0.01$). In conclusion, long-term exposure to 2.4GHz RF radiation (Wi-Fi) does not cause DNA damage of the organs investigated in this study except testes. The results of this study indicated that testes are more sensitive organ to RF radiation.

(E) Akdag M, Dasdag S, Canturk F, Akdag MZ. Exposure to non-ionizing electromagnetic fields emitted from mobile phones induced DNA damage in human ear canal hair follicle cells. Electromagn Biol Med. 37(2):66-75, 2018. (HU, LE, GT)

The aim of this study was to investigate effect of radiofrequency radiation (RFR) emitted from mobile phones on DNA damage in follicle cells of hair in the ear canal. The study was carried out on 56 men (age range: 30-60 years old)in four treatment groups with n = 14 in each group. The groups were defined as follows: people who did not use a mobile phone (Control), people use mobile phones for 0-30 min/day (second group), people use mobile phones for 30-60 min/day (third group) and people use mobile phones for more than 60 min/day (fourth group). Ear canal hair follicle cells taken from the subjects were analyzed by the Comet Assay to determine DNA damages. The Comet Assay parameters measured were head length, tail length, comet length, percentage of head DNA, tail DNA percentage, tail moment, and Olive tail moment. Results of the study showed that DNA damage indicators were higher in the RFR exposure groups than in the control subjects. In addition, DNA damage increased with the daily duration of exposure. In conclusion, RFR emitted from mobile phones has a potential to produce DNA damage in follicle cells of hair in the ear canal. Therefore, mobile phone users have to pay more attention when using wireless phones.

(E) Akhavan-Sigari R, Baf MM, Ariabod V, Rohde V, Rahighi S. Connection between cell phone use, p53 gene expression in different zones of glioblastoma multiforme and survival prognoses. Rare Tumors. 6(3):5350, 2014. (HU, LE, GE)

The aim of this paper is to investigate p53 gene expression in the central and peripheral zones of glioblastoma multiforme using a real-time reverse transcription polymerase chain reaction (RT-PCR) technique in patients who use cell phones ≥3 hours a day and determine its relationship to clinicopathological findings and overall survival. Sixty-three patients (38 males and 25 females), diagnosed with glioblastoma multiforme (GBM), underwent tumor resection between 2008 and 2011. Patient ages ranged from 25 to 88 years, with a mean age of 55. The levels of expression of p53 in the central and peripheral zone of the GBM were quantified by RT-PCR. Data on p53 gene expression from the central and peripheral zone, the related malignancy and the clinicopathalogical findings (age, gender, tumor location and size), as well as overall survival, were analyzed. Forty-one out of 63 patients (65%) with the highest level of cell phone use (≥3 hours/day) had higher mutant type p53 expression in the peripheral zone of the glioblastoma; the difference was statistically significant (P=0.034). Results from the present study on the use of mobile phones for ≥3 hours a day show a consistent pattern of increased risk for the mutant type of p53 gene expression in the peripheral zone of the glioblastoma, and that this increase was significantly correlated with shorter overall survival time. The risk was not higher for ipsilateral

exposure. We found that the mutant type of p53 gene expression in the peripheral zone of the glioblastoma was increased in 65% of patients using cell phones ≥3 hours a day.

(E) Albaqami M, Hammad M, Pooam M, Procopio M, Sameti M, Ritz T, Ahmad M, Martino CF.. Arabidopsis cryptochrome is responsive to Radiofrequency (RF) electromagnetic fields. Sci Rep 10(1):11260, 2020. (VO, AE, GE)

How living systems respond to weak electromagnetic fields represents one of the major unsolved challenges in sensory biology. Recent evidence has implicated cryptochrome, an evolutionarily conserved flavoprotein receptor, in magnetic field responses of organisms ranging from plants to migratory birds. However, whether cryptochromes fulfill the criteria to function as biological magnetosensors remains to be established. Currently, theoretical predictions on the underlying mechanism of chemical magnetoreception have been supported by experimental observations that exposure to radiofrequency (RF) in the MHz range disrupt bird orientation and mammalian cellular respiration. Here we show that, in keeping with certain quantum physical hypotheses, a weak 7 MHz radiofrequency magnetic field significantly reduces the biological responsivity to blue light of the cryptochrome receptor cry1 in Arabidopsis seedlings. Using an in vivo phosphorylation assay that specifically detects activated cryptochrome, we demonstrate that RF exposure reduces conformational changes associated with biological activity. RF exposure furthermore alters cryptochrome-dependent plant growth responses and gene expression to a degree consistent with theoretical predictions. To our knowledge this represents the first demonstration of a biological receptor responding to RF exposure, providing important new implications for magnetosensing as well as possible future applications in biotechnology and medicine.

(E)Alkis ME, Bilgin HM, Akpolat V, Dasdag S, Yegin K, Yavas MC, Akdag MZ. Effect of 900-1800-, and 2100-MHz radiofrequency radiation on DNA and oxidative stress in brain. Electromagn Biol Med. 38(1):32-47, 2019a. (VO, LE, LI, GT, OX)

Ubiquitous and ever increasing use of mobile phones led to the growing concern about the effects of radiofrequency radiation (RFR) emitted by cell phones on biological systems. The aim of this study is to explore whether long-term RFR exposure at different frequencies affects DNA damage and oxidant-antioxidant parameters in the blood and brain tissue of rats. 28 male Sprague Dawley rats were randomly divided into four equal groups (n = 7). They were identified as Group 1: sham-control, Group 2: 900 MHz, Group 3: 1800 MHz, and Group 4: 2100 MHz. Experimental groups of rats were exposed to RFR 2 h/day for 6 months. The sham-control group of rats was subjected to the same experimental condition but generator was turned off. Specific absorption rates (SARs) at brain with 1 g average were calculated as 0.0845 W/kg, 0.04563 W/kg, and 0.03957, at 900 MHz, 1800 MHz, and 2100 MHz, respectively. Additionally, malondialdehyde (MDA), 8-hydroxydeoxyguanosine (8-OHdG), total antioxidant status (TAS), and total oxidant status (TOS) analyses were conducted in the brain tissue samples. Results of the study showed that DNA damage and oxidative stress indicators were found higher in the RFR exposure groups than in the sham-control group. In conclusion, 900-, 1800-, and 2100-MHz RFR emitted from mobile phones may cause oxidative damage, induce increase in lipid

peroxidation, and increase oxidative DNA damage formation in the frontal lobe of the rat brain tissues. Furthermore, 2100-MHz RFR may cause formation of DNA single-strand breaks.

(E) Alkis MS, Akdag MZ, Dasdag S, Yegin K, Akpolat V. Single-strand DNA breaks and oxidative changes in rat testes exposed to radiofrequency radiation emitted from cellular phones, Biotechnology & Biotechnological Equipment, 33:1, 1733-1740, 2019b. (VO, LE, GT, OX)

The testes are a sensitive organ to electromagnetic pollution and people are concerned about the harmful effects of the radiofrequency radiation (RFR) emitted from cellular phones. Therefore, the purpose of this study was to investigate the effects of long-term exposure to different RFR frequencies on single-strand DNA breaks and oxidative changes in rat testicular tissue. Twenty-eight male Sprague–Dawley rats were divided randomly into four groups. Three groups were exposed to radiation emitted from 900, 1800 and 2100 MHz RF generators, 2 h/day for 6 months. The sham-control group was kept under the same experimental conditions but the RFR generator was turned off. Immediately after the last exposure, testes were removed and DNA damage, 8-hydroxydeoxyguanosine (8-OHdG), malondialdehyde (MDA), total antioxidant status (TAS), total oxidant status (TOS) and oxidative stress index (OSI) were analyzed. The results of this study indicated that RFR increased TOS, OSI, MDA and 8-OHdG ($p < 0.05$). TAS levels in the exposed group were lower than in the sham group ($p < 0.05$). In terms of DNA damage, the tail intensities in the comet assay were higher in the exposure groups ($p < 0.05$). This study demonstrated that long-term exposure to RFR emitted by cellular phones may cause oxidative stress and oxidative DNA damage in rat testicular tissue and may generate DNA single-strand breaks at high frequencies (1800 and 2100 MHz). Our results showed that some RFR emitted from cellular phones has potential to lead to cell damage in the testes.

(E) Alkis ME, Akdag MZ, Dasdag S. Effects of Low-Intensity Microwave Radiation on Oxidant-Antioxidant Parameters and DNA Damage in the Liver of Rats**.** Bioelectromagnetics 42(1):76-85, 2021.(VO,LE, OX, GT)

The continuously increasing usage of cell phones has raised concerns about the adverse effects of microwave radiation (MWR) emitted by cell phones on health. Several in vitro and in vivo studies have claimed that MWR may cause various kinds of damage in tissues. The aim of this study is to examine the possible effects of exposure to low-intensity MWR on DNA and oxidative damage in the livers of rats. Eighteen Sprague-Dawley male rats were divided into three equal groups randomly (n = 6). Group 1 (Sham-control): rats were kept under conditions the same as those of other groups, except for MWR exposure. Group 2: rats exposed to 1800 MHz (SAR: 0.62 W/kg) at 0.127 ± 0.04 mW/cm^2 power density, and Group 3: rats exposed to 2,100 MHz (SAR: 0.2 W/kg) at 0.038 ± 0.03 mW/cm^2 power density. Microwave application groups were exposed to MWR 2 h/day for 7 months. At the end of the exposure period, the rats were sacrificed and DNA damage, malondialdehyde (MDA), 8-hydroxydeoxyguanosine (8-OHdG), and total oxidant-antioxidant parameter analyses were conducted in their liver tissue samples. It was found that 1800 and 2100 MHz low-intensity MWR caused a significant increase in MDA, 8-OHdG, total oxidant status, oxidative stress index, and comet assay tail intensity (P <

0.05), while total antioxidant status levels ($P < 0.05$) decreased. The results of our study showed that whole-body exposure to 1800 and 2100 MHz low-intensity MWR emitted by cell phones can induce oxidative stress by altering oxidant-antioxidant parameters and lead to DNA strand breaks and oxidative DNA damage in the liver of rats.

(NE) Al-Serori H, Kundi M, Ferk F, Mišík M, Nersesyan A, Murbach M, Lah TT, Knasmüller S. Evaluation of the potential of mobile phone specific electromagnetic fields (UMTS) to produce micronuclei in human glioblastoma cell lines. Toxicol In Vitro. 40:264-271, 2017. (VT, E, GT)

Some epidemiological studies indicate that mobile phones cause glioblastomas in humans. Since it is known that genomic instability plays a key role in the etiology of cancer, we investigated the effects of the universal mobile telecommunications system radiofrequency (UMTS-RF) signal, which is used in "smart" phones, on micronucleus (MN) formation and other anomalies such as nuclear buds (NBUDs) and nucleoplasmatic bridges (NPBs). MN are formed by structural and numerical aberrations, NBs reflect gene amplification and NPBs are formed from dicentric chromosomes. The experiments were conducted with human glioblastoma cell lines, which differ in regard to their p53 status, namely U87 (wild-type) and U251 (mutated). The cells were cultivated for 16h in presence and absence of fetal calf serum and exposed to different SAR doses (0.25, 0.50 and 1.00W/kg), which reflect the exposure of humans, in presence and absence of mitomycin C as former studies indicate that RF may cause synergistic effects in combination with this drug. We found no evidence for induction of MN and other anomalies. However, with the highest dose, induction of apoptosis was observed in U251 cells on the basis of the morphological features of the cells. Our findings indicate that the UMTS-RF signal does not cause chromosomal damage in glioblastoma cells; the mechanisms which lead to induction of programmed cell death will be investigated in further studies.

(E) Al-Serori H, Ferk F, Kundi M, Bileck A, Gerner C, Mišík M, Nersesyan A, Waldherr M, Murbach M, Lah TT, Herold-Mende C, Collins AR, Knasmüller S. Mobile phone specific electromagnetic fields induce transient DNA damage and nucleotide excision repair in serum-deprived human glioblastoma cells. PLoS One. 13(4):e0193677, 2018. (VT, AE, GT)

Some epidemiological studies indicate that the use of mobile phones causes cancer in humans (in particular glioblastomas). It is known that DNA damage plays a key role in malignant transformation; therefore, we investigated the impact of the UMTS signal which is widely used in mobile telecommunications, on DNA stability in ten different human cell lines (six brain derived cell lines, lymphocytes, fibroblasts, liver and buccal tissue derived cells) under conditions relevant for users (SAR 0.25 to 1.00 W/kg). We found no evidence for induction of damage in single cell gel electrophoresis assays when the cells were cultivated with serum. However, clear positive effects were seen in a p53 proficient glioblastoma line (U87) when the

cells were grown under serum free conditions, while no effects were found in p53 deficient glioblastoma cells (U251). Further experiments showed that the damage disappears rapidly in U87 and that exposure induced nucleotide excision repair (NER) and does not cause double strand breaks (DSBs). The observation of NER induction is supported by results of a proteome analysis indicating that several proteins involved in NER are up-regulated after exposure to UMTS; additionally, we found limited evidence for the activation of the γ-interferon pathway. The present findings show that the signal causes transient genetic instability in glioma derived cells and activates cellular defense systems.

(NE) Antonopoulos A, Eisenbrandt H, Obe G, Effects of high-frequency electromagnetic fields on human lymphocytes in vitro. Mutat Res 395(2-3): 209-214, 1997. (VT, AE, GT)

Human peripheral lymphocytes were incubated in the presence of high-frequency electromagnetic fields of 380, 900 and 1800 MHz. The measured endpoints were cell cycle progression and the frequencies of sister-chromatid exchanges. No differences between treated and control cultures could be found.

(E) Aravinda VSS, Kandregula CR, Muppa R, Krishna MM, Nikitha BS, Yenni M. A cross-sectional and histological analysis to understand the cytological effects of cell phone radiation on buccal mucosa of children. J Indian Soc Pedod Prev Dent 2022;40(1):74-80. (HU, LE, GT)

Context: The ongoing pandemic has affected all the spheres of life and one of the severely affected avenues is the education of a child. The online education has seen an upward curve since the start of COVID-19 pandemic. Schools globally have adopted online class tutorials as the main method to impart education and directly increasing the screen time for a child. **Aim:** The aim of the present study was to evaluate the cytological effects of prolonged mobile phone usage on the buccal mucosa of children. **Settings and design:** Stratified sampling was used for the selection of subjects for the study. After a questionnaire regarding the usage of a mobile phone was distributed among the parents of children. Among them, 90 children were selected on the basis of pattern and frequency of mobile phone usage in the child. **Materials and methodology:** The children were divided into three groups based on the per day hours of viewing of mobile phone, i.e., Group 1: Usage of 1-2 h a day, Group 2: Usage of 3-6 h a day, and Group 3: Usage of >6 h a day. The time frame taken into consideration was 1 year after the pandemic started. This was specifically to understand the impact of the online education. Swab was obtained by using the conventional ice-cream stick method from the buccal mucosa. **Statistical analysis:** The samples were subjected to histological and microscopical analysis to observe for cytological changes. One-way ANOVA was used to determine the statistical significance if any. **Results:** The results obtained clearly showed that Group 3 (>6 h usage per day) showed the highest number of cellular and chromosomal aberrations which was significant. **Conclusion:** The results indicated that impact due to the prolonged screen time on the buccal mucosa is significant. A direct proportionality was seen between the apoptotic changes and chromosomal aberrations and the number of daily hour usage.

(E) Atasoy HI, Gunal MY, Atasoy P, Elgun S, Bugdayci G. Immunohistopathologic demonstration of deleterious effects on growing rat testes of radiofrequency waves emitted from conventional Wi-Fi devices. J Pediatr Urol. 9:223-229, 2013. (VO; LE, GT, OX, RP)

OBJECTIVE: To investigate effects on rat testes of radiofrequency radiation emitted from indoor Wi-Fi Internet access devices using 802.11.g wireless standards. METHODS: Ten Wistar albino male rats were divided into experimental and control groups, with five rats per group. Standard wireless gateways communicating at 2.437 GHz were used as radiofrequency wave sources. The experimental group was exposed to radiofrequency energy for 24 h a day for 20 weeks. The rats were sacrificed at the end of the study. Intracardiac blood was sampled for serum 8-hydroxy-2'-deoxyguanosine levels. Testes were removed and examined histologically and immunohistochemically. Testis tissues were analyzed for malondialdehyde levels and prooxidant-antioxidant enzyme activities. RESULTS: We observed significant increases in serum 8-hydroxy-2'-deoxyguanosine levels and 8-hydroxyguanosine staining in the testes of the experimental group indicating DNA damage due to exposure ($p < 0.05$). We also found decreased levels of catalase and glutathione peroxidase activity in the experimental group, which may have been due to radiofrequency effects on enzyme activity ($p < 0.05$). CONCLUSIONS: These findings raise questions about the safety of radiofrequency exposure from Wi-Fi Internet access devices for growing organisms of reproductive age, with a potential effect on both fertility and the integrity of germ cells.

(E) Atlı Şekeroğlu Z, Akar A, Sekeroğlu V. Evaluation of the cytogenotoxic damage in immature and mature rats exposed to 900 MHz radio frequency electromagnetic fields. Int J Radiat Biol. 89(11):985-992, 2013. (VO, LE, GT, DE)

Purpose: One of the most important issues regarding radio frequency electromagnetic fields (RF-EMF) is their effect on genetic material. Therefore, we investigated the cytogenotoxic effects of 900 MHz radio frequency electromagnetic fields (RF-EMF) and the effect of a recovery period after exposure to RF-EMF on bone marrow cells of immature and mature rats. Materials and methods: The immature and mature rats in treatment groups were exposed to RF-EMF for 2 h/day for 45 days. Average electrical field values for immature and mature rats were 28.1±4.8 V/m and 20.0±3.2 V/m, respectively. Whole-body specific absorption rate (SAR) values for immature and mature rats were in the range of 0.38-0.78 W/kg, and 0.31-0.52 W/kg during the 45 days, respectively. Two recovery groups were kept for 15 days after RF-EMF exposure. Results: Significant differences were observed in chromosome aberrations (CA), micronucleus (MN) frequency, mitotic index (MI) and ratio of polychromatic erythrocytes (PCE) in all treatment and recovery groups. The cytogenotoxic damage in immature rats was statistically higher than the mature rats. The recovery period did not reduce the damage to the same extent as the corresponding control groups. Conclusions: The exposure of RF-EMF leads to cytotoxic and genotoxic damage in immature and mature rats. More sensitive studies are required to elucidate the possible carcinogenic risk of EMF exposure in humans, especially children.

(E) Avendaño C, Mata A, Sanchez Sarmiento CA, Doncel GF. Use of laptop computers connected to internet through Wi-Fi decreases human sperm motility and increases sperm DNA fragmentation. Fertil Steril 97:39-45, 2012. (HU, AE, GT, RP)

OBJECTIVE: To evaluate the effects of laptop computers connected to local area networks wirelessly (Wi-Fi) on human spermatozoa. DESIGN: Prospective in vitro study. SETTING: Center for reproductive medicine. PATIENT(S): Semen samples from 29 healthy donors. INTERVENTION(S): Motile sperm were selected by swim up. Each sperm suspension was divided into two aliquots. One sperm aliquot (experimental) from each patient was exposed to an internet-connected laptop by Wi-Fi for 4 hours, whereas the second aliquot (unexposed) was used as control, incubated under identical conditions without being exposed to the laptop. MAIN OUTCOME MEASURE(S): Evaluation of sperm motility, viability, and DNA fragmentation. RESULT(S): Donor sperm samples, mostly normozoospermic, exposed ex vivo during 4 hours to a wireless internet-connected laptop showed a significant decrease in progressive sperm motility and an increase in sperm DNA fragmentation. Levels of dead sperm showed no significant differences between the two groups. CONCLUSION(S): To our knowledge, this is the first study to evaluate the direct impact of laptop use on human spermatozoa. Ex vivo exposure of human spermatozoa to a wireless internet-connected laptop decreased motility and induced DNA fragmentation by a nonthermal effect. We speculate that keeping a laptop connected wirelessly to the internet on the lap near the testes may result in decreased male fertility. Further in vitro and in vivo studies are needed to prove this contention.

(E)Azimipour F, Zavareh S, Lashkarbolouki T. The Effect of Radiation Emitted by Cell Phone on The Gelatinolytic Activity of Matrix Metalloproteinase-2 and -9 of Mouse Pre-Antral Follicles during In Vitro Culture. Cell J 22(1):1-8, 2020. (VT, AE, GE, RP)

Objective: The unfavorable effects of electromagnetic radiation (EMR) emitted by the cell phone on reproduction health are controversial. Metalloproteinases play a vital role in ovarian follicle development. This study was designed to investigate the effects of exposure to the cell phone on the gelatinolytic activity of *in vitro* cultured mouse pre-antral follicle. **Materials and methods:** In this experimental study, pre-antral follicles were isolated from ovaries of immature mice (n=16) and cultured with or without exposure to the cell phone in talking mode for 60 minutes. The gelatinolytic activity was evaluated through the zymography method, as well as the gene expression of matrix metalloproteinases (MMPs) namely *MMP-2* and -9 and tissue inhibitors of metalloproteinases (TIMPs) namely, *TIMP-1* and -2 by the real-time polymerase chain reaction (PCR) method. Also, in parallel, the development of pre-antral follicles was assessed. **Results:** The maturation parameters of the cell phone-exposed pre-antral follicles were significantly lower compared with the control group ($P<0.05$). The gelatinolytic activity was significantly decreased in the cell phone-exposed preantral follicles compared with the control group ($P<0.05$). The relative mRNA expression of the *MMP-2* gene was significantly ($P<0.05$) increased in the cell phone-exposed pre-antral follicles whereas the expression rate of the MMP-9 gene was considerably ($P<0.05$) reduced when compared with the control group. Conversely, the relative expression of the *TIMP-1* was markedly ($P<0.05$) increased in the cell phone-exposed pre-antral follicles while the expression of the *TIMP-2* was ($P<0.05$) significantly diminished in comparison with the control group. **Conclusion:** Exposure to the cell phone alters the growth and maturation rate of murine ovarian follicle through the changing in the expression of the *MMP-2* and -9 genes, as well as the gelatinolytic activity.

(E) Balakrishnan K, Murali V, Rathika C, Manikandan T, Malini RP, Kumar RA, Krishnan M. Hsp70 is an independent stress marker among frequent users of mobile phones. J Environ Pathol Toxicol Oncol. 33(4):339-347, 2014. (HU, LE, GE)

The aim of this study was to measure the serum concentrations of heat shock protein (HSP) 70 and C-reactive protein (CRP) and the expression levels of the hsp70 gene among frequent users of mobile phones (FUMPs). We enrolled 120 employees of information technology (IT)/IT enabled service companies (FUMPs; IT professionals) and 102 infrequent users of mobile phones (IFUMPs; people from non-IT professions) as controls. The serum concentrations of HSP70 and CRP were measured by enzyme-linked immunosorbant assay and hsp70 gene expression by reverse transcription polymerase chain reaction. Significantly higher concentrations of serum HSP70 ($P < 0.00012$) and CRP ($P < 0.04$) were observed among FUMPs than IFUMPs. A higher level of hsp70 gene expression (fold induction) was observed among FUMPs than IFUMPs ($P < 7.06 \times 10\text{-}13$). In contrast to the duration of exposure-dependent increase of serum concentration of CRP, the serum HSP70 concentration was found to be independent of the duration of exposure to mobile phones. Thus, the study convincingly demonstrated the role of serum HSP and CRP as systemic inflammatory biomarkers for mobile phone-induced radiation.

(E) Balode, Z, Assessment of radio-frequency electromagnetic radiation by the micronucleus test in bovine peripheral erythrocytes. Sci Total Environ 180(1):81-85, 1996. (VO, LE, GT)

Previous bioindicative studies in the Skrunda Radio Location Station area have focused on the somatic influence of electromagnetic radiation on plants, but it is also important to study genetic effects. We have chosen cows as test animals for cytogenetical evaluation because they live in the same general exposure area as humans, are confined to specific locations and are chronically exposed to radiation. Blood samples were obtained from female Latvian Brown cows from a farm close to and in front of the Skrunda Radar and from cows in a control area. A simplified alternative to the Schiff method of DNA staining for identification of micronuclei in peripheral erythrocytes was applied. Microscopically, micronuclei in peripheral blood erythrocytes were round in shape and exhibited a strong red colour. They are easily detectable as the only coloured bodies in the uncoloured erythrocytes. From each individual animal 2000 erythrocytes were examined at a magnification of x 1000 for the presence of micronuclei. The counting of micronuclei in peripheral erythrocytes gave low average incidences, 0.6 per 1000 in the exposed group and 0.1 per 1000 in the control, but statistically significant ($P < 0.01$) differences were found in the frequency distribution between the control and exposed groups.

(E) Baohong Wang, Jiliang H, Lifen J, Deqiang L, Wei Z, Jianlin L, Hongping D. Studying the synergistic damage effects induced by 1.8 GHz radiofrequency field radiation (RFR) with four chemical mutagens on human lymphocyte DNA using comet assay in vitro. Mutat Res 578:149-57, 2005. (VT, AE, GT, IX)

The aim of this investigation was to study the synergistic DNA damage effects in human lymphocytes induced by 1.8GHz radiofrequency field radiation (RFR, SAR of 3W/kg) with four

chemical mutagens, i.e. mitomycin C (MMC, DNA crosslinker), bleomycin (BLM, radiomimetic agent), methyl methanesulfonate (MMS, alkylating agent), and 4-nitroquinoline-1-oxide (4NQO, UV-mimetic agent). The DNA damage of lymphocytes exposed to RFR and/or with chemical mutagens was detected at two incubation time (0 or 21h) after treatment with comet assay in vitro. Three combinative exposure ways were used. Cells were exposed to RFR and chemical mutagens for 2 and 3h, respectively. Tail length (TL) and tail moment (TM) were utilized as DNA damage indexes. The results showed no difference of DNA damage indexes between RFR group and control group at 0 and 21h incubation after exposure (P>0.05). There were significant difference of DNA damage indexes between MMC group and RFR+MMC co-exposure group at 0 and 21h incubation after treatment (P<0.01). Also the significant difference of DNA damage indexes between 4NQO group and RFR+4NQO co-exposure group at 0 and 21h incubation after treatment was observed (P<0.05 or P<0.01). The DNA damage in RFR+BLM co-exposure groups and RFR+MMS co-exposure groups was not significantly increased, as compared with corresponding BLM and MMS groups (P>0.05). The experimental results indicated 1.8GHz RFR (SAR, 3W/kg) for 2h did not induce the human lymphocyte DNA damage effects in vitro, but could enhance the human lymphocyte DNA damage effects induced by MMC and 4NQO. The synergistic DNA damage effects of 1.8GHz RFR with BLM or MMS were not obvious.

(E) Baohong W, Lifen J, Lanjuan L, Jianlin L, Deqiang L, Wei Z, Jiliang H.Evaluating the combinative effects on human lymphocyte DNA damage induced by ultraviolet ray C plus 1.8GHz microwaves using comet assay in vitro. Toxicology. 232(3):311-316, 2007. (VT, AE, GT, IX)

The objective of this study was to observe whether 1.8GHz microwaves (MW) (SAR, 3 W/kg) exposure can influence human lymphocyte DNA damage induced by ultraviolet ray C (UVC). The lymphocytes, which were from three young healthy donors, were exposed to 254 nm UVC at the doses of 0.25, 0.5, 0.75, 1.0, 1.5 and 2.0 J m(-2), respectively. The lymphocytes were irradiated by 1.8GHz MW (SAR, 3 W/kg) for 0, 1.5 and 4 h. The combinative exposure of UVC plus MW was conducted. The treated cells were incubated for 0, 1.5 and 4 h. Finally, comet assay was used to measure DNA damage of above treated lymphocytes. The results indicated that the difference of DNA damage induced between MW group and control group was not significant (P>0.05). The MTLs induced by UVC were 1.71+/-0.09, 2.02+/-0.08, 2.27+/-0.17, 2.27+/-0.06, 2.25+/-0.12, 2.24+/-0.11 microm, respectively, which were significantly higher than that (0.96+/-0.05 microm) of control (P<0.01). MTLs of some sub-groups in combinative exposure groups at 1.5-h incubation were significantly lower that those of corresponding UVC sub-groups (P<0.01 or P<0.05). However, MTLs of some sub-groups in combinative exposure groups at 4-h incubation were significantly higher that those of corresponding UVC sub-groups (P<0.01 or P<0.05). In this experiment it was found that 1.8GHz (SAR, 3 W/kg) MW exposure for 1.5 and 4 h did not enhance significantly human lymphocyte DNA damage, but could reduce and increase DNA damage of human lymphocytes induced by UVC at 1.5-h and 4-h incubation, respectively.

(E) Banerjee S, Singh NN, Sreedhar G, Mukherjee S. Analysis of the genotoxic effects of mobile phone radiation using buccal micronucleus assay: a comparative evaluation. J Clin Diagn Res 10(3):ZC82-85, 2016. (HU, LE, GT)

Introduction: Micronucleus (MN) is considered to be a reliable marker for genotoxic damage and it determines the presence and the extent of the chromosomal damage. The MN is formed due to DNA damage or chromosomal disarrangements. The MN has a close association with cancer incidences. In the new era, mobile phones are constantly gaining popularity specifically in the young generation, but this device uses radiofrequency radiation that may have a possible carcinogenic effect. The available reports related to the carcinogenic effect of mobile radiation on oral mucosa are contradictory. **Aim:** To explore the effects of mobile phone radiation on the MN frequency in oral mucosal cells. **Materials and methods:** The subjects were divided into two major groups: low mobile phone users and high mobile phone users. Subjects who used their mobile phone since less than five years and less than three hours a week comprised of the first group and those who used their mobile since more than five years and more than 10 hours a week comprised of the second group. Net surfing and text messaging was not considered in this study. Exfoliated buccal mucosal cells were collected from both the groups and the cells were stained with DNA-specific stain acridine orange. Thousand exfoliated buccal mucosal cells were screened and the cells which were positive for micronuclei were counted. The micronucleus frequency was represented as mean±SD, and unpaired Student t-test was used for intergroup comparisons. **Results:** The number of micronucleated cells/ 1000 exfoliated buccal mucosal cells was found to be significantly increased in high mobile phone users group than the low mobile phone users group. The use of mobile phone with the associated complaint of warmth around the ear showed a maximum increase in the number of micronucleated cells /1000 exfoliated buccal mucosal cells. **Conclusion:** Mobile phone radiation even in the permissible range when used for longer duration causes significant genotoxicity. The genotoxicity can be avoided to some extent by the regular use of headphones.

(E) Beaubois E, Girard S, Lallechere S, Davies E, Paladian F, Bonnet P, Ledoigt G, Vian A. Intercellular communication in plants: evidence for two rapidly transmitted systemic signals generated in response to electromagnetic field stimulation in tomato. Plant Cell Environ 30(7):834-844. 2007. (VO, AE, GE, LI)

Exposing all of a wild-type tomato plant to electromagnetic radiation evoked rapid and substantial accumulation of basic leucine-zipper transcription factor (bZIP) mRNA in the terminal leaf (#4) with kinetics very similar to that seen in response to wounding, while in the abscisic acid (ABA) mutant (Sitiens), the response was more rapid, but transient. Submitting just the oldest leaf (#1) of a wild-type plant to irradiation evoked bZIP mRNA accumulation both locally in the exposed leaf and systemically in the unexposed (distant) leaf #4, although systemic accumulation was delayed somewhat. Accumulation of Pin2 mRNA was less than bZIP in both the exposed and distant leaves in wild type, but there was no delay in the systemic response. In Sitiens, bZIP mRNA accumulation was far less than in wild type in both local and distant leaves, while Pin2 mRNA accumulation was stronger in the exposed leaf, but totally prevented in the systemic leaf. In the jasmonic acid (JA) mutant (JL-5) and in wild-type plants treated with the ABA biosynthesis inhibitor, naproxen, responses were similar to those in the ABA mutant, while treatment of the exposed leaf with calcium antagonists totally abolished both local and systemic increases in bZIP transcript accumulation.

(E) Bektas H, Dasdag S, Bektas MS. Comparison of effects of 2.4 GHz Wi-Fi and mobile phone exposure on human placenta and cord blood. Biotechn Biotechnological Equipment, 34:1, 154-162, 2020.(HU, CE, OX, GT)

The aim of this study was to investigate the effects of radiofrequency radiation emitted from Wi-Fi systems and mobile phones on cord blood and placenta. The study included 149 pregnant women who were divided in subgroups: unexposed (control), mobile phone exposed, Wi-Fi exposed and mobile phone plus Wi-Fi exposed groups. Immediately after birth, placenta and cord blood samples were collected and protein carbonyl (PCO), malondialdehyde (MDA), total oxidant status (TOS), total antioxidant status, 8-hydroxy-20-deoxyguanosine (8-OHdG) levels and DNA single strand breaks were analysed. The results of the study showed an increase in 8-OHdG, MDA, PCO and TOS in cord blood and placenta in the group exposed to mobile phones during gestation. However, the group exposed to Wi-Fi did not show alterations in the studied oxidative stress parameters. On the other hand, tail intensity and tail moment of DNA in the mobile phone exposure groups were higher than those in the control and Wi-Fi exposure groups. In conclusion, the results of this study indicated that mobile phone exposure during pregnancy could have an important potential to cause oxidative stress and DNA damage in cord blood and placenta. The results of this study also indicated that combined effects of Wi-Fi plus mobile phone exposure have a higher potential to cause synergistic harmful effects.

(E) Belyaev IYa, Alipov YD, Shcheglov VS, Lystsov VN, Resonance effect of microwaves on the genome conformational state of E. coli cells. Z Naturforsch [C] 47(7-8):621-627,1992. (VT, AE, LI, GT)

The effect of low intensity microwaves on the conformational state of the genome of X-irradiated E. coli cells was studied by the method of viscosity anomalous time dependencies. It has been established that within the ranges of 51.62-51.84 GHz and 41.25-41.50 GHz the frequency dependence of the observed effect has a resonance nature with a resonance half-width of the order of 100 MHz. The power dependence of the microwave effect within the range of 0.1-200 microW/cm^2 has shown that a power density of 1 microW/cm2 is sufficient to suppress radiation-induced repair of the genome conformational state. The effect of microwave suppression of repair is well reproduced and does not depend on the sequence of cell exposure to X-rays and microwave radiation in the millimeter band. The results obtained indicate the role of the cell genome in the resonant interaction of cells with low intensity millimeter waves

(E) Belyaev IY, Hillert L, Protopopova M, Tamm C, Malmgren LO, Persson BR, Selivanova G, Harms-Ringdahl M. 915 MHz microwaves and 50 Hz magnetic field affect chromatin conformation and 53BP1 foci in human lymphocytes from hypersensitive and healthy persons. Bioelectromagnetics 26:173-184, 2005. (VT, AE, LI, GT)

We used exposure to microwaves from a global system for mobile communication (GSM) mobile phone (915 MHz, specific absorption rate (SAR) 37 mW/kg) and power frequency magnetic field (50 Hz, 15 muT peak value) to investigate the response of lymphocytes from healthy subjects and from persons reporting hypersensitivity to electromagnetic field (EMF). The hypersensitive and healthy donors were matched by gender and age and the data were analyzed

blind to treatment condition. The changes in chromatin conformation were measured with the method of anomalous viscosity time dependencies (AVTD). 53BP1 protein, which has been shown to colocalize in foci with DNA double strand breaks (DSBs), was analyzed by immunostaining in situ. Exposure at room temperature to either 915 MHz or 50 Hz resulted in significant condensation of chromatin, shown as AVTD changes, which was similar to the effect of heat shock at 41 degrees C. No significant differences in responses between normal and hypersensitive subjects were detected. Neither 915 MHz nor 50 Hz exposure induced 53BP1 foci. On the contrary, a distinct decrease in background level of 53BP1 signaling was observed upon these exposures as well as after heat shock treatments. This decrease correlated with the AVTD data and may indicate decrease in accessibility of 53BP1 to antibodies because of stress-induced chromatin condensation. Apoptosis was determined by morphological changes and by apoptotic fragmentation of DNA as analyzed by pulsed-field gel electrophoresis (PFGE). No apoptosis was induced by exposure to 50 Hz and 915 MHz microwaves. In conclusion, 50 Hz magnetic field and 915 MHz microwaves under specified conditions of exposure induced comparable responses in lymphocytes from healthy and hypersensitive donors that were similar but not identical to stress response induced by heat shock.

(E) Belyaev IY, Koch CB, Terenius O, Roxstromto 915 MHz GSM microwaves induces changes in gene expression but not double stranded DNA breaks or effects on chromatin conformation. Bioelectromagnetics 27:295-306, 2006. (VO, AE, GE)

We investigated whether exposure of rat brain to microwaves (MWs) of global system for mobile communication (GSM) induces DNA breaks, changes in chromatin conformation and in gene expression. An exposure installation was used based on a test mobile phone employing a GSM signal at 915 MHz, all standard modulations included, output power level in pulses 2 W, specific absorption rate (SAR) 0.4 mW/g. Rats were exposed or sham exposed to MWs during 2 h. After exposure, cell suspensions were prepared from brain samples, as well as from spleen and thymus. For analysis of gene expression patterns, total RNA was extracted from cerebellum. Changes in chromatin conformation, which are indicative of stress response and genotoxic effects, were measured by the method of anomalous viscosity time dependencies (AVTD). DNA double strand breaks (DSBs) were analyzed by pulsed-field gel electrophoresis (PFGE). Effects of MW exposure were observed on neither conformation of chromatin nor DNA DSBs. Gene expression profiles were obtained by Affymetrix U34 GeneChips representing 8800 rat genes and analyzed with the Affymetrix Microarray Suite (MAS) 5.0 software. In cerebellum from all exposed animals, 11 genes were upregulated in a range of 1.34-2.74 fold and one gene was downregulated 0.48-fold ($P < .0025$). The induced genes encode proteins with diverse functions including neurotransmitter regulation, blood-brain barrier (BBB), and melatonin production. The data shows that GSM MWs at 915 MHz did not induce PFGE-detectable DNA double stranded breaks or changes in chromatin conformation, but affected expression of genes in rat brain cells

(E) Belyaev IY, Markovà E, Hillert L, Malmgren LO, Persson BR. Microwaves from UMTS/GSM mobile phones induce long-lasting inhibition of 53BP1/gamma-H2AX DNA repair foci in human lymphocytes. Bioelectromagnetics 30:129-141, 2009. (VT, AE, LI, GT, WS)

We have recently described frequency-dependent effects of mobile phone microwaves (MWs) of global system for mobile communication (GSM) on human lymphocytes from persons reporting hypersensitivity to electromagnetic fields and healthy persons. Contrary to GSM, universal global telecommunications system (UMTS) mobile phones emit wide-band MW signals. Hypothetically, UMTS MWs may result in higher biological effects compared to GSM signal because of eventual "effective" frequencies within the wideband. Here, we report for the first time that UMTS MWs affect chromatin and inhibit formation of DNA double-strand breaks co-localizing 53BP1/gamma-H2AX DNA repair foci in human lymphocytes from hypersensitive and healthy persons and confirm that effects of GSM MWs depend on carrier frequency. Remarkably, the effects of MWs on 53BP1/gamma-H2AX foci persisted up to 72 h following exposure of cells, even longer than the stress response following heat shock. The data are in line with the hypothesis that the type of signal, UMTS MWs, may have higher biological efficiency and possibly larger health risk effects compared to GSM radiation emissions. No significant differences in effects between groups of healthy and hypersensitive subjects were observed, except for the effects of UMTS MWs and GSM-915 MHz MWs on the formation of the DNA repair foci, which were different for hypersensitive ($P < 0.02$[53BP1]//0.01[gamma-H2AX]) but not for control subjects ($P > 0.05$). The non-parametric statistics used here did not indicate specificity of the differences revealed between the effects of GSM and UMTS MWs on cells from hypersensitive subjects and more data are needed to study the nature of these differences.

Bhargav H, Srinivasan TM, Varambally S, Gangadhar BN, Koka P. Effect of mobile phone-induced electromagnetic field on brain hemodynamics and human stem cell functioning: possible mechanistic link to cancer risk and early diagnostic value of electronphotonic imaging. J Stem Cells. 10(4):287-294, 2015. (review)

The mobile phones (MP) are low power radio devices which work on electromagnetic fields (EMFs), in the frequency range of 900-1800 MHz. Exposure to MPEMFs may affect brain physiology and lead to various health hazards including brain tumors. Earlier studies with positron emission tomography (PET) have found alterations in cerebral blood flow (CBF) after acute exposure to MPEMFs. It is widely accepted that DNA double-strand breaks (DSBs) and their misrepair in stem cells are critical events in the multistage origination of various leukemia and tumors, including brain tumors such as gliomas. Both significant misbalance in DSB repair and severe stress response have been triggered by MPEMFs and EMFs from cell towers. It has been shown that stem cells are most sensitive to microwave exposure and react to more frequencies than do differentiated cells. This may be important for cancer risk assessment and indicates that stem cells are the most relevant cellular model for validating safe mobile communication signals. Recently developed technology for recording the human bio-electromagnetic (BEM) field using Electron photonic Imaging (EPI) or Gas Discharge Visualisation (GDV) technique provides useful information about the human BEM. Studies have recorded acute effects of Mobile Phone Electromagnetic Fields (MPEMFs) using EPI and found quantifiable effects on human BEM field. Present manuscript reviews evidences of altered brain physiology and stem cell functioning due to mobile phone/cell tower radiations, its association with increased cancer risk and explores early diagnostic value of EPI imaging in detecting EMF induced changes on human BEM.

(NE) Bisht KS, Moros EG, Straube WL, Baty JD, Roti Roti JL, The Effect of 835.62 MHz FDMA or 847.74 MHz CDMA modulated radiofrequency radiation on the induction of micronuclei in C3H 10T½ cells. Radiat. Res. 157: 506–515, 2002.(VT, AE, GT)

To determine if radiofrequency (RF) radiation induces the formation of micronuclei, C3H 10T½ cells were exposed to 835.62 MHz frequency division multiple access (FDMA) or 847.74 MHz code division multiple access (CDMA) modulated RF radiation. After the exposure to RF radiation, the micronucleus assay was performed by the cytokinesis block method using cytochalasin B treatment. The micronuclei appearing after mitosis were scored in binucleated cells using acridine orange staining. The frequency of micronuclei was scored both as the percentage of binucleated cells with micronuclei and as the number of micronuclei per 100 binucleated cells. Treatment of cells with cytochalasin B at a concentration of 2 μg/ml for 22 h was found to yield the maximum number of binucleated cells in C3H 10T½ cells. The method used for the micronucleus assay in the present study detected a highly significant dose response for both indices of micronucleus production in the dose range of 0.1–1.2 Gy and it was sensitive enough to detect a significant ($P > 0.05$) increase in micronuclei after doses of 0.3 Gy in exponentially growing cells and after 0.9 Gy in plateau-phase cells. Exponentially growing cells or plateau-phase cells were exposed to CDMA (3.2 or 4.8 W/kg) or FDMA (3.2 or 5.1 W/kg) RF radiation for 3, 8, 16 or 24 h. In three repeat experiments, no exposure condition was found by analysis of variance to result in a significant increase relative to sham-exposed cells either in the percentage of binucleated cells with micronuclei or in the number of micronuclei per 100 binucleated cells. In this study, data from cells exposed to different RF signals at two SARs were compared to a common sham-exposed sample. We used the Dunnett's test, which is specifically designed for this purpose, and found no significant exposure-related differences for either plateau-phase cells or exponentially growing cells. Thus the results of this study are not consistent with the possibility that these RF radiations induce micronuclei.

(E) Bourdineaud JP, Šrut M, Štambuk A, Tkalec M, Brèthes D, Malarić K, Klobučar GIV. Electromagnetic fields at a mobile phone frequency (900 MHz) trigger the onset of general stress response along with DNA modifications in Eisenia fetida earthworms. Arh Hig Rada Toksikol. 68(2):142-152, 2017. (VO, AE, LI, GT, OX)

Eisenia fetida earthworms were exposed to electromagnetic field (EMF) at a mobile phone frequency (900 MHz) and at field levels ranging from 10 to 120 V m-1 for a period of two hours (corresponding to specific absorption rates ranging from 0.13 to 9.33 mW kg-1). Potential effects of longer exposure (four hours), field modulation, and a recovery period of 24 h after two hours of exposure were addressed at the field level of 23 V m-1. All exposure treatments induced significant DNA modifications as assessed by a quantitative random amplified polymorphic DNA-PCR. Even after 24 h of recovery following a two hour-exposure, the number of probe hybridisation sites displayed a significant two-fold decrease as compared to untreated control earthworms, implying a loss of hybridisation sites and a persistent genotoxic effect of EMF. Expression of genes involved in the response to general stress (HSP70 encoding the 70 kDa heat

shock protein, and MEKK1 involved in signal transduction), oxidative stress (CAT, encoding catalase), and chemical and immune defence (LYS, encoding lysenin, and MYD, encoding a myeloid differentiation factor) were up-regulated after exposure to 10 and modulated 23 V m-1 field levels. Western blots showing an increased quantity of HSP70 and MTCO1 proteins confirmed this stress response.

(NE) Bourthoumieu S, Joubert V, Marin B, Collin A, Leveque P, Terro F, Yardin C. Cytogenetic studies in human cells exposed in vitro to GSM-900 MHz radiofrequency radiation using R-banded karyotyping. Radiat Res 174:712-718, 2010. (VT, AE, GT)

It is important to determine the possible effects of exposure to radiofrequency (RF) radiation on the genetic material of cells since damage to the DNA of somatic cells may be linked to cancer development or cell death and damage to germ cells may lead to genetic damage in next and subsequent generations. The objective of this study was to investigate whether exposure to radiofrequency radiation similar to that emitted by mobile phones of second-generation standard Global System for Mobile Communication (GSM) induces genotoxic effects in cultured human cells. The cytogenetic effects of GSM-900 MHz (GSM-900) RF radiation were investigated using R-banded karyotyping after in vitro exposure of human cells (amniotic cells) for 24 h. The average specific absorption rate (SAR) was 0.25 W/kg. The exposures were carried out in wire-patch cells (WPCs) under strictly controlled conditions of temperature. The genotoxic effect was assessed immediately or 24 h after exposure using four different samples. One hundred metaphase cells were analyzed per assay. Positive controls were provided by using bleomycin. We found no direct cytogenetic effects of GSM-900 either 0 h or 24 h after exposure. To the best of our knowledge, our work is the first to study genotoxicity using complete R-banded karyotyping, which allows visualizing all the chromosomal rearrangements, either numerical or structural.

(NE) Bourthoumieu S, Terro F, Leveque P, Collin A, Joubert V, Yardin C. Aneuploidy studies in human cells exposed in vitro to GSM-900 MHz radiofrequency radiation using FISH. Int J Radiat Biol 87:400-408, 2011. (VT, AE, GT)

PURPOSE: Since previous research found an increase in the rate of aneuploidies in human lymphocytes exposed to radiofrequencies, it seems important to perform further studies. The objective of this study was then to investigate whether the exposure to RF (radiofrequency) radiation similar to that emitted by mobile phones of a second generation standard, i.e., Global System for Mobile communication (GSM) may induce aneuploidy in cultured human cells. MATERIALS AND METHODS: The potential induction of genomic instability by GSM-900 MHz radiofrequency (GSM-900) was investigated after in vitro exposure of human amniotic cells for 24 h to average-specific absorption rates (SAR) of 0.25, 1, 2 and 4 W/kg in the temperature range of 36.3-39.7°C. The exposures were carried out in a wire-patch cell (WPC). The rate of aneuploidy of chromosomes 11 and 17 was determined by interphase FISH (Fluorescence In Situ Hybridisation) immediately after independent exposure of three different donors for 24 h. At least 100 interphase cells were analysed per assay. RESULTS: No significant change in the rate of aneuploidy of chromosomes 11 and 17 was found following exposure to GSM-900 for 24 h at average SAR up to 4 W/kg. CONCLUSION: Our study did not show any in vitro aneuploidogenic effect of GSM using FISH and is not in agreement with the results of previous research.

(NE) Bourthoumieu S, Magnaudeix A, Terro F, Leveque P, Collin A, Yardin C. Study of p53 expression and post-transcriptional modifications after GSM-900 radiofrequency exposure of human amniotic cells. Bioelectromagnetics. 3(1):52-60, 2013. (VT, AE, GT)

The potential effects of radiofrequency (RF) exposure on the genetic material of cells are very important to determine since genome instability of somatic cells may be linked to cancer development. In response to genetic damage, the p53 protein is activated and can induce cell cycle arrest allowing more time for DNA repair or elimination of damaged cells through apoptosis. The objective of this study was to investigate whether the exposure to RF electromagnetic fields, similar to those emitted by mobile phones of the second generation standard, Global System for Mobile Communications (GSM), may induce expression of the p53 protein and its activation by post-translational modifications in cultured human cells. The potential induction of p53 expression and activation by GSM-900 was investigated after in vitro exposure of human amniotic cells for 24 h to average specific absorption rates (SARs) of 0.25, 1, 2, and 4 W/kg in the temperature range of 36.3-39.7 °C. The exposures were carried out using a wire-patch cell (WPC) under strictly controlled conditions of temperature. Expression and activation of p53 by phosphorylation at serine 15 and 37 were studied using Western blot assay immediately after three independent exposures of cell cultures provided from three different donors. Bleomycin-exposed cells were used as a positive control. According to our results, no significant changes in the expression and activation of the p53 protein by phosphorylation at serine 15 and 37 were found following exposure to GSM-900 for 24 h at average SARs up to 4 W/kg in human embryonic cells.

(E) Brech A, Kubinyi G, Németh Z, Bakos J, Fiocchi S, Thuróczy G. Genotoxic effects of intermediate frequency magnetic fields on blood leukocytes in vitro. Mutat Res Genet Toxicol Environ Mutagen 845:403060, 2019.(VT, AE, GT)

The widespread presence of electromagnetic sources in daily life has initiated several studies on the effects of radiofrequency and power frequency fields. Only few investigations on the genotoxic effects of exposure to intermediate frequency magnetic fields (IF-MF) have been done so far. Therefore, the aim of this study was to evaluate possible genotoxic effects of exposure to 123.90 kHz and 250.80 kHz IF-MF on canine and human blood. Blood was exposed to IF-MF at 630 A/m (0.79 mT) and 80 A/m (0.10 m T) with exposure durations of 1-5 h (hourly), 20 and 24 h. Cylindrically divided Petri dish system was developed for in vitro exposures where different induced current could be achieved in the samples at the same magnetic flux density level. For the assessment of genotoxicity the alkaline comet assay was applied. We detected a statistically significant increase in DNA damage only following 20 h exposure to IF-MF.

(E) Burlaka A, Tsybulin O, Sidorik E, Lukin S, Polishuk V, Tsehmistrenko S, Yakymenko I. Overproduction of free radical species in embryonal cells exposed to low intensity radiofrequency radiation. Exp Oncol. 35(3):219-225, 2013. (VO, LE, GT, LI, DE, OX)

Aim: Long-term exposure of humans to low intensity radiofrequency electromagnetic radiation (RF-EMR) leads to a statistically significant increase in tumor incidence. Mechanisms of such

the effects are unclear, but features of oxidative stress in living cells under RF-EMR exposure were previously reported. Our study aims to assess a production of initial free radical species, which lead to oxidative stress in the cell. Materials and Methods: Embryos of Japanese quails were exposed in ovo to extremely low intensity RF-EMR of GSM 900 MHz (0.25 μW/cm2) during 158-360 h discontinuously (48 c - ON, 12 c - OFF) before and in the initial stages of development. The levels of superoxide (O2·-), nitrogen oxide (NO·), thiobarbituric acid reactive substances (TBARS), 8-oxo-2'-deoxyguanosine (8-oxo-dG) and antioxidant enzymes' activities were assessed in cells/tissues of 38-h, 5- and 10-day RF-EMR exposed and unexposed embryos. Results: The exposure resulted in a significant persistent overproduction of superoxide and nitrogen oxide in embryo cells during all period of analyses. As a result, significantly increased levels of TBARS and 8-oxo-dG followed by significantly decreased levels of superoxide dismutase and catalase activities were developed in the exposed embryo cells. Conclusion: Exposure of developing quail embryos to extremely low intensity RF-EMR of GSM 900 MHz during at least one hundred and fifty-eight hours leads to a significant overproduction of free radicals/reactive oxygen species and oxidative damage of DNA in embryo cells. These oxidative changes may lead to pathologies up to oncogenic transformation of cells.

(E) Busljeta I, Trosic I, Milkovic-Kraus S. Erythropoietic changes in rats after 2.45 GJz nonthermal irradiation. Int J Hyg Environ Health. 207(6):549-554, 2004.(VO. LE, GT)

The purpose of this study was to observe the erythropoietic changes in rats subchronically exposed to radiofrequency microwave (RF/MW) irradiation at nonthermal level. Adult male Wistar rats (N=40) were exposed to 2.45 GHz continuous RF/MW fields for 2 hours daily, 7 days a week, at 5-10 mW/cm2. Exposed animals were divided into four subgroups (n=10 animals in each subgroup) in order to be irradiated for 2, 8, 15 and 30 days. Animals were sacrified on the final irradiation day of each treated subgroup. Unexposed rats were used as control (N=24). Six animals were included into the each control subgroup. Bone marrow smears were examined to determine absolute counts of anuclear cells and erythropoietic precursor cells. The absolute erythrocyte count, haemoglobin and haematocrit values were observed in the peripheral blood by an automatic cell counter. The bone marrow cytogenetic analysis was accomplished by micronucleus (MN) tests. In the exposed animals erythrocyte count, haemoglobin and haematocrit were increased in peripheral blood on irradiation days 8 and 15. Concurrently, anuclear cells and erythropoietic precursor cells were significantly decreased ($p < 0.05$) in the bone marrow on day 15, but micronucleated cells' frequency was increased. In the applied experimental condition, RF/MW radiation might cause disturbance in red cell maturation and proliferation, and induce micronucleus formation in erythropoietic cells.

(E) Buttiglione M, Roca L, Montemurno E, Vitiello F, Capozzi V, Cibelli G. Radiofrequency radiation (900 MHz) induces Egr-1 gene expression and affects cell-cycle control in human neuroblastoma cells. J Cell Physiol. 213(3):759-767, 2007. (VT, AE, GE)

Many environmental signals, including ionizing radiation and UV rays, induce activation of Egr-1 gene, thus affecting cell growth and apoptosis. The paucity and the controversial knowledge about the effect of electromagnetic fields (EMF) exposure of nerve cells prompted us to

investigate the bioeffects of radiofrequency (RF) radiation on SH-SY5Y neuroblastoma cells. The effect of a modulated RF field of 900 MHz, generated by a wire patch cell (WPC) antenna exposure system on Egr-1 gene expression, was studied as a function of time. Short-term exposures induced a transient increase in Egr-1 mRNA level paralleled with activation of the MAPK subtypes ERK1/2 and SAPK/JNK. The effects of RF radiations on cell growth rate and apoptosis were also studied. Exposure to RF radiation had an anti-proliferative activity in SH-SY5Y cells with a significant effect observed at 24 h. RF radiation impaired cell cycle progression, reaching a significant G2-M arrest. In addition, the appearance of the sub-G1 peak, a hallmark of apoptosis, was highlighted after a 24-h exposure, together with a significant decrease in mRNA levels of Bcl-2 and survivin genes, both interfering with signaling between G2-M arrest and apoptosis. Our results provide evidence that exposure to a 900 MHz-modulated RF radiation affect both Egr-1 gene expression and cell regulatory functions, involving apoptosis inhibitors like Bcl-2 and survivin, thus providing important insights into a potentially broad mechanism for controlling in vitro cell viability.

(E) Cam ST, Seyhan N. Single-strand DNA breaks in human hair root cells exposed to mobile phone radiation. Int J Radiat Biol 88(5):420-424, 2012. (HU, AE, GT)

Purpose: To analyze the short term effects of radiofrequency radiation (RFR) exposure on genomic deoxyribonucleic acid (DNA) of human hair root cells. Subjects and methods: Hair samples were collected from 8 healthy human subjects immediately before and after using a 900-MHz GSM (Global System for Mobile Communications) mobile phone for 15 and 30 minutes. Single-strand DNA breaks of hair root cells from the samples were determined using the 'comet assay'. Results: The data showed that talking on a mobile phone for 15 or 30 minutes significantly increased ($p < .05$) single-strand DNA breaks in cells of hair roots close to the phone. Comparing the 15-min and 30-min data using the paired t-test also showed that significantly more damages resulted after 30 minutes than after 15 minutes of phone use. Conclusions: A short-term exposure (15 and 30 minutes) to RFR (900-MHz) from a mobile phone caused a significant increase in DNA single-strand breaks in human hair root cells located around the ear which is used for the phone calls.

(E) Campisi A, Gulino M, Acquaviva R, Bellia P, Raciti G, Grasso R, Musumeci F, Vanella A, Triglia A. Reactive oxygen species levels and DNA fragmentation on astrocytes in primary culture after acute exposure to low intensity microwave electromagnetic field. Neurosci Lett 473:52-55. 2010. (VT, AE, GT, LI, OX, WS)

The exposure of primary rat neocortical astroglial cell cultures to acute electromagnetic fields (EMF) in the microwave range was studied. Differentiated astroglial cell cultures at 14 days in vitro were exposed for 5, 10, or 20 min to either 900 MHz continuous waves or 900 MHz waves modulated in amplitude at 50 Hz using a sinusoidal waveform and 100% modulation index. The strength of the electric field (rms value) at the sample position was 10V/m. No change in cellular viability evaluated by MTT test and lactate dehydrogenase release was observed. A significant increase in ROS levels and DNA fragmentation was found only after exposure of the astrocytes to modulated EMF for 20 min. No evident effects were detected when shorter time intervals or continuous waves were used. The irradiation conditions allowed the exclusion of any possible thermal effect. Our data demonstrate, for the first time, that even acute exposure to low intensity

EMF induces ROS production and DNA fragmentation in astrocytes in primary cultures, which also represent the principal target of modulated EMF. Our findings also suggest the hypothesis that the effects could be due to hyperstimulation of the glutamate receptors, which play a crucial role in acute and chronic brain damage. Furthermore, the results show the importance of the amplitude modulation in the interaction between EMF and neocortical astrocytes.

(E) Cantu JC, Butterworth JW, Peralta XG, Payne JA, Echchgadda I. Analysis of global DNA methylation changes in human keratinocytes immediately following exposure to a 900 MHz radiofrequency field. Bioelectromagnetics 2023 Mar 31. doi: 10.1002/bem.22439. Online ahead of print. (VT, AE, LI, GE, EP)

The increasing use of nonionizing radiofrequency electromagnetic fields (RF-EMFs) in a wide range of technologies necessitates studies to further understanding of biological effects from exposures to such forms of electromagnetic fields. While previous studies have described mechanisms for cellular changes occurring following exposure to low-intensity RF-EMFs, the role of molecular epigenetics has not been thoroughly investigated. Specifically unresolved is the effect of RF-EMFs on deoxyribonucleic acid (DNA) methylation, which is a powerful epigenetic process, used by cells to regulate gene expression. DNA methylation is dynamic and can be rapidly triggered in response to external stimuli such as exposure to RF-EMFs. In the present study, we performed a global analysis of DNA methylation patterns in human keratinocytes exposed to 900 MHz RF-EMFs for 1 h at a low dose rate (estimated mean specific absorption rate (SAR) < 10 mW/kg). We used a custom system to allow stable exposure of cell cultures to RF-EMFs under biologically relevant conditions (37 °C, 5% CO_2 , 95% humidity). We performed whole genome bisulfite sequencing directly following RF-EMF exposure to examine the immediate changes in DNA methylation patterns and identify early differentially methylated genes in RF-EMF-exposed keratinocytes. By correlating global gene expression to whole genome bisulfite sequencing, we identified six common targets that were both differentially methylated and differentially expressed in response to RF-EMF exposure. The results highlight a potential epigenetic role in the cellular response to RF-EMFs. Particularly, the six identified targets may potentially be developed as epigenetic biomarkers for immediate responses to RF-EMF exposure.

(E) Cappucci, U., Assunta Maria Casale, Mirena Proietti, Fiorenzo Marinelli, Livio Giuliani, Lucia Piacentini. WiFi Related Radiofrequency Electromagnetic Fields Promote Transposable Element Dysregulation and Genomic Instability in *Drosophila melanogaster.* Cells 11(24):4036, 2022. (VO, LE, LI, GT, OX)

Exposure to artificial radio frequency electromagnetic fields (RF-EMFs) has greatly increased in recent years, thus promoting a growing scientific and social interest in deepening the biological impact of EMFs on living organisms. The current legislation governing the exposure to RF-EMFs is based exclusively on their thermal effects, without considering the possible non-thermal adverse health effects from long term exposure to EMFs. In this study we investigated the biological non-thermal effects of low-level indoor exposure to RF-EMFs produced by WiFi wireless technologies, using *Drosophila melanogaster* as the model system. Flies were exposed to 2.4 GHz radiofrequency in a Transverse Electromagnetic (TEM) cell device to ensure homogenous controlled fields. Signals were continuously monitored during the experiments and

regulated at non thermal levels. The results of this study demonstrate that WiFi electromagnetic radiation causes extensive heterochromatin decondensation and thus a general loss of transposable elements epigenetic silencing in both germinal and neural tissues. Moreover, our findings provide evidence that WiFi related radiofrequency electromagnetic fields can induce reactive oxygen species (ROS) accumulation, genomic instability, and behavioural abnormalities. Finally, we demonstrate that WiFi radiation can synergize with RasV12 to drive tumor progression and invasion. All together, these data indicate that radiofrequency radiation emitted from WiFi devices could exert genotoxic effects in *Drosophila* and set the stage to further explore the biological effects of WiFi electromagnetic radiation on living organisms..

(E) Cervellati F, Valacchi G, Lunghi L, Fabbri E, Valbonesi P, Marci R, Biondi C, Vesce F. 17-β-estradiol counteracts the effects of high frequency electromagnetic fields on trophoblastic connexins and integrins. Oxid Med Cell Longev. 2013:280850, 2013. (VT, AE, GE, IX)

We investigated the effect of high-frequency electromagnetic fields (HF-EMFs) and 17-β-estradiol on connexins (Cxs), integrins (Ints), and estrogen receptor (ER) expression, as well as on ultrastructure of trophoblast-derived HTR-8/SVneo cells. HF-EMF, 17-β-estradiol, and their combination induced an increase of Cx40 and Cx43 mRNA expression. HF-EMF decreased Int alpha1 and β 1 mRNA levels but enhanced Int alpha5 mRNA expression. All the Ints mRNA expressions were increased by 17-β-estradiol and exposure to both stimuli. ER-β mRNA was reduced by HF-EMF but augmented by 17-β-estradiol alone or with HF-EMF. ER-β immunofluorescence showed a cytoplasmic localization in sham and HF-EMF exposed cells which became nuclear after treatment with hormone or both stimuli. Electron microscopy evidenced a loss of cellular contact in exposed cells which appeared counteracted by 17-β-estradiol. We demonstrate that 17-β-estradiol modulates Cxs and Ints as well as ER-β expression induced by HF-EMF, suggesting an influence of both stimuli on trophoblast differentiation and migration.

(E) Chandel S, Kaur S, Issa M, Singh HP, Batish DR, Kohli RK. Exposure to mobile phone radiations at 2350 MHz incites cyto- and genotoxic effects in root meristems of *Allium cepa*. J Environ Health Sci Eng. 17(1):97-104, 2019a. (VO, AE, GT)

BACKGROUND: The exponential increase of electromagnetic field radiations (EMF-r) in the natural environment has raked up the controversies regarding their biological effects. Concern regarding the putative capacity of EMF-r to affect living beings has been growing due to the ongoing elevation in the use of high frequency EMF-r in communication systems, e.g. Mobile phones. METHODS: In the present study, we tried to examine the cyto- and genotoxic potential of mobile phone EMF-r at 2350 MHz using onions (*Allium cepa* L.). Fresh adventitious onion roots were exposed to continuous EMF-r at 2350 MHz for different time periods (1 h, 2 h and 4 h). The evaluation of cytotoxicity was done in terms of mitotic index (MI), phase index and chromosomal aberrations. Genotoxicity was investigated employing comet assay in terms of changes in % HDNA (head DNA) and % TDNA (tail DNA), TM (tail moment) and OTM (olive tail moment). Data were analyzed using one-way ANOVA and mean values were separated using post hoc Tukey's test. RESULTS: The results manifested a significant increase of MI and

chromosomal aberrations (%) upon 4 h, and ≥ 2 h of exposure, respectively, as compared to the control. No specific changes in phase index in response to EMF-r exposure were observed. The % HDNA and % TDNA values exhibited significant changes in contrast to that of control upon 2 h and 4 h of exposure, respectively. However, TM and OTM did not change significantly. CONCLUSIONS: Our results infer that continuous exposures of radiofrequency EMF-r (2350 MHz) for long durations have a potential of inciting cyto- and genotoxic effects in onion root meristems.

(E) Chandel S, Kaur S, Issa M, Singh HP, Batish DR, Kohli RK. Appraisal of immediate and late effects of mobile phone radiations at 2100 MHz on mitotic activity and DNA integrity in root meristems of Allium cepa. Protoplasma. 256(5):1399-1407, 2019b. (VO, AE, GT)

The present study evaluated the potential of 2100 MHz radiofrequency radiations to act as cytotoxic and genotoxic agent. Fresh onion (Allium cepa L.) roots were exposed to electromagnetic field radiations (EMF-r) for different durations (1 h and 4 h) and evaluated for mitotic index (MI), phase index, chromosomal aberrations, and DNA damage. DNA damage was investigated with the help of the comet assay by assessing various parameters like % head DNA (HDNA), % tail DNA (TDNA), tail moment (TM), and olive tail moment (OTM). Effects of EMF-r exposure were also compared with that of methyl methanesulfonate (MMS; 90 μM), which acted as a positive control. The post-exposure effects of EMF-r after providing the test plants with an acclimatization period of 24 h were also evaluated. Compared to the control, a significant increase in the MI and aberration percentage was recorded upon 4 h of exposure. However, no specific trend of phase index in response to exposure was detected. EMF-r exposure incited DNA damage with a significant decrease in HDNA accompanied by an increase in TDNA upon exposure of 4 h. However, TM and OTM did not change significantly upon exposure as compared to that of control. Analysis of the post-exposure effects of EMF-r did not show any significant change/recovery. Our data, thus, suggest the potential cytotoxic and genotoxic nature of 2100 MHz EMF-r. Our study bears great significance in view of the swiftly emergent EMF-r in the surrounding environment and their potential for inciting aberrations at the chromosomal level, thus posing a genetic hazard.

(NE) Chang SK, Choi JS, Gil HW, Yang JO, Lee EY, Jeon YS, Lee ZW, Lee M, Hong MY, Ho Son T, Hong SY. Genotoxicity evaluation of electromagnetic fields generated by 835-MHz mobile phone frequency band. Eur J Cancer Prev 14:175-179, 2005. (VT, AE, GT) (Some interaction effects with chemicals are reported in this paper.)

It is still unclear whether the exposure to electromagnetic fields (EMFs) generated by mobile phone radiation is directly linked to cancer. We examined the biological effects of an EMF at 835 MHz, the most widely used communication frequency band in Korean CDMA mobile phone networks, on bacterial reverse mutation (Ames assay) and DNA stability (in vitro DNA degradation). In the Ames assay, tester strains alone or combined with positive mutagen were applied in an artificial mobile phone frequency EMF generator with continuous waveform at a specific absorption rate (SAR) of 4 W/kg for 48 h. In the presence of the 835-MHz EMF radiation, incubation with positive mutagen 4-nitroquinoline-1-oxide and cumene hydroxide further increased the mutation rate in Escherichia coli WP2 and TA102, respectively, while the

contrary results in Salmonella typhimurium TA98 and TA1535 treated with 4-nitroquinoline-1-oxide and sodium azide, respectively, were shown as antimutagenic. However, these mutagenic or co-mutagenic effects of 835-MHz radiation were not significantly repeated in other relevant strains with same mutation type. In the DNA degradation test, the exposure to 835-MHz EMF did not change the rate of degradation observed using plasmid pBluescriptSK(+) as an indicator. Thus, we suggest that 835-MHz EMF under the conditions of our study neither affected the reverse mutation frequency nor accelerated DNA degradation in vitro.

(E) Chaturvedi CM, Singh VP, Singh P, Basu P, Singaravel M, Shukla RK, Dhawan A, Pati AK, Gangwar RK, and Singh SP. 2.45 GHZ (CW) microwave irradiation alters circadian organization, spatial memory, DNA structure in the brain cells and blood cell counts of male mice, Mus musculus. Progress In Electromagnetics Research B, Vol. 29, 23-42, 2011. (VO, LE, GT)

Present study examines biological effects of 2.45 GHz microwave radiation in Parkes strain mice. Forty-day-old mice were exposed to CW (continuous wave) microwave radiation (2 h/day for 30 days). Locomotor activity was recorded on running wheel for 12 days prior to microwave exposure (pre-exposure), 7 days during the first week of exposure (short-term exposure) and another 7-day spell during the last week of the 30-day exposure period (long-term exposure). Morris water maze test was performed from 17th to 22nd day of exposure. At the termination of the exposure, blood was processed for hematological parameters, brain for comet assay, epididymis for sperm count and motility and serum for SGOT (serum glutamate oxaloacetate transaminase) and SGPT (serum glutamate pyruvate transaminase). The results show that long-term radiation-exposed group exhibited a positive ψ (phase angle difference) for the onset of activity with reference to lights-off timing and most of the activity occurred within the light fraction of the LD (light: dark) cycle. Microwave radiation caused an increase in erythrocyte and leukocyte counts, a significant DNA strand break in brain cells and the loss of spatial memory in mice. This report for the first time provides experimental evidence that continuous exposure to low intensity microwave radiation may have an adverse effect on the brain function by altering circadian system and rate of DNA damage.

(NE) Chauhan V, Mariampillai A, Bellier PV, Qutob SS, Gajda GB, Lemay E, Thansandote A, McNamee JP. Gene expression analysis of a human lymphoblastoma cell line exposed in vitro to an intermittent 1.9 GHz pulse-modulated radiofrequency field. Radiat Res. 165(4):424-429, 2006a. (VT, AE, GE)

This study was designed to determine whether radiofrequency (RF) fields of the type used for wireless communications could elicit a cellular stress response. As general indicators of a cellular stress response, we monitored changes in proto-oncogene and heat-shock protein expression. Exponentially growing human lymphoblastoma cells (TK6) were exposed to 1.9 GHz pulse-modulated RF fields at average specific absorption rates (SARs) of 1 and 10 W/kg. Perturbations in the expression levels of the proto-oncogenes FOS, JUN and MYC after exposure to sham and RF fields were assessed by real-time RT-PCR. In addition, the transcript levels of the cellular stress proteins HSP27 and inducible HSP70 were also monitored. We demonstrated that transcript levels of these genes in RF-field-exposed cells showed no significant difference in

relation to the sham treatment group. However, concurrent positive (heat-shock) control samples displayed a significant elevation in the expression of HSP27, HSP70, FOS and JUN. Conversely, the levels of MYC mRNA were found to decline in the positive (heat-shock) control. In conclusion, our study found no evidence that the 1.9 GHz RF-field exposure caused a general stress response in TK6 cells under our experimental conditions.

(NE) Chauhan V, Mariampillai A, Gajda GB, Thansandote A, McNamee JP. Analysis of proto-oncogene and heat-shock protein gene expression in human derived cell-lines exposed in vitro to an intermittent 1.9 GHz pulse-modulated radiofrequency field. Int J Radiat Biol. 82(5):347-354, 2006b. (VT, AE, GE)

Purpose: Several studies have reported that radiofrequency (RF) fields, as emitted by mobile phones, may cause changes in gene expression in cultured human cell-lines. The current study was undertaken to evaluate this possibility in two human-derived immune cell-lines.Materials and methods: HL-60 and Mono-Mac-6 (MM6) cells were individually exposed to intermittent (5 min on, 10 min off) 1.9 GHz pulse-modulated RF fields at a average specific absorption rate (SAR) of 1 and 10 W/kg at 37 +/- 0.5 degrees C for 6 h. Concurrent negative and positive (heat-shock for 1 h at 43 degrees C) controls were conducted with each experiment. Immediately following RF field exposure (T = 6 h) and 18 h post-exposure (T = 24 h), cell pellets were collected from each of the culture dishes and analyzed for transcript levels of proto-oncogenes (c-jun, c-myc and c-fos) and the stress-related genes (heat shock proteins (HSP) HSP27 and HSP70B) by quantitative reverse transcriptase polymerase chain reaction (RT-PCR).Results: No significant effects were observed in mRNA expression of HSP27, HSP70, c-jun, c-myc or c-fos between the sham and RF-exposed groups, in either of the two cell-lines. However, the positive (heat-shock) control group displayed a significant elevation in the expression of HSP27, HSP70, c-fos and c-jun in both cell-lines at T = 6 and 24 h, relative to the sham and negative control groups.Conclusion: This study found no evidence that exposure of cells to non-thermalizing levels of 1.9 GHz pulse-modulated RF fields can cause any detectable change in stress-related gene expression.

(NE) Chauhan V, Qutob SS, Lui S, Mariampillai A, Bellier PV, Yauk CL, Douglas GR, Williams A, McNamee JP. Analysis of gene expression in two human-derived cell lines exposed in vitro to a 1.9 GHz pulse-modulated radiofrequency field. Proteomics. 7(21):3896-3905, 2007. (VT, AE, GE)

There is considerable controversy surrounding the biological effects of radiofrequency (RF) fields, as emitted by mobile phones. Previous work from our laboratory has shown no effect related to the exposure of 1.9 GHz pulse-modulated RF fields on the expression of 22,000 genes in a human glioblastoma-derived cell-line (U87MG) at 6 h following a 4 h RF field exposure period. As a follow-up to this study, we have now examined the effect of RF field exposure on the possible expression of late onset genes in U87MG cells after a 24 h RF exposure period. In addition, a human monocyte-derived cell-line (Mono-Mac-6, MM6) was exposed to intermittent (5 min ON, 10 min OFF) RF fields for 6 h and then gene expression was assessed immediately after exposure and at 18 h postexposure. Both cell lines were exposed to 1.9 GHz pulse-modulated RF fields for 6 or 24 h at specific absorption rates (SARs) of 0.1-10.0 W/kg. In

support of our previous results, we found no evidence that nonthermal RF field exposure could alter gene expression in either cultured U87MG or MM6 cells, relative to nonirradiated control groups. However, exposure of both cell-lines to heat-shock conditions (43 degrees C for 1 h) caused an alteration in the expression of a number of well-characterized heat-shock proteins.

(E) Chavdoula ED, Panagopoulos DJ, Margaritis LH. Comparison of biological effects between continuous and intermittent exposure to GSM-900-MHz mobile phone radiation: detection of apoptotic cell-death features. Mutat Res 700:51-61, 2010. (VO, LE, GT, RP)

In the present study we used a 6-min daily exposure of dipteran flies, Drosophila melanogaster, to GSM-900 MHz (Global System for Mobile Telecommunications) mobile phone electromagnetic radiation (EMR), to compare the effects between the continuous and four different intermittent exposures of 6min total duration, and also to test whether intermittent exposure provides any cumulative effects on the insect's reproductive capacity as well as on the induction of apoptotic cell death. According to our previous experiments, a 6-min continuous exposure per day for five days to GSM-900 MHz and DCS-1800 MHz (Digital Cellular System) mobile phone radiation, brought about a large decrease in the insect's reproductive capacity, as defined by the number of F pupae. This decrease was found to be non thermal and correlated with an increased percentage of induced fragmented DNA in the egg chambers' cells at early- and mid-oogenesis. In the present experiments we show that intermittent exposure also decreases the reproductive capacity and alters the actin cytoskeleton network of the egg chambers, another known aspect of cell death that was not investigated in previous experiments, and that the effect is also due to DNA fragmentation. Intermittent exposures with 10-min intervals between exposure sessions proved to be almost equally effective as continuous exposure of the same total duration, whereas longer intervals between the exposures seemed to allow the organism the time required to recover and partly overcome the above-mentioned effects of the GSM exposure.

(NE) Chemeris NK, Gapeyev AB, Sirota NP, Gudkova OY, Kornienko NV, Tankanag AV, Konovalov IV, Buzoverya ME, Suvorov VG, Logunov VA. DNA damage in frog erythrocytes after in vitro exposure to a high peak-power pulsed electromagnetic field. Mutat Res. 558(1-2): 27-34, 2004. (VT, AE, GT)

Till the present time, the genotoxic effects of high peak-power pulsed electromagnetic fields (HPPP EMF) on cultured cells have not been studied. We investigated possible genotoxic effects of HPPP EMF (8.8 GHz, 180 ns pulse width, peak power 65 kW, repetition rate 50 Hz) on erythrocytes of the frog Xenopus laevis. We used the alkaline comet assay, which is a highly sensitive method to assess DNA single-strand breaks and alkali-labile lesions. Blood samples were exposed to HPPP EMF for 40 min in rectangular wave guide. The specific absorption rate (SAR) calculated from temperature kinetics was about 1.6 kW/kg (peak SAR was about 300 MW/kg). The temperature rise in the blood samples at steady state was 3.5 +/- 0.1 degrees C. The data show that the increase in DNA damage after exposure of erythrocytes to HPPP EMF was induced by the rise in temperature in the exposed cell suspension. This was confirmed in experiments in which cells were incubated for 40 min under the corresponding temperature

conditions. The results allow us to conclude that HPPP EMF-exposure at the given modality did not cause any a-thermal genotoxic effect on frog erythrocytes in vitro.

(NE) Chemeris NK, Gapeyev AB, Sirota NP, Gudkova OYu, Tankanag AV, Konovalov IV, Buzoverya ME, Suvorov VG, Logunov VA. Lack of direct DNA damage in human blood leukocytes and lymphocytes after in vitro exposure to high power microwave pulses. Bioelectromagnetics 27(3):197-203, 2006. (VT, AE, GT)

Currently, the potential genotoxicity of high power microwave pulses (HPMP) is not clear. Using the alkaline single cell gel electrophoresis assay, also known as the alkaline comet assay, we studied the effects of HPMP (8.8 GHz, 180 ns pulse width, peak power 65 kW, pulse repetition frequency 50 Hz) on DNA of human whole-blood leukocytes and isolated lymphocytes. The cell suspensions were exposed to HPMP for 40 min in a rectangular waveguide. The average SAR calculated from the temperature kinetics was about 1.6 kW/kg (peak SAR was about 300 MW/kg). The steady-state temperature rise in the 50 microl samples exposed to HPMP was 3.5 +/- 0.1 degrees C. In independent experiments, we did not find any statistically significant DNA damage manifested immediately after in vitro HPMP exposure of human blood leukocytes or lymphocytes or after HPMP exposure of leukocytes subsequently incubated at 37 degrees C for 30 min. Our results indicate that HPMP under the given exposure conditions did not induce DNA strand breaks, alkali-labile sites, and incomplete excision repair sites, which could be detected by the alkaline comet assay.

(E) Chen C, Ma Q, Deng P, Lin M, Gao P, He M, Lu Y, Pi H, He Z, Zhou C, Zhang Y, Yu Z, Zhang L 1800 MHz Radiofrequency Electromagnetic Field Impairs Neurite Outgrowth Through Inhibiting EPHA5 Signaling. Front Cell Dev Biol 9:657623, 2021. (VT, AE, GE)

The increasing intensity of environmental radiofrequency electromagnetic fields (RF-EMF) has increased public concern about its health effects. Of particular concern are the influences of RF-EMF exposure on the development of the brain. The mechanisms of how RF-EMF acts on the developing brain are not fully understood. Here, based on high-throughput RNA sequencing techniques, we revealed that transcripts related to neurite development were significantly influenced by 1800 MHz RF-EMF exposure during neuronal differentiation. Exposure to RF-EMF remarkably decreased the total length of neurite and the number of branch points in neural stem cells-derived neurons and retinoic acid-induced Neuro-2A cells. The expression of Eph receptors 5 (EPHA5), which is required for neurite outgrowth, was inhibited remarkably after RF-EMF exposure. Enhancing EPHA5 signaling rescued the inhibitory effects of RF-EMF on neurite outgrowth. Besides, we identified that cAMP-response element-binding protein (CREB) and RhoA were critical downstream factors of EPHA5 signaling in mediating the inhibitory effects of RF-EMF on neurite outgrowth. Together, our finding revealed that RF-EMF exposure impaired neurite outgrowth through EPHA5 signaling. This finding explored the effects and key mechanisms of how RF-EMF exposure impaired neurite outgrowth and also provided a new clue to understanding the influences of RF-EMF on brain development.

(E) Chen G, Lu D, Chiang H, Leszczynski D, Xu Z. Using model organism Saccharomyces cerevisiae to evaluate the effects of ELF-MF and RF-EMF exposure on global gene expression. Bioelectromagnetics. 33(7):550-560, 2012 . (VO, AE, GE)

The potential health hazard of exposure to electromagnetic fields (EMF) continues to cause public concern. However, the possibility of biological and health effects of exposure to EMF remains controversial and their biophysical mechanisms are unknown. In the present study, we used Saccharomyces cerevisiae to identify genes responding to extremely low frequency magnetic fields (ELF-MF) and to radiofrequency EMF (RF-EMF) exposures. The yeast cells were exposed for 6 h to either 0.4 mT 50 Hz ELF-MF or 1800 MHz RF-EMF at a specific absorption rate of 4.7 W/kg. Gene expression was analyzed by microarray screening and confirmed using real-time reverse transcription-polymerase chain reaction (RT-PCR). We were unable to confirm microarray-detected changes in three of the ELF-MF responsive candidate genes using RT-PCR ($P > 0.05$). On the other hand, out of the 40 potential RF-EMF responsive genes, only the expressions of structural maintenance of chromosomes 3 (SMC3) and aquaporin 2 (AQY2 (m)) were confirmed, while three other genes, that is, halotolerance protein 9 (HAL9), yet another kinase 1 (YAK1) and one function-unknown gene (open reading frame: YJL171C), showed opposite changes in expression compared to the microarray data ($P < 0.05$). In conclusion, the results of this study suggest that the yeast cells did not alter gene expression in response to 50 Hz ELF-MF and that the response to RF-EMF is limited to only a very small number of genes. The possible biological consequences of the gene expression changes induced by RF-EMF await further investigation.

(E)Cheon H, Hur JK, Hwang W, Yang H-J, Son J-H. Epigenetic modification of gene expression in cancer cells by terahertz demethylation. Sci Rep 13(1):4930, 2023. (VT, AE, GE, EP)

Terahertz (THz) radiation can affect the degree of DNA methylation, the spectral characteristics of which exist in the terahertz region. DNA methylation is an epigenetic modification in which a methyl (CH_3) group is attached to cytosine, a nucleobase in human DNA. Appropriately controlled DNA methylation leads to proper regulation of gene expression. However, abnormal gene expression that departs from controlled genetic transcription through aberrant DNA methylation may occur in cancer or other diseases. In this study, we demonstrate the modification of gene expression in cells by THz demethylation using resonant THz radiation. Using an enzyme-linked immunosorbent assay, we observed changes in the degree of global DNA methylation in the SK-MEL-3 melanoma cell line under irradiation with 1.6-THz radiation with limited spectral bandwidth. Resonant THz radiation demethylated living melanoma cells by 19%, with no significant occurrence of apurinic/apyrimidinic sites, and the demethylation ratio was linearly proportional to the power of THz radiation. THz demethylation downregulates FOS, JUN, and CXCL8 genes, which are involved in cancer and apoptosis pathways. Our results show that THz demethylation has the potential to be a gene expression modifier with promising applications in cancer treatment.

(NE) Choi J, Min K, Jeon S, Kim N, Pack JK, Song K. Continuous exposure to 1.7 GHz LTE electromagnetic fields increases intracellular reactive oxygen species to decrease human cell proliferation and induce senescence. Sci Rep. 10(1):9238, 2020. (VT, AE, GT, OX)

Due to the rapid development of mobile phone technology, we are continuously exposed to 1.7 GHz LTE radio frequency electromagnetic fields (RF-EMFs), but their biological effects have not been clarified. Here, we investigated the non-thermal cellular effects of these RF-EMFs on human cells, including human adipose tissue-derived stem cells (ASCs), Huh7 and Hep3B liver cancer stem cells (CSCs), HeLa and SH-SY5Y cancer cells, and normal fibroblast IMR-90 cells. When continuously exposed to 1.7 GHz LTE RF-EMF for 72 h at 1 and 2 SAR, cell proliferation was consistently decreased in all the human cells. The anti-proliferative effect was higher at 2 SAR than 1 SAR and was less severe in ASCs. The exposure to RF-EMF for 72 h at 1 and 2 SAR did not induce DNA double strand breaks or apoptotic cell death, but did trigger a slight delay in the G1 to S cell cycle transition. Cell senescence was also clearly observed in ASC and Huh7 cells exposed to RF-EMF at 2 SAR for 72 h. Intracellular ROS increased in these cells and the treatment with an ROS scavenger recapitulated the anti-proliferative effect of RF-EMF. These observations strongly suggest that 1.7 GHz LTE RF-EMF decrease proliferation and increase senescence by increasing intracellular ROS in human cells.

(NE) Ciaravino V, Meltz ML, Erwin DN, Absence of a synergistic effect between moderate power radio-frequency electromagnetic radiation and adriamycin on cell-cycle progression and sister-chromatid exchange. Bioelectromagnetics 12(5):289-298, 1991.(VT, AE, GT)

In our laboratories we are conducting investigations of potential interactions between radio-frequency electromagnetic radiation (RFR) and chemicals that are toxic by different mechanisms to mammalian cells. The RFR is being tested at frequencies in the microwave range and at different power levels. We report here on the 1) ability of simultaneous RFR exposures to alter the distribution of cells in first and second mitoses from that after treatment by adriamycin alone, and 2) on the ability of simultaneous RFR exposure to alter the extent of sister chromatid exchanges (SCEs) induced by adriamycin alone. This chemical was selected because of its reported mechanism of action and because it is of interest in the treatment of cancer. In our studies, Chinese hamster ovary (CHO) cells were exposed for 2 h simultaneously to adriamycin and pulsed RFR at a frequency of 2,450 MHz and a specific absorption rate of 33.8 W/Kg. The maximal temperature (in the tissue-culture medium) was 39.7 +/- 0.2 degrees C. The experiments were controlled for chemical and RFR exposures, as well as for temperature. Verified statistically, the data indicate that the RFR did not affect changes in cell progression caused by adriamycin, and the RFR did not change the number of SCEs that were induced by the adriamycin, which adriamycin is known to affect cells by damaging their membranes and DNA.

(E) Costantini E, Aielli L, Serra F, De Dominicis L, Falasca K, Di Giovanni P, Reale M. Evaluation of Cell Migration and Cytokines Expression Changes under the Radiofrequency Electromagnetic Field on Wound Healing In Vitro Model. Int J Mol Sci. 23(4):2205, 2022. (VT, AE, GE)

Wound healing (WH) proceeds through four distinct phases: hemostasis, inflammation, proliferation, and remodeling. Impaired WH may be the consequence of the alteration of one of these phases and represents a significant health and economic burden to millions of individuals. Thus, new therapeutic strategies are the topics of intense research worldwide. Although radiofrequency electromagnetic field (RF-EMF) has many medical applications in rehabilitation, pain associated with musculoskeletal disorders, and degenerative joint disorders, its impact on WH is not fully understood. The process of WH begins just after injury and continues during the inflammatory and proliferative phases. A thorough understanding of the mechanisms by which RF-EMF can improve WH is required before it can be used as a non-invasive, inexpensive, and easily self-applicable therapeutic strategy. Thus, the aim of this study is to explore the therapeutic potential of different exposure setups of RF-EMF to drive faster healing, evaluating the keratinocytes migration, cytokines, and matrix metalloproteinases (MMPs) expression. The results showed that RF-EMF treatment promotes keratinocytes' migration and regulates the expression of genes involved in healing, such as MMPs, tissue inhibitors of metalloproteinases, and pro/anti-inflammatory cytokines, to improve WH.

(E) Czyz J, Guan K, Zeng Q, Nikolova T, Meister A, Schönborn F, Schuderer J, Kuster N, Wobus AM. High frequency electromagnetic fields (GSM signals) affect gene expression levels in tumor suppressor p53-deficient embryonic stem cells. Bioelectromagnetics 25(4):296-307, 2004. (VT, AE, GE)

Effects of electromagnetic fields (EMF) simulating exposure to the Global System for Mobile Communications (GSM) signals were studied using pluripotent embryonic stem (ES) cells in vitro. Wild-type ES cells and ES cells deficient for the tumor suppressor p53 were exposed to pulse modulated EMF at 1.71 GHz, lower end of the uplink band of GSM 1800, under standardized and controlled conditions, and transcripts of regulatory genes were analyzed during in vitro differentiation. Two dominant GSM modulation schemes (GSM-217 and GSM-Talk), which generate temporal changes between GSM-Basic (active during talking phases) and GSM-DTX (active during listening phases thus simulating a typical conversation), were applied to the cells at and below the basic safety limits for local exposures as defined for the general public by the International Commission on Nonionizing Radiation Protection (ICNIRP). GSM-217 EMF induced a significant upregulation of mRNA levels of the heat shock protein, hsp70 of p53-deficient ES cells differentiating in vitro, paralleled by a low and transient increase of c-jun, c-myc, and p21 levels in p53-deficient, but not in wild-type cells. No responses were observed in either cell type after EMF exposure to GSM-Talk applied at similar slot-averaged specific absorption rates (SAR), but at lower time-averaged SAR values. Cardiac differentiation and cell cycle characteristics were not affected in embryonic stem and embryonic carcinoma cells after exposure to GSM-217 EMF signals. Our data indicate that the genetic background determines cellular responses to GSM modulated EMF.

(E) D'ambrosio G, Lioi MB, Massa R, Scarfi MR, Zeni O. Genotoxic Effects of amplitude-modulated microwaves on human lymphocytes exposed in vitro under controlled conditions. Electro- and Magnetiobiology 14:157-164, 1995. (VT, AE, GT, WS)

Peripheral human blood from 23 healthy donors aged between 23 and 95 years was exposed to continuous wave (CW) or 50 Hz amplitude modulated (AM) microwave radiation and was cultured for 72 h. Other exposure parameters were: frequency 9 GHz, specific absorption rate (SAR) 90 mW/g, exposure duration 10 min. The possible genotoxic effect was evaluated by means of cytokinesis-block micronucleus method. A significant ($p < 0.05$) increase in micronuclei was found following AM microwave exposure

(E) d'Ambrosio G, Massa R, Scarfi MR, Zeni O, Cytogenetic damage in human lymphocytes following GMSK phase modulated microwave exposure. Bioelectromagnetics 23:7-13, 2002. (VT, AE, GT, WS)

The present study investigated, using in vitro experiments on human lymphocytes, whether exposure to a microwave frequency used for mobile communication, either unmodulated or in presence of phase only modulation, can cause modification of cell proliferation kinetics and/or genotoxic effects, by evaluating the cytokinesis block proliferation index and the micronucleus frequency. In the GSM 1800 mobile communication systems the field is both phase (Gaussian minimum shift keying, GMSK) and amplitude (time domain multiple access, TDMA) modulated. The present study investigated only the effects of phase modulation, and no amplitude modulation was applied. Human peripheral blood cultures were exposed to 1.748 GHz, either continuous wave (CW) or phase only modulated wave (GMSK), for 15 min. The maximum specific absorption rate (~5 W/kg) was higher than that occurring in the head of mobile phone users; however, no changes were found in cell proliferation kinetics after exposure to either CW or GMSK fields. As far as genotoxicity is concerned, the micronucleus frequency result was not affected by CW exposure; however, a statistically significant micronucleus effect was found following exposure to phase modulated field. These results would suggest a genotoxic power of the phase modulation per se

(NE) Danese E, LippiG, Buonocore R, Benati M, Bovo C, Bonaguri C, Salvagno GL, Brocco G, Roggenbuck D, Martina Montagnana M. Mobile phone radiofrequency exposure has no effect on DNA double strand breaks (DSB) in human lymphocytes. Ann Transl Med 5(13):272, 2017. (VT, AE, GT) (See also: Mortazavi SMJ. Comment on "Mobile phone radiofrequency exposure has no effect on DNA double strand breaks (DSB) in human lymphocytes" Ann Transl Med 5(21):441, 2017.)

Background: The use of mobile phones has been associated with an increased risk of developing certain type of cancer, especially in long term users. Therefore, this study was aimed to investigate the potential genotoxic effect of mobile phone radiofrequency exposure on human peripheral blood mononuclear cells *in vitro.* **Methods:** The study population consisted in 14 healthy volunteers. After collection of two whole blood samples, the former was placed in a plastic rack, 1 cm from the chassis of a commercial mobile phone (900 MHz carrier frequency), which was activated by a 30-min call. The second blood sample was instead maintained far from

mobile phones or other RF sources. The influence of mobile phone RF on DNA integrity was assessed by analyzing γ-H2AX foci in lymphocytes using immunofluorescence staining kit on AKLIDES. **Results:** No measure of γ-H2AX foci was significantly influenced by mobile phone RF exposure, nor mobile phone exposure was associated with significant risk of genetic damages *in vitro* (odds ratio comprised between 0.27 and 1.00). **Conclusions:** The results of this experimental study demonstrate that exposure of human lymphocytes to a conventional 900 MHz RF emitted by a commercial mobile phone for 30 min does not significantly impact DNA integrity.

(E) Dasdag S, Akdag MZ, Erdal ME, Erdal N, Ay OI, Ay ME, Yilmaz SG, Tasdelen B, Yegin K. Long term and excessive use of 900 MHz radiofrequency radiation alter microRNA expression in brain. Int J Radiat Biol 91(4):306-311, 2015a. (VO, LE, GE)

Purpose: We still do not have any information on the interaction between radiofrequency radiation (RF) and miRNA, which play paramount role in growth, differentiation, proliferation and cell death by suppressing one or more target genes. The purpose of this study was to bridge this gap by investigating effects of long-term 900 MHz mobile phone exposure on some of the miRNA in brain tissue. **Materials and methods:** The study was carried out on 14 Wistar Albino adult male rats by dividing them into two groups: Sham (n = 7) and exposure (n = 7). Rats in the exposure group were exposed to 900 MHz RF radiation for 3 h per day (7 days a week) for 12 months (one year). The same procedure was applied to the rats in the sham group except the generator was turned off. Immediately after the last exposure, rats were sacrificed and their brains were removed. rno-miR-9-5p, rno-miR-29a-3p, rno-miR-106b-5p, rno-miR-107 and rno-miR-125a-3p in brain were investigated in detail. **Results:** Results revealed that long-term exposure of 900 MHz RF radiation only decreased rno-miR107 (adjP* = 0.045) value where the whole body (rms) SAR value was 0.0369 W/kg. However, our results indicated that other microRNA evaluated in this study was not altered by 900 MHz RF radiation. **Conclusion:** 900 MHz RF radiation can alter some of the miRNA, which, in turn, may lead to adverse effects. Therefore, further studies should be performed.

(E) Dasdag S, Akdag MZ, Erdal ME, Erdal N, Ay OI, Ay ME, Yilmaz SG, Tasdelen B, Yegin K. Effects of 2.4 GHz radiofrequency radiation emitted from Wi-Fi equipment on microRNA expression in brain tissue. Int J Radiat Biol 91(7):555-561, 2015b. (VO, LE, GE)

Purpose: MicroRNAs (miRNA) play a paramount role in growth, differentiation, proliferation and cell death by suppressing one or more target genes. However, their interaction with radiofrequencies is still unknown. The aim of this study was to investigate the long-term effects of radiofrequency radiation emitted from a Wireless Fidelity (Wi-Fi) system on some of the miRNA in brain tissue. **Materials and methods:** The study was carried out on 16 Wistar Albino adult male rats by dividing them into two groups such as sham (n = 8) and exposure (n = 8). Rats in the exposure group were exposed to 2.4 GHz radiofrequency (RF) radiation for 24 hours a day for 12 months (one year). The same procedure was applied to the rats in the sham group except the Wi-Fi system was turned off. Immediately after the last exposure, rats were sacrificed and their brains were removed. miR-9-5p, miR-29a-3p, miR-106b-5p, miR-107, miR-125a-3p in

brain were investigated in detail. **Results:** The results revealed that long-term exposure of 2.4 GHz Wi-Fi radiation can alter expression of some of the miRNAs such as miR-106b-5p (adj p* = 0.010) and miR-107 (adj p* = 0.005). We observed that mir 107 expression is 3.3 times and miR- 106b-5p expression is 3.65 times lower in the exposure group than in the control group. However, miR-9-5p, miR-29a-3p and miR-125a-3p levels in brain were not altered. **Conclusion:** Long-term exposure of 2.4 GHz RF may lead to adverse effects such as neurodegenerative diseases originated from the alteration of some miRNA expression and more studies should be devoted to the effects of RF radiation on miRNA expression levels.

(E) Dasdag S, Erdal ME, Erdal N, Tasdelen B, Kiziltug MT, Yegin K, Akdag MZ. 900 MHz Radiofrequency Radiation Has Potential to Increase the Expression of rno-miR-145-5p in Brain. J Int Dent Med Res 12(4): 1652-1658, 2019. (VO, LE, GE)

Interaction between radiofrequency radiation (RFR) and miRNA which plays paramount role in growth, differentiation, proliferation, and cell death by suppressing one or more target genes, is still unknown. Therefore, the purpose of this study is to investigate the effects of long term 900 MHz mobile phone exposure on some of the miRNA in brain tissues (sham: 7; Exposure : 7)., which were kept in appropriate laboratory conditions for the second phase of our previous study [5]. It is remembered that rats in the exposure group were exposed to 900 MHz RFR for 3 h per day (7 days a week) for one year. For the aim of this study, expression of miRNAs such as rno-miR-22-3p, rnomiR-24-1-3p, rno-miR-132-3p, rno-miR-145-5p, rno-miR-181a-5p, rno-miR-186-5p, rno-miR-195- 5p, rno-miR-219a-5p, rno-miR-221-3p and rno-miR-222-3p were investigated. Results indicated that long-term exposure of 900 MHz RF radiation increased only expression of rno-miR-145-5p (adj P * = 0.047) value where 1g average SAR value in brain was 0.198 W/kg. Our results indicated that chronic exposure of 900 MHz RFR has potential to increase expression of rno-miR-145-5p. Therefore, further studies are necessary to understand the relation between 900 MHz mobile phone exposure and diseases related to the expression of rno-miR-145-5p.

(E) Dasdag S, Akdag MZ, Bashan M, Kizmaz V, Erdal N, Erdal ME, Kiziltug MT, Yegin K. Role of 2.4 GHz radiofrequency radiation emitted from Wi-Fi on some miRNA and fatty acids composition in brain, Electromagnetic Biology and Medicine 41(1):281-292, 2022. (VO, LE, GE)

The purpose of this study is to investigate the effects of 2.4 GHz Wi-Fi exposure, which is continuously used in the internet connection by mobile phones, computers and other wireless equipment, on microRNA and membrane and depot fatty acid composition of brain cells. Sixteen Wistar Albino rats were divided equally into two groups such as sham and exposure. The rats in the experimental group (n = 8) were exposed to 2.4 GHz RFR emitted from a Wi-Fi generator for 24 h/day for one year. The animals in the control group (n = 8) were kept under the same

conditions as the experimental group, but the Wi-Fi generator was turned off. At the end of the study, rats were sacrificed and brains were removed to analyze miRNA expression and membrane and depot fatty acids of brain cells. We analyzed the situation of ten different miRNA expressions and nineteen fatty acid patterns in this study. We observed that long-term and excessive exposure of 2.4 GHz Wi-Fi radiation increased rno-miR-181a-5p, phosphatidylserine (PS) and triacylglycerol (TAG) in the brain. In conclusion, 2.4 GHz Wi-Fi exposure has the potential to alter rno-miR-181a-5p expression and the fatty acid percentage of some membrane lipids such as phospholipid (PL), phosphatidylserine (PS) and triacylglycerol (TAG), which are depot fats in the brain. However, the uncontrolled use of RFRs, whose use and diversity have reached incredible levels with each passing day and which are increasing in the future, may be paving the way for many diseases that we cannot connect with today.

(E) Dasgupta S, Leong C, Simonich MT, Truong L, Liu H, Tanguay RL. Transcriptomic and Long-Term Behavioral Deficits Associated with Developmental 3.5 GHz Radiofrequency Radiation Exposures in Zebrafish. Environ Sci Technol Lett 9(4):327-332, 2022. (VO, AE, GE, DE)

The rapid deployment of the fifth-generation (5G) spectrum by the telecommunication industry is intended to promote better connectivity and data integration among various industries. However, concerns among the public about the safety and health effects of radiofrequency radiations (RFRs) emitted from the newer-generation cell phone frequencies remain, partly due to the lack of robust scientific data. Previously, we used developmental zebrafish to model the bioactivity of 3.5 GHz RFR, a frequency used by 5G-enabled cell phones, in a novel RFR exposure chamber. With RFR exposures from 6 h post-fertilization (hpf) to 48 hpf, we observed that, despite no teratogenic effects, embryos showed subtle hypoactivity in a startle response behavior assay, suggesting abnormal sensorimotor behavior. This study builds upon the previous one by investigating the transcriptomic basis of RFR-associated behavior effects and their persistence into adulthood. Using mRNA sequencing, we found a modest transcriptomic disruption at 48 hpf, with 28 differentially expressed genes. KEGG pathway analysis showed that biochemical pathways related to metabolism were significantly perturbed. Embryos were grown to adulthood, and then a battery of behavioral assays suggested subtle but significant abnormal responses in RFR-exposed fish across the different assays evaluated that suggest potential long-term behavioral effects. Overall, our study suggests the impacts of RFRs on the developing brain, behavior, and the metabolome should be further explored.

(NE) Dawe AS, Nylund R, Leszczynski D, Kuster N, Reader T, De Pomerai DI. Continuous wave and simulated GSM exposure at 1.8 W/kg and 1.8 GHz do not induce hsp16-1 heat-shock gene expression in Caenorhabditis elegans. Bioelectromagnetics. 29(2):92-99, 2008. (VO, AE, GE)

Recent data suggest that there might be a subtle thermal explanation for the apparent induction by radiofrequency (RF) radiation of transgene expression from a small heat-shock protein (hsp16-1) promoter in the nematode, Caenorhabditis elegans. The RF fields used in the C. elegans study were much weaker (SAR 5-40 mW kg(-1)) than those routinely tested in many

other published studies (SAR approximately 2 W kg(-1)). To resolve this disparity, we have exposed the same transgenic hsp16-1::lacZ strain of C. elegans (PC72) to higher intensity RF fields (1.8 GHz; SAR approximately 1.8 W kg(-1)). For both continuous wave (CW) and Talk-pulsed RF exposures (2.5 h at 25 degrees C), there was no indication that RF exposure could induce reporter expression above sham control levels. Thus, at much higher induced RF field strength (close to the maximum permitted exposure from a mobile telephone handset), this particular nematode heat-shock gene is not up-regulated. However, under conditions where background reporter expression was moderately elevated in the sham controls (perhaps as a result of some unknown co-stressor), we found some evidence that reporter expression may be reduced by approximately 15% following exposure to either Talk-pulsed or CW RF fields.

(E) De Amicis A, Sanctis SD, Cristofaro SD, Franchini V, Lista F, Regalbuto E, Giovenale E, Gallerano GP, Nenzi P, Bei R, Fantini M, Benvenuto M, Masuelli L, Coluzzi E, Cicia C, Sgura A. Biological effects of in vitro THz radiation exposure in human foetal fibroblasts. Mutat Res Genet Toxicol Environ Mutagen. 793:150-160, 2015. (VT, AE, GT)

In recent years, terahertz (THz) radiation has been widely used in a variety of applications: medical, security, telecommunications and military areas. However, few data are available on the biological effects of this type of electromagnetic radiation and the reported results, using different genetic or cellular assays, are quite discordant. This multidisciplinary study focuses on potential genotoxic and cytotoxic effects, evaluated by several end-points, associated with THz radiation. For this purpose, in vitro exposure of human foetal fibroblasts to low frequency THz radiation (0.1-0.15THz) was performed using a Compact Free Electron Laser. We did not observe an induction of DNA damage evaluated by Comet assay, phosphorylation of H2AX histone or telomere length modulation. In addiction, no induction of apoptosis or changes in pro-survival signalling proteins were detected. Moreover, our results indicated an increase in the total number of micronuclei and centromere positive micronuclei induction evaluated by CREST analysis, indicating that THz radiation could induce aneugenic rather than clastogenic effects, probably leading to chromosome loss. Furthermore, an increase of actin polymerization observed by ultrastructural analysis after THz irradiation, supports the hypothesis that an abnormal assembly of spindle proteins could lead to the observed chromosomal malsegregation.

(E) De Iuliis GN, Newey RJ, King BV, Aitken RJ. Mobile phone radiation induces reactive oxygen species production and DNA damage in human spermatozoa in vitro. PLoS One 4:e6446, 2009. (VT, AE, GT, OX, RP)

BACKGROUND: In recent times there has been some controversy over the impact of electromagnetic radiation on human health. The significance of mobile phone radiation on male reproduction is a key element of this debate since several studies have suggested a relationship between mobile phone use and semen quality. The potential mechanisms involved have not been established, however, human spermatozoa are known to be particularly vulnerable to oxidative stress by virtue of the abundant availability of substrates for free radical attack and the lack of cytoplasmic space to accommodate antioxidant enzymes. Moreover, the induction of oxidative stress in these cells not only perturbs their capacity for fertilization but also contributes to sperm DNA damage. The latter has, in turn, been linked with poor fertility, an increased incidence of

miscarriage and morbidity in the offspring, including childhood cancer. In light of these associations, we have analyzed the influence of RF-EMR on the cell biology of human spermatozoa in vitro. PRINCIPAL FINDINGS: Purified human spermatozoa were exposed to radio-frequency electromagnetic radiation (RF-EMR) tuned to 1.8 GHz and covering a range of specific absorption rates (SAR) from 0.4 W/kg to 27.5 W/kg. In step with increasing SAR, motility and vitality were significantly reduced after RF-EMR exposure, while the mitochondrial generation of reactive oxygen species and DNA fragmentation were significantly elevated ($P<0.001$). Furthermore, we also observed highly significant relationships between SAR, the oxidative DNA damage bio-marker, 8-OH-dG, and DNA fragmentation after RF-EMR exposure. CONCLUSIONS: RF-EMR in both the power density and frequency range of mobile phones enhances mitochondrial reactive oxygen species generation by human spermatozoa, decreasing the motility and vitality of these cells while stimulating DNA base adduct formation and, ultimately DNA fragmentation. These findings have clear implications for the safety of extensive mobile phone use by males of reproductive age, potentially affecting both their fertility and the health and wellbeing of their offspring.

(NE) de Oliveira FM, Carmona AM, Ladeira C. Is mobile phone radiation genotoxic? An analysis of micronucleus frequency in exfoliated buccal cells. Mutat Res. 822:41-46, 2017. (HU. CE, GT)

Electromagnetic fields (EMF) are classified as "possibly carcinogenic" by the International Agency for Research on Cancer (IARC). Some publications have reported associations between EMF exposure and DNA damage, but many other studies contradict such findings. Cytomorphological changes, such as micronuclei (MN), indicative of genomic damage, are biomarkers of genotoxicity. To test whether mobile phone-associated EMF exposure affects the MN frequency in exfoliated buccal cells, we obtained cells smears from the left and right inner cheeks of healthy mobile phone users, aged 18-30 (n=86), who also completed a characterization survey. MN frequencies were tested for potential confounding factors and for duration of phone use and preferential side of mobile phone use. No relationship was observed between MN frequency and duration of mobile phone use in daily calls. Cells ipsilateral to mobile phone use did not present a statistically significantly higher MN frequency, compared to cells contralateral to exposure. A highly statistically significant ($p<0.0001$) increase in MN frequency was found in subjects reporting regular exposure to genotoxic agents. Therefore, our results suggest that mobile phone-associated EMF do not to induce MN formation in buccal cells at the observed exposure levels.

(E) Del Re B, Bersani F, Giorgi G. Effect of electromagnetic field exposure on the transcription of repetitive DNA elements in human cells. Electromagn Biol Med. 38(4):262-270, 2019. (VT, AE, GE, CS)

Repetitive DNA (RE-DNA) was long thought to be silent and inert; only recent research has shown that it can be transcribed and that transcription alteration can be induced by environmental stress conditions, causing human pathological effects. The aim of this study was to determine whether exposure to radiofrequency electromagnetic fields (RF-EMF) could affect the transcription of RE-DNA. To this purpose, three different human cell lines (HeLa, BE(2)C and

SH-SY5Y) were exposed to 900 MHz GSM-modulated RF-EMF at specific absorption rate of 1 W/kg or to sham. After exposure, mRNA levels of RE-DNA were evaluated through quantitative real-time PCR. The following RE-DNA types were investigated: Long Interspersed nucleotide Element 1, DNA alpha satellite and Human Endogenous Retroviruses-like sequences. When comparing cells exposed to RF-EMF versus control samples, different results were found for the three cell lines evaluated, indicating that RF-EMF exposure can significantly affect RE-DNA transcription and that the effects strongly depend on the cellular context and the tissue type. Further studies are needed to elucidate which molecular mechanisms could be involved.

(E) Del Vecchio G, Giuliani A, Fernandez M, Mesirca P, Bersani F, Pinto R, Ardoino L, Lovisolo GA, Giardino L, Calzà L. Continuous exposure to 900MHz GSM-modulated EMF alters morphological maturation of neural cells. Neurosci Lett. 455(3):173-177, 2009. (VT, AE, GE, DE)

The effects of radiofrequency electromagnetic field (RF-EMF) exposure on neuronal phenotype maturation have been studied in two different in vitro models: murine SN56 cholinergic cell line and rat primary cortical neurons. The samples were exposed at a dose of 1W/kg at 900 MHz GSM modulated. The phenotype analysis was carried out at 48 and 72 h (24 and 48 h of SN56 cell line differentiation) or at 24, 72, 120 h (2, 4 and 6 days in vitro for cortical neurons) of exposure, on live and immunolabeled neurons, and included the morphological study of neurite emission, outgrowth and branching. Moreover, cortical neurons were studied to detect alterations in the expression pattern of cytoskeleton regulating factors, e.g. beta-thymosin, and of early genes, e.g. c-Fos and c-Jun through real-time PCR on mRNA extracted after 24h exposure to EMF. We found that RF-EMF exposure reduced the number of neurites generated by both cell systems, and this alteration correlates to increased expression of beta-thymosin mRNA.

(E) Demsia G, Vlastos D, Matthopoulos DP. Effect of 910-MHz electromagnetic field on rat bone marrow. ScientificWorld Journal 4 Suppl 2:48-54, 2004. (VO, LE, GT)

Aiming to investigate the possibility of electromagnetic fields (EMF) developed by nonionizing radiation to be a noxious agent capable of inducing genotoxicity to humans, in the current study we have investigated the effect of 910-MHz EMF in rat bone marrow. Rats were exposed daily for 2 h over a period of 30 consecutive days. Studying bone marrow smears from EMF-exposed and sham-exposed animals, we observed an almost threefold increase of micronuclei (MN) in polychromatic erythrocytes (PCEs) after EMF exposure. An induction of MN was also observed in polymorphonuclear cells. The induction of MN in female rats was less than that in male rats. The results indicate that 910-MHz EMF could be considered as a noxious agent capable of producing genotoxic effects.

(E) Deshmukh PS, Megha K, Banerjee BD, Ahmed RS, Chandna S, Abegaonkar MP, Tripathi AK. Detection of low level microwave radiation induced deoxyribonucleic acid damage vis-à-vis genotoxicity in brain of Fischer rats. Toxicol Int. 20(1):19-24, 2013. (VO, LE, GT, LI)

BACKGROUND: Non-ionizing radiofrequency radiation has been increasingly used in industry, commerce, medicine and especially in mobile phone technology and has become a matter of serious concern in present time. OBJECTIVE: The present study was designed to investigate the possible deoxyribonucleic acid (DNA) damaging effects of low-level microwave radiation in brain of Fischer rats. MATERIALS AND METHODS: Experiments were performed on male Fischer rats exposed to microwave radiation for 30 days at three different frequencies: 900, 1800 and 2450 MHz. Animals were divided into 4 groups: Group I (Sham exposed): Animals not exposed to microwave radiation but kept under same conditions as that of other groups, Group II: Animals exposed to microwave radiation at frequency 900 MHz at specific absorption rate (SAR) 5.953 × 10(-4) W/kg, Group III: Animals exposed to 1800 MHz at SAR 5.835 × 10(-4) W/kg and Group IV: Animals exposed to 2450 MHz at SAR 6.672 × 10(-4) W/kg. At the end of the exposure period animals were sacrificed immediately and DNA damage in brain tissue was assessed using alkaline comet assay. RESULTS: In the present study, we demonstrated DNA damaging effects of low level microwave radiation in brain. CONCLUSION: We concluded that low SAR microwave radiation exposure at these frequencies may induce DNA strand breaks in brain tissue.

(E) Deshmukh PS, Nasare N, Megha K, Banerjee BD, Ahmed RS, Singh D, Abegaonkar MP, Tripathi AK, Mediratta PK. Cognitive Impairment and Neurogenotoxic Effects in Rats Exposed to Low-Intensity Microwave Radiation. Int J Toxicol. 34(3):24-290, 2015. (VO, LE, GT, LI)

The health hazard of microwave radiation (MWR) has become a recent subject of interest as a result of the enormous increase in mobile phone usage. The present study aimed to investigate the effects of chronic low-intensity microwave exposure on cognitive function, heat shock protein 70 (HSP70), and DNA damage in rat brain. Experiments were performed on male Fischer rats exposed to MWR for 180 days at 3 different frequencies, namely, 900, 1800 MHz, and 2450 MHz. Animals were divided into 4 groups: group I: sham exposed; group II: exposed to MWR at 900 MHz, specific absorption rate (SAR) 5.953×10^{-4} W/kg; group III: exposed to 1800 MHz, SAR 5.835×10^{-4} W/kg; and group IV: exposed to 2450 MHz, SAR 6.672×10^{-4} W/kg. All the rats were tested for cognitive function at the end of the exposure period and were subsequently sacrificed to collect brain. Level of HSP70 was estimated by enzyme-linked immunotarget assay and DNA damage was assessed using alkaline comet assay in all the groups. The results showed declined cognitive function, elevated HSP70 level, and DNA damage in the brain of microwave-exposed animals. The results indicated that, chronic low-intensity microwave exposure in the frequency range of 900 to 2450 MHz may cause hazardous effects on the brain.

(E) Deshmukh PS, Megha K, Nasare N, Banerjee BD, Ahmed RS, Abegaonkar MP, Tripathi AK, Mediratta PK. Effect of low level subchronic microwave radiation on rat brain. Biomed Environ Sci. 29(12):858-867, 2016. (VO, LE, GT, LI)

OBJECTIVE: The present study was designed to investigate the effects of subchronic low level microwave radiation (MWR) on cognitive function, heat shock protein 70 (HSP70) level and DNA damage in brain of Fischer rats. METHODS: Experiments were performed on male Fischer rats exposed to microwave radiation for 90 days at three different frequencies: 900, 1800, and

2450 MHz. Animals were divided into 4 groups: Group I: Sham exposed, Group II: animals exposed to microwave radiation at 900 MHz and specific absorption rate (SAR) 5.953 × 10-4 W/kg, Group III: animals exposed to 1800 MHz at SAR 5.835 × 10-4 W/kg and Group IV: animals exposed to 2450 MHz at SAR 6.672 × 10-4 W/kg. All the animals were tested for cognitive function using elevated plus maze and Morris water maze at the end of the exposure period and subsequently sacrificed to collect brain tissues. HSP70 levels were estimated by ELISA and DNA damage was assessed using alkaline comet assay. RESULTS: Microwave exposure at 900-2450 MHz with SAR values as mentioned above lead to decline in cognitive function, increase in HSP70 level and DNA damage in brain. CONCLUSION: The results of the present study suggest that low level microwave exposure at frequencies 900, 1800, and 2450 MHz may lead to hazardous effects on brain.

(E) Diem E, Schwarz C, Adlkofer F, Jahn O, Rudiger H. Non-thermal DNA breakage by mobile-phone radiation (1800MHz) in human fibroblasts and in transformed GFSH-R17 rat granulosa cells in vitro. Mutat Res. 583:178-183, 2005. (VT, AE, GT, WS)

Cultured human diploid fibroblasts and cultured rat granulosa cells were exposed to intermittent and continuous radiofrequency electromagnetic fields (RF-EMF) used in mobile phones, with different specific absorption rates (SAR) and different mobile-phone modulations. DNA strand breaks were determined by means of the alkaline and neutral comet assay. RF-EMF exposure (1800MHz; SAR 1.2 or 2W/kg; different modulations; during 4, 16 and 24h; intermittent 5min on/10min off or continuous wave) induced DNA single- and double-strand breaks. Effects occurred after 16h exposure in both cell types and after different mobile-phone modulations. The intermittent exposure showed a stronger effect in the comet assay than continuous exposure. Therefore we conclude that the induced DNA damage cannot be based on thermal effects.

(E) Ding Z, Xiang X, Li J, Wu S. Molecular Mechanism of Malignant Transformation of Balb/c-3T3 Cells Induced by Long-Term Exposure to 1800 MHz Radiofrequency Electromagnetic Radiation (RF-EMR). Bioengineering (Basel) 9(2):43, 2022. (VT, LE, GE)

Purpose: We aimed to investigate RF-EMR-induced cell malignant transformation.
Methods: We divided Balb/c-3T3 cells into sham and expo groups. The expo groups were exposed to a 1800 MHz RF continuous wave for 40 and 60 days, for 4 h per day. The sham group was sham-exposed. Cells were harvested for a cell transformation assay, transplantation in severe combined immune deficient (SCID) mice, soft agar clone formation detection, and a transwell assay. The mRNA microarray assay was used to declare key genes and pathways.
Results: The exposed Balb/c-3T3 cells showed a strong increase in cell proliferation and migration. Malignant transformation was observed in expo Balb/c-3T3 cells exposed for 40 days and 60 days, which was symbolized with visible foci and clone formation. Expo Balb/c-3T3 cells that were exposed for 40 days and 60 days produced visible tumors in the SCID mice. Lipid metabolism was the key biological process and pathway involved. The mevalonate (MVA) pathway was the key metabolic pathway. The interacted miRNAs could be further research targets to examine the molecular mechanism of the carcinogenic effects of long-term exposure.

Conclusion: Exposure for 40 and 60 days to 1800 MHz RF-EMR induced malignant transformation in Balb/c-3T3 cells at the SAR of 8.0 W/kg. We declared that lipid metabolism was the pivotal biological process and pathway. The MVA pathway was the key metabolic pathway.

(E) D'Silva MH, Swer RT, Anbalagan J, Rajesh B. Effect of Radiofrequency Radiation Emitted from 2G and 3G Cell Phone on Developing Liver of Chick Embryo - A Comparative Study. J Clin Diagn Res. 11(7):AC05-AC09, 2017. (CE, GE, IU, ME)

INTRODUCTION: The increasing scientific evidence of various health hazards on exposure of Radiofrequency Radiation (RFR) emitted from both the **cell phones** and base stations have caused significant media attention and public discussion in recent years. The mechanism of interaction of RF fields with developing tissues of children and fetuses may be different from that of adults due to their smaller physical size and variation in tissue electromagnetic properties. The present study may provide an insight into the basic mechanisms by which RF fields interact with developing tissues in an embryo. AIM: To evaluate the possible tissue and DNA damage in developing liver of chick embryo following chronic exposure to Ultra-High Frequency/Radiofrequency Radiation (UHF/RFR) emitted from 2G and 3G **cell phone**. MATERIALS AND METHODS: Fertilized chick embryos were incubated in four groups. Group A-experimental group exposed to 2G radiation (60 eggs), Group B- experimental group exposed to 3G radiation (60 eggs), Group C- sham exposed control group (60 eggs) and Group D- control group (48 eggs). On completion of scheduled duration, the embryos were collected and processed for routine histological studies to check structural changes in liver. The nuclear diameter and karyorrhexis changes of hepatocytes were analysed using oculometer and square reticule respectively. The liver procured from one batch of eggs from all the four groups was subjected to alkaline comet assay technique to assess DNA damage. The results were compared using one-way ANOVA test. RESULTS: In our study, the exposure of developing chick embryos to 2G and 3G **cell phone** radiations caused structural changes in liver in the form of dilated sinusoidal spaces with haemorrhage, increased vacuolations in cytoplasm, increased nuclear diameter and karyorrhexis and significantly increased DNA damage. CONCLUSION: The chronic exposure of chick embryo liver to RFR emitted from 2G and 3G **cell phone** resulted in various structural changes and DNA damage. The changes were more pronounced in 3G experimental group. Based on these findings it is necessary to create awareness among public about the possible ill effects of RFR exposure from **cell phone**.

(E) D'Silva MH, Swer RT, Anbalagan J, Bhargavan R. Assessment of DNA Damage in Chick Embryo Brains Exposed to 2G and 3G Cell Phone Radiation using Alkaline Comet Assay Technique. J Clin of Diagn Res. 15(1):AC01-AC04, 2021. (VO, LE, GT)

Introduction The cellular phones/mobile phones have emerged as the fastest growing man-made phenomenon ever discovered in the history. Controversies still exist among the scientific community regarding the ill-effects of Radiofrequency Radiation (RFR) exposure from cell phones on biological tissues. The present study will provide an insight into the basic mechanisms by which RF fields interact with developing brain in an embryo. Aim To assess the possible

Deoxyribonucleic Acid (DNA) damage in developing brain of chick embryo following chronic exposure to Ultra-High Frequency/Radiofrequency Radiation (UHF/RFR) emitted from 2G and 3G cell phone.Materials and Methods Fertilised hen eggs were divided into three groups. Experimental Group A (exposed to 2G radiation, 24 eggs), Experimental Group B (exposed to 3G radiation, 24 eggs) and Group C sham exposed control group (24 eggs). After the completion of scheduled duration of exposure (72 minutes per day), the chick embryos were sacrificed from 9^{th}-12^{th} day and the brains were dissected out. The chick embryo brains were then subjected to alkaline comet assay technique to assess the DNA damage. The results were statistically compared using one-way Analysis of Variance (ANOVA). Results In the present study, the exposure of chick embryo brains to 2G and 3G cell phone radiation caused increased mean comet length ($p<0.001$), mean tail length ($p<0.001$), mean percentage of DNA in the tail ($p<0.001$) and mean tail moment ($p<0.01$) suggestive of increased DNA damage. Conclusion The present study concludes that the RFR exposure caused significant increase in DNA damage in developing brain of chick embryos with changes more pronounced in 3G exposure group

(E) Duan W, Liu C, Zhang L, He M, Xu S, Chen C, Pi H, Gao P, Zhang Y, Zhong M, Yu Z, Zhou Z. Comparison of the genotoxic effects induced by 50 Hz extremely low-frequency electromagnetic fields and 1800 MHz radiofrequency electromagnetic fields in GC-2 cells. Radiat Res. 183(3):305-314, 2015. (VT, AE, GT, OX, WS, RP)

Extremely low-frequency electromagnetic fields (ELF-EMF) and radiofrequency electromagnetic fields (RF-EMF) have been considered to be possibly carcinogenic to humans. However, their genotoxic effects remain controversial. To make experiments controllable and results comparable, we standardized exposure conditions and explored the potential genotoxicity of 50 Hz ELF-EMF and 1800 MHz RF-EMF. A mouse spermatocyte-derived GC-2 cell line was intermittently (5 min on and 10 min off) exposed to 50 Hz ELF-EMF at an intensity of 1, 2 or 3 mT or to RF-EMF in GSM-Talk mode at the specific absorption rates (SAR) of 1, 2 or 4 W/kg. After exposure for 24 h, we found that neither ELF-EMF nor RF-EMF affected cell viability using Cell Counting Kit-8. Through the use of an alkaline comet assay and immunofluorescence against γ-H2AX foci, we found that ELF-EMF exposure resulted in a significant increase of DNA strand breaks at 3 mT, whereas RF-EMF exposure had insufficient energy to induce such effects. Using a formamidopyrimidine DNA glycosylase (FPG)-modified alkaline comet assay, we observed that RF-EMF exposure significantly induced oxidative DNA base damage at a SAR value of 4 W/kg, whereas ELF-EMF exposure did not. Our results suggest that both ELF-EMF and RF-EMF under the same experimental conditions may produce genotoxicity at relative high intensities, but they create different patterns of DNA damage. Therefore, the potential mechanisms underlying the genotoxicity of different frequency electromagnetic fields may be different.

(NE) Durdik M, Kosik P, Markova E, Somsedikova A, Gajdosechova B, Nikitina E, Horvathova E, Kozics K, Davis D, Belyaev I. Microwaves from mobile phone induce reactive oxygen species but not DNA damage, preleukemic fusion genes and apoptosis in hematopoietic stem/progenitor cells. Sci Rep. 9(1):16182, 2019. (VT, AE, GT)

Exposure to electromagnetic fields (EMF) has been associated with the increased risk of childhood leukemia, which arises from mutations induced within hematopoietic stem cells often through preleukemic fusion genes (PFG). In this study we investigated whether exposure to microwaves (MW) emitted by mobile phones could induce various biochemical markers of cellular damage including reactive oxygen species (ROS), DNA single and double strand breaks, PFG, and apoptosis in umbilical cord blood (UCB) cells including CD34+ hematopoietic stem/progenitor cells. UCB cells were exposed to MW pulsed signals from GSM900/UMTS test-mobile phone and ROS, apoptosis, DNA damage, and PFG were analyzed using flow cytometry, automated fluorescent microscopy, imaging flow cytometry, comet assay, and RT-qPCR. In general, no persisting difference in DNA damage, PFG and apoptosis between exposed and sham-exposed samples was detected. However, we found increased ROS level after 1 h of UMTS exposure that was not evident 3 h post-exposure. We also found that the level of ROS rise with the higher degree of cellular differentiation. Our data show that UCB cells exposed to pulsed MW developed transient increase in ROS that did not result in sustained DNA damage and apoptosis.

(E) Eker ED, Arslan B, Yildirim M, Akar A, Aras N. The effect of exposure to 1800 MHz radiofrequency radiation on epidermal growth factor, caspase-3, Hsp27 and p38MAPK gene expressions in the rat eye. Bratisl Lek Listy. 119(9):588-592, 2018. (VO, LE, LI, GE)

Radiofrequency electromagnetic fields (RF-EMF) may induce DNA damage and oxidative stress in human lens epithelial cells (LECs). We aimed to investigate the expression levels of heat shock protein 27 (Hsp27), p38 mitogen-activated protein kinase (p38MAPK), epidermal growth factor receptor (EGFR) and caspase-3 gene expression levels in rat eye that was exposed to 1800 MHz RF-EMF. METHODS: Thirty-seven female Wistar albino rats were divided into three groups. The rats in the study group (n = 9) were exposed to 1800 MHz RF-EMF at an electric field 6.8 ± 0.1 V/m and 0.06 W/kg specific absorption rate (SAR) for 2 hours per day for eight weeks. Sham group (n = 9) was kept under similar conditions as the exposed group without exposure to RF-EMF. The rats in all three groups were sacrificed and their eyes were removed. Hsp27, p38MAPK, EGFR, caspase-3 gene expression levels were investigated in detail with real-time polymerase chain reactions (Real-Time PCR). RESULTS: caspase-3 and p38MAPK gene expression were significantly upregulated in the ocular tissues following exposure to RF-EMF ($p < 0.05$). CONCLUSION: According to our findings, eye cells recognize EMF as a stress factor, and in response, activate caspase-3 and p38MAPK gene expressions. These results confirm that RF-EMF can cause cellular damage in rat ocular cells (Tab. 2, Fig. 3, Ref. 37).

(E) Engelmann JC, Deeken R, Müller T, Nimtz G, Roelfsema MR, Hedrich R. Is gene activity in plant cells affected by UMTS-irradiation? A whole genome approach. Adv Appl Bioinform Chem. 1:71-83, 2008. (VT, AE, GE)

Mobile phone technology makes use of radio frequency (RF) electromagnetic fields transmitted through a dense network of base stations in Europe. Possible harmful effects of RF fields on humans and animals are discussed, but their effect on plants has received little attention. In search for physiological processes of plant cells sensitive to RF fields, cell suspension cultures of Arabidopsis thaliana were exposed for 24 h to a RF field protocol representing typical microwave exposition in an urban environment. mRNA of exposed cultures and controls was

used to hybridize Affymetrix-ATH1 whole genome microarrays. Differential expression analysis revealed significant changes in transcription of 10 genes, but they did not exceed a fold change of 2.5. Besides that 3 of them are dark-inducible, their functions do not point to any known responses of plants to environmental stimuli. The changes in transcription of these genes were compared with published microarray datasets and revealed a weak similarity of the microwave to light treatment experiments. Considering the large changes described in published experiments, it is questionable if the small alterations caused by a 24 h continuous microwave exposure would have any impact on the growth and reproduction of whole plants.

(E) Ergun DD, Ozsobaci NP, Yilmaz T, Ozcelik D, Kalkan MT. Zinc affects nuclear factor kappa b and DNA methyltransferase activity in C3H cancer fibroblast cells induced by a 2100 MHz electromagnetic field. Electromagn Biol Med 41(1):93-100, 2022. (VT, AE, GE, IX, EP)

The use of mobile phones is becoming widespread with the development of technology, and as a result, its effects on human health are becoming more and more important every day. Studies have reported that the electromagnetic field (EMF) emitted by mobile phones may have adverse effects on the biological systems. In order to evaluate the effect of zinc (Zn) on C3H cancer fibroblast cells exposed to 2100 MHz EMF, we analyzed cell viability%, nuclear factor kappa b (NF-κB) and DNA methyltransferase (DNMT) activities. Cells were divided to following groups: Control, sham control, 2100 MHz EMF, 50 μM Zn + 2100 MHz EMF, 100 μM Zn + 2100 MHz EMF, and 200 μM Zn + 2100 MHz EMF for 2 h. We measurement cell viability, NF-κB and DNMT activities. There was increased cell viability % in the 2100 MHz EMF group compared to the control group, while the cell viability % was decreased in the 50, 100 and 200 μM Zn + 2100 MHz EMF groups compared to 2100 MHz EMF. NF-κB and DNMT activities were a significant increase in the 2100 MHz EMF group compared to the control group, although were statistically decreased in the 50, 100 and 200 μM Zn + 2100 MHz EMF groups compared to the 2100 MHz EMF group. Our results demonstrate that 2100 MHz EMF exposure in cancer fibroblast cells induce NF-κB and DNMT activities, whereas zinc supplementation reduce NF-κB and DNMT activities-induced 2100 MHz EMF.

(E) Esmekaya MA, Aytekin E, Ozgur E, Güler G, Ergun MA, Omeroğlu S, Seyhan N. Mutagenic and morphologic impacts of 1.8GHz radiofrequency radiation on human peripheral blood lymphocytes (hPBLs) and possible protective role of pre-treatment with Ginkgo biloba (EGb 761). Sci Total Environ. 410-411:59-64, 2011. (VT, AE, GT, OX)

The mutagenic and morphologic effects of 1.8GHz Global System for Mobile Communications (GSM) modulated RF (radiofrequency) radiation alone and in combination with Ginkgo biloba (EGb 761) pre-treatment in human peripheral blood lymphocytes (hPBLs) were investigated in this study using Sister Chromatid Exchange (SCE) and electron microscopy. Cell viability was assessed with 3-(4, 5-dimethylthiazol-2-yl)-2, 5-diphenyltetrazolium bromide (MTT) reduction assay. The lymphocyte cultures were exposed to GSM modulated RF radiation at 1.8GHz for 6, 8, 24 and 48h with and without EGb 761. We observed morphological changes in pulse-modulated RF radiated lymphocytes. Longer exposure periods led to destruction of organelle and nucleus structures. Chromatin change and the loss of mitochondrial crista occurred in cells

exposed to RF for 8h and 24h and were more pronounced in cells exposed for 48h. Cytoplasmic lysis and destruction of membrane integrity of cells and nuclei were also seen in 48h RF exposed cells. There was a significant increase ($p<0.05$) in SCE frequency in RF exposed lymphocytes compared to sham controls. EGb 761 pre-treatment significantly decreased SCE from RF radiation. RF radiation also inhibited cell viability in a time dependent manner. The inhibitory effects of RF radiation on the growth of lymphoctes were marked in longer exposure periods. EGb 761 pre-treatment significantly increased cell viability in RF+EGb 761 treated groups at 8 and 24h when compared to RF exposed groups alone. The results of our study showed that RF radiation affects cell morphology, increases SCE and inhibits cell proliferation. However, EGb 761 has a protective role against RF induced mutagenity. We concluded that RF radiation induces chromosomal damage in hPBLs but this damage may be reduced by EGb 761 pre-treatment.

(NE) Falzone N, Huyser C, Franken DR, Leszczynski D. Mobile phone radiation does not induce pro-apoptosis effects in human spermatozoa. Radiat Res 174:169-176, 2010. (VT, AE. GT, OX)

Recent reports suggest that mobile phone radiation may diminish male fertility. However, the effects of this radiation on human spermatozoa are largely unknown. The present study examined effects of the radiation on induction of apoptosis-related properties in human spermatozoa. Ejaculated, density-purified, highly motile human spermatozoa were exposed to mobile phone radiation at specific absorption rates (SARs) of 2.0 and 5.7 W/kg. At various times after exposure, flow cytometry was used to examine caspase 3 activity, externalization of phosphatidylserine (PS), induction of DNA strand breaks, and generation of reactive oxygen species. Mobile phone radiation had no statistically significant effect on any of the parameters studied. This suggests that the impairment of fertility reported in some studies was not caused by the induction of apoptosis in spermatozoa.

(E) Ferreira AR, Knakievicz T, de Bittencourt Pasquali MA, Gelain DP, Dal-Pizzol F, Fernandez CE, de Almeida de Salles AA, Ferreira HB, Moreira JC. Ultra high frequency-electromagnetic field irradiation during pregnancy leads to an increase in erythrocytes micronuclei incidence in rat offspring. Life Sci 80: 43-50, 2006. (VO, LE, GT, OX, DE)

Mobile telephones and their base stations are an important ultra high frequency-electromagnetic field (UHF-EMF) source and their utilization is increasing all over the world. Epidemiological studies suggested that low energy UHF-EMF emitted from a cellular telephone may cause biological effects, such as DNA damage and changes on oxidative metabolism. An in vivo mammalian cytogenetic test, the micronucleus (MN) assay, was used to investigate the occurrence of chromosomal damage in erythrocytes from rat offspring exposed to a non-thermal UHF-EMF from a cellular phone during their embryogenesis; the irradiated group showed a significant increase in MN occurrence. In order to investigate if UHF-EMF could also alter oxidative parameters in the peripheral blood and in the liver - an important hematopoietic tissue in rat embryos and newborns - we also measured the activity of antioxidant enzymes, quantified total sulfhydryl content, protein carbonyl groups, thiobarbituric acid-reactive species and total non-enzymatic antioxidant defense. No significant differences were found in any oxidative parameter of offspring blood and liver. The average number of pups in each litter has also not

been significantly altered. Our results suggest that, under our experimental conditions, UHF-EMF is able to induce a genotoxic response in hematopoietic tissue during the embryogenesis through an unknown mechanism.

(NE) Finnie JW, Cai Z, Blumbergs PC, Manavis J, Kuchel TR. Expression of the immediate early gene, c-fos, in fetal brain after whole of gestation exposure of pregnant mice to global system for mobile communication microwaves. Pathology. 38(4):333-335, 2006. (VO, LE, GE, DE)

AIMS: To study immediate early gene, c-fos, expression as a marker of neural stress after whole of gestation exposure of the fetal mouse brain to mobile telephone-type radiofrequency fields. METHODS: Using a purpose-designed exposure system at 900 MHz, pregnant mice were given a single, far-field, whole body exposure at a specific absorption rate of 4 W/kg for 60 min/day from day 1 to day 19 of gestation. Pregnant control mice were sham-exposed or freely mobile in a cage without further restraint. Immediately prior to parturition on gestational day 19, fetal heads were collected, fixed in 4% paraformaldehyde and paraffin embedded. Any stress response in the brain was detected by c-fos immunohistochemistry in the cerebral cortex, basal ganglia, thalamus, hippocampus, midbrain, cerebellum and medulla. RESULTS: c-fos expression was of limited, but consistent, neuroanatomical distribution and there was no difference in immunoreactivity between exposed and control brains. CONCLUSION: In this animal model, no stress response was detected in the fetal brain using c-fos immunohistochemistry after whole of gestation exposure to mobile telephony.

(E) Fragopoulou AF, Polyzos A, Papadopoulou MD, Sansone A, Manta AK, Balafas E, Kostomitsopoulos N, Skouroliakou A, Chatgilialoglu C, Georgakilas A, Stravopodis DJ, Ferreri C, Thanos D, Margaritis LH. Hippocampal lipidome and transcriptome profile alterations triggered by acute exposure of mice to GSM 1800 MHz mobile phone radiation: An exploratory study. Brain Behav. 8(6):e01001, 2018. (VO, AE, GE)

Background: The widespread use of wireless devices during the last decades is raising concerns about adverse health effects of the radiofrequency electromagnetic radiation (RF-EMR) emitted from these devices. Recent research is focusing on unraveling the underlying mechanisms of RF-EMR and potential cellular targets. The "omics" high-throughput approaches are powerful tools to investigate the global effects of RF-EMR on cellular physiology. **Methods:** In this work, C57BL/6 adult male mice were whole-body exposed ($n_{Exp} = 8$) for 2 hr to GSM 1800 MHz mobile phone radiation at an average electric field intensity range of 4.3-17.5 V/m or sham-exposed ($n_{SE} = 8$), and the RF-EMR effects on the hippocampal lipidome and transcriptome profiles were assessed 6 hr later. **Results:** The data analysis of the phospholipid fatty acid residues revealed that the levels of four fatty acids [16:0, 16:1 (6c + 7c), 18:1 9c, eicosapentaenoic acid omega-3 (EPA, 20:5 ω3)] and the two fatty acid sums of saturated and monounsaturated fatty acids (SFA and MUFA) were significantly altered ($p < 0.05$) in the exposed group. The observed changes indicate a membrane remodeling response of the tissue phospholipids after nonionizing radiation exposure, reducing SFA and EPA, while increasing MUFA residues. The microarray data analysis demonstrated that the expression of 178 genes changed significantly ($p < 0.05$) between the two groups, revealing an impact on genes involved in critical biological processes, such as cell cycle, DNA replication and repair, cell death, cell

signaling, nervous system development and function, immune system response, lipid metabolism, and carcinogenesis. **Conclusions:** This study provides preliminary evidence that mobile phone radiation induces hippocampal lipidome and transcriptome changes that may explain the brain proteome changes and memory deficits previously shown by our group.

(E) Franchini V, Regalbuto E, De Amicis A, De Sanctis S, Di Cristofaro S, Coluzzi E, Marinaccio J, Sgura A, Ceccuzzi S, Doria A, Gallerano GP, Giovenale E, Ravera GL, Bei R, Benvenuto M, Modesti A, Masuelli L, Lista F. Genotoxic effects in human fibroblasts exposed to microwave radiation. Health Phys. 115(1):126-139, 2018a. (VT, AE, GT)

In the last decades, technological development has led to an increasing use of devices and systems based on microwave radiation. The increased employment of these devices has elicited questions about the potential long-term health consequences associated with microwave radiation exposure. From this perspective, biological effects of microwave radiation have been the focus of many studies, but the reported scientific data are unclear and contradictory. The aim of this study is to evaluate the potential genotoxic and cellular effects associated with in vitro exposure of human fetal and adult fibroblasts to microwave radiation at the frequency of 25 GHz. For this purpose, several genetic and biological end points were evaluated. Results obtained from comet assay, phosphorylation of H2AX histone, and antikinetochore antibody (CREST)-negative micronuclei frequency excluded direct DNA damage to human fetal and adult fibroblasts exposed to microwaves. No induction of apoptosis or changes in prosurvival signalling proteins were detected. Moreover, CREST analysis showed for both the cell lines an increase in the total number of micronuclei and centromere positive micronuclei in exposed samples, indicating aneuploidy induction due to chromosome loss.

(E) Franchini V, De Sanctis S, Marinaccio J, De Amicis A, Coluzzi E, Di Cristofaro S, Lista F, Regalbuto E, Doria A, Giovenale E, Gallerano GP, Bei R, Benvenuto M, Masuelli L, Udroiu I, Sgura A. Study of the effects of 0.15 terahertz radiation on genome integrity of adult fibroblasts. Environ Mol Mutagen. 59(6):476-487, 2018b. (VT, AE, GT)

The applications of Terahertz (THz) technologies have significantly developed in recent years, and the complete understanding of the biological effects of exposure to THz radiation is becoming increasingly important. In a previous study, we found that THz radiation induced genomic damage in fetal fibroblasts. Although these cells demonstrated to be a useful model, exposure of human foetuses to THz radiation is highly improbable. Conversely, THz irradiation of adult dermal tissues is cause of possible concern for some professional and nonprofessional categories. Therefore, we extended our study to the investigation of the effects of THz radiation on adult fibroblasts (HDF). In this work, the effects of THz exposure on HDF cells genome integrity, cell cycle, cytological ultrastructure and proteins expression were assessed. Results of centromere-negative micronuclei frequencies, phosphorylation of H2AX histone, and telomere length modulation indicated no induction of DNA damage. Concordantly, no changes in the expression of proteins associated with DNA damage sensing and repair were detected. Conversely, our results showed an increase of centromere-positive micronuclei frequencies and chromosomal nondisjunction events, indicating induction of aneuploidy. Therefore, our results indicate that THz radiation

exposure may affect genome integrity through aneugenic effects, and not by DNA breakage. Our findings are compared to published studies, and possible biophysical mechanisms are discussed.

(E) Franzellitti S, Valbonesi P, Contin A, Biondi C, Fabbri E. HSP70 expression in human trophoblast cells exposed to different 1.8 Ghz mobile phone signals. Radiat Res. 170(4):488-497, 2008. (VT, AE, GE)

The heat-shock proteins (HSPs) are important cellular stress markers and have been proposed as candidates to infer biological effects of high-frequency electromagnetic fields (EMFs). In the current study, HSP70 gene and protein expression were evaluated in cells of the human trophoblast cell line HTR-8/SVneo after prolonged exposure (4 to 24 h) to 1.8 GHz continuous-wave (CW) and different GSM signals (GSM-217Hz and GSM-Talk) to assess the possible effects of time and modulation schemes on cell responses. Inducible HSP70 protein expression was not modified by high-frequency EMFs under any condition tested. The inducible HSP70A, HSP70B and the constitutive HSC70 transcripts did not change in cells exposed to high-frequency EMFs with the different modulation schemes. Instead, levels of the inducible HSP70C transcript were significantly enhanced after 24 h exposure to GSM-217Hz signals and reduced after 4 and 16 h exposure to GSM-Talk signals. As in other cell systems, in HTR-8/SVneo cells the response to high-frequency EMFs was detected at the mRNA level after exposure to amplitude-modulated GSM signals. The present results suggest that the expression analysis for multiple transcripts, though encoding the same or similar protein products, can be highly informative and may account for subtle changes not detected at the protein level.

(E) Franzellitti S, Valbonesi P, Ciancaglini N, Biondi C, Contin A, Bersani F, Fabbri E. Transient DNA damage induced by high-frequency electromagnetic fields (GSM 1.8 GHz) in the human trophoblast HTR-8/SVneo cell line evaluated with the alkaline comet assay. Mutat Res 683(1-2):35-42, 2010. (VT, AE, GT, WS)

One of the most controversial issue regarding high-frequency electromagnetic fields (HF-EMF) is their putative capacity to affect DNA integrity. This is of particular concern due to the increasing use of HF-EMF in communication technologies, including mobile phones. Although epidemiological studies report no detrimental effects on human health, the possible disturbance generated by HF-EMF on cell physiology remains controversial. In addition, the question remains as to whether cells are able to compensate their potential effects. We have previously reported that a 1-h exposure to amplitude-modulated 1.8 GHz sinusoidal waves (GSM-217 Hz, SAR=2 W/kg) largely used in mobile telephony did not cause increased levels of primary DNA damage in human trophoblast HTR-8/SVneo cells. Nevertheless, further investigations on trophoblast cell responses after exposure to GSM signals of different types and durations were considered of interest. In the present work, HTR-8/SVneo cells were exposed for 4, 16 or 24h to 1.8 GHz continuous wave (CW) and different GSM signals, namely GSM-217 Hz and GSM-Talk (intermittent exposure: 5 min field on, 10 min field off). The alkaline comet assay was used to evaluate primary DNA damages and/or strand breaks due to uncompleted repair processes in HF-EMF exposed samples. The amplitude-modulated signals GSM-217 Hz and GSM-Talk

induced a significant increase in comet parameters in trophoblast cells after 16 and 24h of exposure, while the un-modulated CW was ineffective. However, alterations were rapidly recovered and the DNA integrity of HF-EMF exposed cells was similar to that of sham-exposed cells within 2h of recovery in the absence irradiation. Our data suggest that HF-EMF with a carrier frequency and modulation scheme typical of the GSM signal may affect the DNA integrity.

(NE) Fritze K, Wiessner C, Kuster N, Sommer C, Gass P, Hermann DM, Kiessling M, Hossmann KA. Effect of global system for mobile communication microwave exposure on the genomic response of the rat brain. Neuroscience. 81(3):627-639, 1997. (VO, AE, GE)

The acute effect of global system for mobile communication (GSM) microwave exposure on the genomic response of the central nervous system was studied in rats by measuring changes in the messenger RNAs of hsp70, the transcription factor genes c-fos and c-jun and the glial structural gene GFAP using in situ hybridization histochemistry. Protein products of transcription factors, stress proteins and marker proteins of astroglial and microglial activation were assessed by immunocytochemistry. Cell proliferation was evaluated by bromodeoxyuridine incorporation. A special GSM radiofrequency test set, connected to a commercial cellular phone operating in the discontinuous transmission mode, was used to simulate GSM exposure. The study was conducted at time averaged and brain averaged specific absorption rates of 0.3 W/kg (GSM exposure), 1.5 W/kg (GSM exposure) and 7.5 W/kg (continuous wave exposure), respectively. Immediately after exposure, in situ hybridization revealed slight induction of hsp70 messenger RNA in the cerebellum and hippocampus after 7.5 W/kg exposure, but not at lower intensities. A slightly increased expression of c-fos messenger RNA was observed in the cerebellum, neocortex and piriform cortex of all groups subjected to immobilization, but no differences were found amongst different exposure conditions. C-jun and GFAP messenger RNAs did not increase in any of the experimental groups. 24 h after exposure, immunocytochemical analysis of FOS and JUN proteins (c-FOS, FOS B, c-JUN JUN B, JUN D), of HSP70 or of KROX-20 and -24 did not reveal any alterations. Seven days after exposure, neither increased cell proliferation nor altered expression of astroglial and microglial marker proteins were observed. In conclusion, acute high intensity microwave exposure of immobilized rats may induce some minor stress response but does not result in lasting adaptive or reactive changes of the brain.

(E) Fucic A, Garaj-Vrhovac V, Skara M, Dimitrovic B, X-rays, microwaves and vinyl chloride monomer: their clastogenic and aneugenic activity, using the micronucleus assay on human lymphocytes. Mutat Res 282(4):265-271, 1992. (VT. AE, GT)

Chromosome aberration assays, sister-chromatid exchange techniques and micronucleus assays are commonly used methods for biomonitoring genetic material damaged by chemical or physical agents. On the other hand, their aneugenic activity, which can lead to hypoploidy and may also be associated with carcinogenesis, has not been thoroughly investigated. In our study we chose the micronucleus assay with a new mathematical approach to separate clastogenic from aneugenic activity of three well-known mutagens (vinyl chloride monomer, X-rays and microwaves) on the genome of human somatic cells. The comparison of frequencies of size distribution of micronuclei in the lymphocytes of humans exposed to each of these three mutagens showed that X-rays and microwaves were preferentially clastogens while vinyl

chloride monomer showed aneugenic activity as well. Microwaves possess some mutagenic characteristics typical of chemical mutagens

(E) Furtado-Filho OV, Borba JB, Dallegrave A, Pizzolato TM, Henriques JA, Moreira JC, Saffi J. Effect of 950 MHz UHF electromagnetic radiation on biomarkers of oxidative damage, metabolism of UFA and antioxidants in the livers of young rats of different ages. Int J Radiat Biol. 90(2):159-168, 2014. (VO, LE, GT, OX)

Purpose: To assess the effect of 950 MHz ultra-high-frequency electromagnetic radiation (UHF EMR) on biomarkers of oxidative damage, as well as to verify the concentration of unsaturated fatty acids (UFA) and the expression of the catalase in the livers of rats of different ages. Materials and methods: Twelve rats were equally divided into two groups as controls (CR) and exposed (ER), for each age (0, 6, 15 and 30 days). Radiation exposure lasted half an hour per day for up to 51 days (21 days of gestation and 6, 15 or 30 days of life outside the womb). The specific absorption rate (SAR) ranged from 1.3-1.0 W/kg. The damage to lipids, proteins and DNA was verified by thiobarbituric acid reactive substances (TBARS), protein carbonyls and comets, respectively. UFA were determined by gas chromatography with a flame ionization detector. The expression of catalase was by Western blotting. Results: The neonates had low levels of TBARS and concentrations of UFA after exposure. There was no age difference in the accumulation of protein carbonyls for any age. The DNA damage of ER 15 or 30 days was different. The exposed neonates exhibited lower expression of catalase. Conclusions: 950 MHz UHF EMR does not cause oxidative stress (OS), and it is not genotoxic to the livers of neonates or those of 6 and 15 day old rats, but it changes the concentrations of polyunsaturated fatty acid (PUFA) in neonates. For rats of 30 days, no OS, but it is genotoxic to the livers of ER to total body irradiation.

(NE) Furtado-Filho OV, Borba JB, Maraschin T, Souza LM, Jose JA, Moreira CF, Saffi J. Effects of chronic exposure to 950 MHz ultra-high-frequency electromagnetic radiation on reactive oxygen species metabolism in the right and left cerebral cortex of young rats of different ages. Int J Radiat Biol. 91(11):891-897, 2015. (VO, LE, GT)

PURPOSE: To assess the effect of 950 MHz ultra-high-frequency electromagnetic radiation (UHF-EMR) on biomarkers of oxidative damage to DNA, proteins and lipids in the left cerebral cortex (LCC) and right cerebral cortex (RCC) of neonate and 6-day-old rats. MATERIALS AND METHODS: Twelve rats were equally divided into two groups as controls (CR) and exposed (ER), for each age (0 and 6 days). The LCC and RCC were examined in ER and CR after exposure. Radiation exposure lasted half an hour per day for up to 27 days (throughout pregnancy and 6 days postnatal). The specific absorption rate ranged from 1.32 - 1.14 W/kg. The damage to lipids, proteins and DNA was verified by thiobarbituric acid reactive substances, carbonylated proteins (CP) and comets, respectively. The concentration of glucose in the peripheral blood of the rats was measured by the Accu-Chek Active Kit due to increased CP in RCC. RESULTS: In neonates, no modification of the biomarkers tested was detected. On the other hand, there was an increase in the levels of CP in the RCC of the 6-day-old ER. Interestingly, the

concentration of blood glucose was decreased in this group. CONCLUSIONS: Our results indicate that there is no genotoxicity and oxidative stress in neonates and 6 days rats. However, the RCC had the highest concentration of CP that do not seem to be a consequence of oxidative stress. This study is the first to demonstrate the use of UHF-EMR causes different damage responses to proteins in the LCC and RCC.

(E) Gadhia PK, Shah T, Mistry A, Pithawala M, Tamakuwala D. A preliminary study to assess possible chromosomal damage among users of digital mobile phones. Electromag Biol Med 22:149-159, 2003. (HU, LE, GT)

In a preliminary study to examine possible lymphocyte chromosomal damage, we have tested two cytogenetic endpoints, namely, chromosomal aberrations (CA) and sister chromatid exchange frequencies (SCE), in 24 mobile phone users (12 nonsmoker–nonalcoholic subjects and 12 smoker–alcoholics), who used digital mobile phones for at least 2 years, employing Gaussian Minimum Shift Keying modulations with uplink frequencies at 935–960 MHz. and downlinks at 890–915 MHz. For comparison, the control study group included another 24 individuals, matched according to their age, sex, drinking and smoking habits, as well as similar health status, working habits, and professional careers; but did not use mobile phones. Blood samples of 12 mobile users (6 smoker–alcoholic and 6 nonsmoker–nonalcoholic) and 12 controls (identical to mobile users in every respect) were further treated with a known mutagen Mitomycin-C (MMC) to find out comutagenic/synergistic effect. A complete blood picture for each individual was assessed with an automatic particle cell counter. There was a significant increase ($P < 0.05$) in dicentric chromosomes among mobile users who were smoker–alcoholic as compared to nonsmoker–nonalcoholic; the same held true for controls of both types. After MMC treatment, there was a significant increase in dicentrics ($P < 0.05$) and ring chromosomes ($P < 0.001$) in both smoker–alcoholic and nonsmoker–nonalcoholic mobile users when compared with the controls. Although SCEs showed a significant increase among mobile users, no change in cell cycle progression was noted. The hematological picture showed only minor variations between mobile users and controls.

(E) Gajski G, Garaj-Vrhovac V. Radioprotective effects of honeybee venom (Apismellifera) against 915-MHz microwave radiation-induced DNA damage in wistar rat lymphocytes: in vitro study. Int J Toxicol 28:88-98, 2009. (VT, AE, GT, OX, IX)

The aim of this study is to investigate the radioprotective effect of bee venom against DNA damage induced by 915-MHz microwave radiation (specific absorption rate of 0.6 W/kg) in Wistar rats. Whole blood lymphocytes of Wistar rats are treated with 1 microg/mL bee venom 4 hours prior to and immediately before irradiation. Standard and formamidopyrimidine-DNA glycosylase (Fpg)-modified comet assays are used to assess basal and oxidative DNA damage produced by reactive oxygen species. Bee venom shows a decrease in DNA damage compared with irradiated samples. Parameters of Fpg-modified comet assay are statistically different from controls, making this assay more sensitive and suggesting that oxidative stress is a possible mechanism of DNA damage induction. Bee venom is demonstrated to have a radioprotective

effect against basal and oxidative DNA damage. Furthermore, bee venom is not genotoxic and does not produce oxidative damage in the low concentrations used in this study.

(E) Gandhi G, Anita, Genetic damage in mobile phone users: some preliminary findings. Ind J Hum Genet 11:99-104, 2005. (HU, LE, GT)

BACKGROUND: The impact of microwave (MW)/radio frequency radiation (RFR) on important biological parameters is probably more than a simply thermal one. Exposure to radio frequency (RF) signals generated by the use of cellular telephones have increased dramatically and reported to affect physiological, neurological, cognitive and behavioural changes and to induce, initiate and promote carcinogenesis. Genotoxicity of RFR has also been reported in various test systems after in vitro and/or in vivo exposure but none in mobile phone users. AIMS: In the present study, DNA and chromosomal damage investigations were carried out on the peripheral blood lymphocytes of individuals using mobile phones, being exposed to MW frequency ranging from 800 to 2000 MHz. METHODS: DNA damage was assessed using the single cell gel electrophoresis assay and aneugenic and clastogenic damage by the in vivo capillary blood micronucleus test (MNT) in a total of 24 mobile phone users. RESULTS: Mean comet tail length (26.76 ± 0.054 mm; 39.75% of cells damaged) in mobile phone users was highly significant from that in the control group. The in vivo capillary blood MNT also revealed highly significant (0.25) frequency of micronucleated (MNd) cells. CONCLUSIONS: These results highlight a correlation between mobile phone use (exposure to RFR) and genetic damage and require interim public health actions in the wake of widespread use of mobile telephony.

(E) Gandhi G, Singh P. Cytogenetic damage in mobile phone users: preliminary data. Int J Hum Genet 5:259-265, 2005. (HU, LE, GT)

Mobile telephones, sometimes called cellular (cell) phones or handies, are now an integral part of modern life. The mobile phone handsets are low-powered radiofrequency transmitters, emitting maximum powers in the range of 0.2 to 0.6 watts. Scientific concenrns have increased sufficiently over the possible hazard to health from using cell phones. The reported adverse health effects include physiological, behavioural and cognitive changes as well as tumour formation and genetic damage. However findings are controversial and no consensus exists. Genotoxicity has been observed either in lower organisms or in vitro studies. The aim of the present study hence was to detect any cytogenertic damage in mobile phone users by analysing short term peripheral lymphocyte cultures for chromosomal aberrations and the buccal mucosal cells for micronuclei (aneugenicity and clastogenicity). The results revealed increased number of micronucleated buccal cells and cytological abnormalities in cultured lymphocytes indicating the genotoxic response from mobile phone use.

(E) Gandhi G, Kaur G, Nisar U. A cross-sectional case control study on genetic damage in individuals residing in the vicinity of a mobile phone base station. Electromagn Biol Med. 34(4):344-354, 2015a. (HU, LE, GT)

Mobile phone base stations facilitate good communication, but the continuously emitting radiations from these stations have raised health concerns. Hence in this study, genetic damage using the single cell gel electrophoresis (comet) assay was assessed in peripheral blood leukocytes of individuals residing in the vicinity of a mobile phone base station and comparing it

to that in healthy controls. The power density in the area within 300 m from the base station exceeded the permissive limits and was significantly ($p = 0.000$) higher compared to the area from where control samples were collected. The study participants comprised 63 persons with residences near a mobile phone tower, and 28 healthy controls matched for gender, age, alcohol drinking and occupational sub-groups. Genetic damage parameters of DNA migration length, damage frequency (DF) and damage index were significantly ($p = 0.000$) elevated in the sample group compared to respective values in healthy controls. The female residents ($n = 25$) of the sample group had significantly ($p = 0.004$) elevated DF than the male residents ($n = 38$). The linear regression analysis further revealed daily mobile phone usage, location of residence and power density as significant predictors of genetic damage. The genetic damage evident in the participants of this study needs to be addressed against future disease-risk, which in addition to neurodegenerative disorders, may lead to cancer.

(E) Gandhi, G., Singh P, Kaur G. Perspectives revisited - The buccal cytome assay in mobile phone users. Int. J. Hum. genet. 15:173–182, 2015b. (HU, LE, GT)

Buccal cell preparations previously scored for micronuclei were re-investigated for genomic instability and other biomarkers to assess DNA damage, cell-proliferation and cell-death in healthy mobile phone users (n=25; 30.96±2.09y) using mobile phones for 3-5y and the non-mobile phones users (n=25; 32.28±2.01y) according to the buccal micronucleus cytome (BMCyt) assay which was then not available. The frequency of micronuclei (13.66x), nuclear buds (2.57x), basal (1.34x), karyorrhectic (1.26x), karyolytic (2.44x), pyknotic (1.77x) and condensed chromatin (2.08x) cells were highly significantly (p=0.000) increased in mobile phone users whereas the binucleated cells (4.03x) and repair index (8.36x) showed significant decrease (p=0.000). DNA damage and nuclear anomalies scored in BMCyt assay are indicative of genetic damage that has not been repaired and this may predispose the mobile phone users to malignancy and cytotoxicity ramifications. Therefore, despite the benefits of communication technology, measures need to be taken so that better connectivity is not at expense of health.

(E) Gapeyev AB, Lukyanova NA, Gudkov SV. Hydrogen peroxide induced by modulated electromagnetic radiation protects the cells from DNA damage. Central European Journal of Biology 9:915–921(2014) (VT, AE, GT, IX, WS)

It is believed that non-ionizing electromagnetic radiation (EMR) and low-level hydrogen peroxide (H_2O_2) may change nonspecific resistance and modify DNA damage caused by ionizing radiation. To check this assumption, the combined effects of extremely high-frequency EMR (EHF EMR) and X-rays on induction of DNA damage in mouse whole blood leukocytes were studied. The cells were exposed to X-rays with or without preliminary treatment with EHF EMR or low-level H_2O_2. With the use of enhanced chemiluminescence, it was shown for the first time that pulse-modulated EHF EMR (42.2 GHz, incident power density of 0.1 mW/cm^2, exposure duration of 20 min, modulation frequency of 1 Hz) induced H_2O_2 at a concentration of 4.6 ± 0.3 nM L^{-1} in physiological saline. With the use of an alkaline comet assay, it was found that the exposure of cells to the pulse-modulated EHF EMR, 25 min prior to treatment with X-rays at a dose of 4 Gy reduced the level of ionizing radiation-induced DNA damage. Continuous EHF

EMR was inefficient. In turn, it was shown that low-level H_2O_2 (30–500 nM L^{-1}) protected the cells against X-irradiation. Thus, the mechanisms of radiation protective effect of EHF EMR are connected with the induction of the adaptive response by nanomolar concentrations of reactive oxygen species formed by pulse-modulated EHF EMR.

(E) Garaj-Vrhovac V, Horvat D, Koren Z, The effect of microwave radiation on the cell genome. Mutat Res 243(2):87-93, 1990.(VT, AE, GT)

Cultured V79 Chinese hamster cells were exposed to continuous radiation, frequency 7.7 GHz, power density 30 mW/cm2 for 15, 30, and 60 min. The parameters investigated were the incorporation of [3H]thymidine and the frequency of chromosome aberrations. Data obtained by 2 methods (the incorporation of [3H]thymidine into DNA and autoradiography) showed that the inhibition of [3H]thymidine incorporation took place by complete prevention of DNA from entering into the S phase. The normal rate of incorporation of [3H]thymidine was recovered within 1 generation cycle of V79 cells. Mutagenic tests performed concurrently showed that even DNA macromolecules were involved in the process. In comparison with the control samples there was a higher frequency of specific chromosome lesions in cells that had been irradiated. Results discussed in this study suggest that microwave radiation causes changes in the synthesis as well as in the structure of DNA molecules.

(E) Garaj-Vrhovac V, Horvat D, Koren Z, The relationship between colony-forming ability, chromosome aberrations and incidence of micronuclei in V79 Chinese hamster cells exposed to microwave radiation. Mutat Res 263(3):143-149, 1991. (VT, AE, GT)

Cultured V79 Chinese hamster fibroblast cells were exposed to continuous radiation, frequency 7.7 GHz, power density 0.5 mW/cm2 for 15, 30 and 60 min. The effect of microwave radiation on cell survival and on the incidence and frequency of micronuclei and structural chromosome aberrations was investigated. The decrease in the number of irradiated V79 cell colonies was related to the power density applied and to the time of exposure. In comparison with the control samples there was a significantly higher frequency of specific chromosome aberrations such as dicentric and ring chromosomes in irradiated cells. The presence of micronuclei in irradiated cells confirmed the changes that had occurred in chromosome structure. These results suggest that microwave radiation can induce damage in the structure of chromosomal DNA.

(E) Garaj-Vrhovac V, Fucic A, Horvat D, The correlation between the frequency of micronuclei and specific chromosome aberrations in human lymphocytes exposed to microwave radiation in vitro. Mutat Res 281(3):181-186, 1992. (VT, AE, GT)

Human whole-blood samples were exposed to continuous microwave radiation, frequency 7.7 GHz, power density 0.5, 10 and 30 mW/cm2 for 10, 30 and 60 min. A correlation between specific chromosomal aberrations and the incidence of micronuclei after in vitro exposure was observed. In all experimental conditions, the frequency of all types of chromosomal aberrations was significantly higher than in the control samples.

In the irradiated samples the presence of dicentric and ring chromosomes was established. The incidence of micronuclei was also higher in the exposed samples. The results of the structural chromosome aberration test and of the micronucleus test were comparatively analyzed. The values obtained showed a positive correlation between micronuclei and specific chromosomal aberrations (acentric fragments and dicentric chromosomes). The results of the study indicate that microwave radiation causes changes in the genome of somatic human cells and that the applied tests are equally sensitive for the detection of the genotoxicity of microwaves.

(E) Garaj-Vrhovac V, Fucic A. The rate of elimination of chromosomal aberrations after accidental exposure to microwave radiation. Bioelectrochem. Bioenerg. 30: 319-325, 1993. (HU, AE, GT)

Analysis of structural chromosome aberrations was performed in a group of radar station personnel who were engaged in repairing radar devices a couple of days earlier. Test results showed a major decline from the values recorded by regular mutagenic monitoring in terms of a significantly increased number of chromosome breaks, acentric fragments, dicentric and polycentric chromosomes with accompanying fragments, ring chromosomes and chromatid interchange. Multiply repeated mutagenic testing demonstrated for all subjects a fall in the total number of chromosome aberrations as a function of time. During a 30-week-long follow-up study a decrease in the total number of chromosome aberrations was observed. In the same period the presence of unstable aberrations such as dicentrics and ring chromosomes persisted, together with a relatively unchanged incidence of stable aberrations.

(E) Garaj-Vrhovac, V, Micronucleus assay and lymphocyte mitotic activity in risk assessment of occupational exposure to microwave radiation. Chemosphere 39(13):2301-2312, 1999. (HU, LE, GT)

The effects of radiofrequency electromagnetic radiation (RFR) on the cell kinetics and genome damages in peripheral blood lymphocytes were determined in lymphocytes of 12 subjects occupationally exposed to microwave radiation. Results showed an increase in frequency of micronuclei (MN) as well as disturbances in the distribution of cells over the first, second and third mitotic division in exposed subjects compared to controls. According to previous reports micronucleus assay can serve as a suitable indicator for the assessment of exposure to genotoxic agents (such as RFR) and the analysis of mitotic activity as an additional parameter for the efficient biomonitoring.

(E) Garaj-Vrhovac V, Orescanin V. Assessment of DNA sensitivity in peripheral blood leukocytes after occupational exposure to microwave radiation: the alkaline comet assay and chromatid breakage assay. Cell Biol Toxicol. 25(1):33-43, 2009a. (HU, LE, GT)

DNA sensitivity in peripheral blood leukocytes of radar-facility workers daily exposed to microwave radiation and an unexposed control subjects was investigated. The study was carried out on clinically healthy male workers employed on radar equipment and antenna system service within a microwave field of 10 muW/cm(2)-20 mW/cm(2) with frequency range of 1,250-1,350 MHz. The control group consisted of subjects of similar age. The evaluation of DNA damage

and sensitivity was performed using alkaline comet assay and chromatid breakage assay (bleomycin-sensitivity assay). The levels of DNA damage in exposed subjects determined by alkaline comet assay were increased compared to control group and showed inter-individual variations. After short exposure of cultured lymphocytes to bleomycin cells of subjects occupationally exposed to microwave (MW) radiation responded with high numbers of chromatid breaks. Almost three times higher number of bleomycin-induced chromatid breaks in cultured peripheral blood lymphocytes were determined in comparison with control group. The difference in break per cell (b/c) values recorded between smokers and non-smokers was statistically significant in the exposed group. Regression analyses showed significant positive correlation between the results obtained with two different methods. Considering the correlation coefficients, the number of metaphase with breaks was a better predictor of the comet assay parameters compared to b/c ratio. The best correlation was found between tail moment and number of chromatid with breaks. Our results indicate that MW radiation represents a potential DNA-damaging hazard using the alkaline comet assay and chromatid breakage assay as sensitive biomarkers of individual cancer susceptibility.

(E) Garaj-Vrhovac V, Gajski G, Trosić I, Pavicić I. Evaluation of basal DNA damage and oxidative stress in Wistar rat leukocytes after exposure to microwave radiation. Toxicology. 259(3):107-112, 2009b. (VO, LE, GT, OX)

The aim of this study was to assess whether microwave-induced DNA damage is basal or it is also generated through reactive oxygen species (ROS) formation. After having irradiated Wistar rats with 915MHz microwave radiation, we assessed different DNA alterations in peripheral leukocytes using standard and formamidopyrimidine DNA-glycosylase (Fpg)-modified comet assay. The first is a sensitive tool for detecting primary DNA damage, and the second is much more specific for detecting oxidative damage. The animals were irradiated for 1h a day for 2 weeks at a field power density of 2.4W/m(2), and the whole-body average specific absorption rate (SAR) of 0.6W/kg. Both the standard and the Fpg-modified comet assay detected increased DNA damage in blood leukocytes of the exposed rats. The significant increase in Fpg-detected DNA damage in the exposed rats suggests that oxidative stress is likely to be responsible. DNA damage detected by the standard comet assay indicates that some other mechanisms may also be involved. In addition, both methods served proved sensitive enough to measure basal and oxidative DNA damage after long-term exposure to 915MHz microwave radiation in vivo.

(E) Garaj-Vrhovac V, Gajski G, Pažanin S, Sarolić A, Domijan AM, Flajs D, Peraica M. Assessment of cytogenetic damage and oxidative stress in personnel occupationally exposed to the pulsed microwave radiation of marine radar equipment. Int J Hyg Environ Health. 4(1):59-65, 2011. (HU, LE, GT, LI, OX)

Due to increased usage of microwave radiation, there are concerns of its adverse effect in today's society. Keeping this in view, study was aimed at workers occupationally exposed to pulsed microwave radiation, originating from marine radars. Electromagnetic field strength was measured at assigned marine radar frequencies (3 GHz, 5.5 GHz and 9.4 GHz) and corresponding specific absorption rate values were determined. Parameters of the comet assay and micronucleus test were studied both in the exposed workers and in corresponding unexposed

subjects. Differences between mean tail intensity (0.67 vs. 1.22) and moment (0.08 vs. 0.16) as comet assay parameters and micronucleus test parameters (micronuclei, nucleoplasmic bridges and nuclear buds) were statistically significant between the two examined groups, suggesting that cytogenetic alterations occurred after microwave exposure. Concentrations of glutathione and malondialdehyde were measured spectrophotometrically and using high performance liquid chromatography. The glutathione concentration in exposed group was significantly lower than in controls (1.24 vs. 0.53) whereas the concentration of malondialdehyde was significantly higher (1.74 vs. 3.17), indicating oxidative stress. Results suggests that pulsed microwaves from working environment can be the cause of genetic and cell alterations and that oxidative stress can be one of the possible mechanisms of DNA and cell damage.

(NE) Garson OM, McRobert TL, Campbell LJ, Hocking BA, Gordon I. A chromosomal study of workers with long-term exposure to radio-frequency radiation. Med J Aust 155(5):289-292, 1991. (HU, LE, GT)

OBJECTIVE: To examine whether an increased level of chromosome damage occurs in the stimulated lymphocytes of radio-linemen after long-term but intermittent exposure to radio-frequency radiation (RFR) during the course of their work. DESIGN AND PARTICIPANTS: Chromosome studies were performed on blood samples from 38 radio-linemen matched by age with 38 controls, all of whom were employed by Telecom Australia. The radio-linemen had all worked with RFR in the range 400 kHz-20 GHz with exposures at or below the Australian occupational limits, and the controls were members of the clerical staff who had no exposure to RFR. Two hundred metaphases from each subject were studied and chromosome damage was scored by an observer who was blind to the status of the subjects. RESULTS: The ratio of the rate of aberrant cells in the radio-linemen group to that in the control group was 1.0 (95% confidence interval, 0.8-1.3). There were no statistically significant differences in the types of aberrations that were scored. CONCLUSION: Exposure to RFR at or below the described limits did not appear to cause any increase in chromosomal damage in circulating lymphocytes.

(E) Ghandehari M, Sadri D, Farhadi S. Micronucleus Assay in Cell Phone Users: Importance of Oral Mucosa Screening. Int J Prev Med. 12:125, 2021. (HU, LE, GT)

Background: One of the concerns of cell phone users is prolonged exposure to harmful and potentially carcinogenic waves. This study was aimed to investigate the correlation between amount of cell phone use and related factors with percentage of micronucleus containing cells. Methods: This descriptive study was conducted on selected patients referring to Islamic Azad University Faculty of Dentistry using cell phones regarding related inclusion and exclusion criteria. Papanicolaou staining method was approached for mucosal smears of samples and frequency of micronucleus containing cells and also, frequency of micronucleus in each cell were recorded for each sample; then, correlation of these findings with amount of daily cell phone usage was statistically analyzed using the calculation of Pearson correlation coefficient and preparation of regression analysis (backward) with significant level of lower than 0.05. Results: Of 100 samples, the frequency of micronucleus containing cells was 2.94% ± 1.89% and the frequency of micronucleus in each cell was 1.02% ± 1.68%. The amount of cell phone usage

was significantly correlated with the frequency of micronucleus containing cells (r = 0.70, P = 0.0001) and also with the frequency of micronucleus in each cell (r = 0.57, P = 0.0001). Also, age and sex were not significantly correlated with the frequency of micronucleus containing cells (P = 0.47 and 0.32) and also with the frequency of micronucleus in each cell, respectively (P = 0.16 and 0.27). Conclusions: The present study showed that the increased amount of cell phone usage had a strong and significant correlation with the higher frequency of the micronucleus containing cells and the higher frequency of micronucleus in each cell in the buccal mucosa. Also, the related factors as age and sex were not significantly correlated with the frequency of micronucleus containing buccal mucosa cells.

(E) Ghatei N, Nabavi AS, Toosi MHB, Azimian H, Homayoun M, Targhi RG, Haghir H. Evaluation of *bax*, *bcl-2*, *p21* and *p53* genes expression variations on cerebellum of BALB/c mice before and after birth under mobile phone radiation exposure. Iran J Basic Med Sci. 20(9):1037-1043, 2017. (VO, LE, GE, DE)

Objectives: The increasing rate of over using cell phones has been considerable in youths and pregnant women. We examined the effect of mobile phones radiation on genes expression variation on cerebellum of BALB/c mice before and after of the birth. **Materials and methods:** In this study, a mobile phone jammer, which is an instrument to prevent receiving signals between cellular phones and base transceiver stations (two frequencies 900 and 1800 MHz) for exposure was used and twelve pregnant mice (BALB/c) divided into two groups (n=6), first group irradiated in pregnancy period (19th day), the second group did not irradiate in pregnancy period. After childbirth, offspring were classified into four groups (n=4): Group1: control, Group 2: B1 (Irradiated after birth), Group 3: B2 (Irradiated in pregnancy period and after birth), Group 4: B3 (Irradiated in pregnancy period). When maturity was completed (8-10 weeks old), mice were dissected and cerebellum was isolated. The expression level of *bax*, bcl-2, p21 and *p53* genes examined by real-time reverse transcription polymerase chain reaction (Real-Time RT- PCR). **Results:** The data showed that mobile phone radio waves were ineffective on the expression level of bcl-2 and p53 genes) $P>0.05$(. Also gene expression level of *bax* decreased and gene expression level of *p21* increased comparing to the control group ($P<0.05$). **Conclusion:** From the obtained data it could be concluded that the mobile phone radiations did not induce apoptosis in cells of the cerebellum and the injured cells can be repaired by cell cycle arrest.

(NE) Gläser K, Rohland M, Kleine-Ostmann T, Schrader T, Stopper H, Hintzsche H. Effect of Radiofrequency Radiation on Human Hematopoietic Stem Cells. Radiat Res. 186(5):455-465, 2016. (VT, AE, GT)

Exposure to electromagnetic fields in the radiofrequency range is ubiquitous, mainly due to the worldwide use of mobile communication devices. With improving technologies and affordability, the number of cell phone subscriptions continues to increase. Therefore, the potential effect on biological systems at low-intensity radiation levels is of great interest. While a number of studies have been performed to investigate this issue, there has been no consensus reached based on the results. The goal of this study was to elucidate the extent to which cells of the hematopoietic system, particularly human hematopoietic stem cells (HSC), were affected by

mobile phone radiation. We irradiated HSC and HL-60 cells at frequencies used in the major technologies, GSM (900 MHz), UMTS (1,950 MHz) and LTE (2,535 MHz) for a short period (4 h) and a long period (20 h/66 h), and with five different intensities ranging from 0 to 4 W/kg specific absorption rate (SAR). Studied end points included apoptosis, oxidative stress, cell cycle, DNA damage and DNA repair. In all but one of these end points, we detected no clear effect of mobile phone radiation; the only alteration was found when quantifying DNA damage. Exposure of HSC to the GSM modulation for 4 h caused a small but statistically significant decrease in DNA damage compared to sham exposure. To our knowledge, this is the first published study in which putative effects (e.g., genotoxicity or influence on apoptosis rate) of radiofrequency radiation were investigated in HSC. Radiofrequency electromagnetic fields did not affect cells of the hematopoietic system, in particular HSC, under the given experimental conditions.

(E) Gökçek-Saraç C, Akçay G, Karakurt S, Ateş K, Özen S, Derin N. Possible effects of different doses of 2.1 GHz electromagnetic radiation on learning, and hippocampal levels of cholinergic biomarkers in Wistar rats. Electromagn Biol Med 40:179-190, 2021. (VO, LE, GE)

The present study evaluated whether short-term exposure to different doses of 2.1 GHz radiofrequency electromagnetic radiation (RF-EMR) has different effects on rats' behaviour and hippocampal levels of central cholinergic biomarkers. Animals were divided into three equal groups namely; group 1 was sham-exposed group, group 2-3 were exposed to 45 V/m and 65 V/m doses of 2.1 GHz frequency for 1 week respectively. Numerical dosimetry simulations were carried out. Object location and Y-maze were used as behavioural tasks. The protein and mRNA expression levels of AChE, ChAT, and VAChT, in the hippocampus were tested using Western Blotting and Real-Time PCR. The impairment performance of rats subjected to 65 V/m dose of 2.1 GHz RF-EMR in both object location and Y-maze tasks was observed. The hippocampal levels of AChE, ChAT, and VAChT, were significantly lower in rats exposed to 65 V/m dose of 2.1 GHz RF-EMR than others. The stronger effect of "65 V/m" dose on both rat's hippocampal-dependent behavioural performances and hippocampal levels of cholinergic biomarkers may be due to the stronger effect of "65 V/m" dose where rats' snouts were located at the nearest distance from the monopole antenna. Furthermore, the simulated SAR values were high for 65 V/m electric-field strengths. For the first time, we report the potential dose-dependent effects of short-term exposure to 2.1 GHz radiation on rat's behavioural performances as well as hippocampal levels of cholinergic biomarkers. Further studies are needed to understand the mechanisms by which RF-EMR influences the function of the central cholinergic system in the brain.

(NE) Görlitz B-D, Müller M, Ebert S, Hecker H, Kuster N, Dasenbrock C. Effects of 1-week and 6-week exposure to GSM/DCS radiofrequency radiation on micronucleus formation in B6C3F1 mice. Radiat Res 164(4 Pt 1):431-439, 2005. (VO, LE, GT)

The aim of this study was to examine the possible induction of micronuclei in erythrocytes of the peripheral blood and bone marrow and in keratinocytes and spleen lymphocytes of mice exposed to radiofrequency (RF) radiation for 2 h per day over periods of 1 and 6 weeks, respectively. The

applied signal simulated the exposure from GSM900 and DCS1800 handsets, including the low-frequency amplitude-modulation components as they occur during speaking (GSM Basic), listening (DTX) and moving within the environment (handovers, power control). The carrier frequency was set to the center of the system's uplink band, i.e., 902 MHz for GSM and 1747 MHz for DCS. Uniform whole-body exposure was achieved by restraining the mice in tubes at fixed positions in the exposure setup. Mice were exposed to slot-averaged whole-body SARs of 33.2, 11.0, 3.7 and 0 mW/g during the 1-week study and 24.9, 8.3, 2.8 and 0 mW/g during the 6-week study. Exposure levels for the 1- and 6-week studies were determined in a pretest to confirm that no thermal effect was present that could influence the genotoxic end points. During both experiments and for both frequencies, no clinical abnormalities were detected in the animals. Cells of the bone marrow from the femur (1-week study), erythrocytes of the peripheral blood (6-week study), keratinocytes from the tail root, and lymphocytes from the spleen (both studies) were isolated on slides and stained for micronucleus analysis. Two thousand cells per animal were scored in erythrocyte and keratinocyte samples. In spleen lymphocytes, 1000 binucleated lymphocytes were scored for each animal. The RF-field exposure had no influence on the formation of red blood cells. After 1 week of exposure, the ratio of polychromatic to normochromatic erythrocytes was unchanged in the treated groups compared to the sham-exposed groups. Furthermore, the RF-field exposure of mice did not induce an increase in the number of micronuclei in erythrocytes of the bone marrow or peripheral blood, in keratinocytes, or in spleen lymphocytes compared to the sham-treated control.

(E) Gorpinchenko I, Nikitin O, Banyra O, Shulyak A. The influence of direct mobile phone radiation on sperm quality. Cent European J Urol. 67(1):65-71, 2014. (VT, AE, GT, RP)

INTRODUCTION: It is impossible to imagine a modern socially-active man who does not use mobile devices and/or computers with Wi-Fi function. The effect of mobile phone radiation on male fertility is the subject of recent interest and investigations. The aim of this study was to investigate the direct in vitro influence of mobile phone radiation on sperm DNA fragmentation and motility parameters in healthy subjects with normozoospermia. **MATERIAL AND METHODS:** 32 healthy men with normal semen parameters were selected for the study. Each sperm sample was divided into two equal portions (A and B). Portions A of all involved men were placed for 5 hours in a thermostat, and portions B were placed into a second thermostat for the same period of time, where a mobile phone in standby/talk mode was placed. After 5 hours of incubation the sperm samples from both thermostats were re-evaluated regarding basic motility parameters. The presence of DNA fragmentation in both A and B portions of each sample was determined each hour using a standard sperm chromatin dispersion test. **RESULTS:** The number of spermatozoa with progressive movement in the group, influenced by electromagnetic radiation, is statistically lower than the number of spermatozoa with progressive movement in the group under no effect of the mobile phone. The number of non-progressive movement spermatozoa was significantly higher in the group, which was influenced by cell phone radiation. The DNA fragmentation was also significantly higher in this group. **CONCLUSIONS:** A correlation exists between mobile phone radiation exposure, DNA-fragmentation level and decreased sperm motility.

(E)Goswami PC, Albee LD, Parsian AJ, Baty JD, Moros EG, Pickard WF, Roti Roti JL, Hunt CR. Proto-oncogene mRNA levels and activities of multiple transcription factors

in C3H 10T 1/2 murine embryonic fibroblasts exposed to 835.62 and 847.74 MHz cellular phone communication frequency radiation. Radiat Res 151(3):300-309, 1999. (VT, AE, GE)

This study was designed to determine whether two differently modulated radiofrequencies of the type generally used in cellular phone communications could elicit a general stress response in a biological system. The two modulations and frequencies studied were a frequency-modulated continuous wave (FMCW) with a carrier frequency of 835.62 MHz and a code division multiple-access (CDMA) modulation centered on 847.74 MHz. Changes in proto-oncogene expression, determined by measuring Fos, Jun, and Myc mRNA levels as well as by the DNA-binding activity of the AP1, AP2 and NF-kappaB transcription factors, were used as indicators of a general stress response. The effect of radiofrequency exposure on proto-oncogene expression was assessed (1) in exponentially growing C3H 10T 1/2 mouse embryo fibroblasts during their transition to plateau phase and (2) during transition of serum-deprived cells to the proliferation cycle after serum stimulation. Exposure of serum-deprived cells to 835.62 MHz FMCW or 847.74 MHz CDMA microwaves (at an average specific absorption rate, SAR, of 0.6 W/kg) did not significantly change the kinetics of proto-oncogene expression after serum stimulation. Similarly, these exposures did not affect either the Jun and Myc mRNA levels or the DNA-binding activity of AP1, AP2 and NF-kappaB in exponential cells during transit to plateau-phase growth. Therefore, these results suggest that the radiofrequency exposure is unlikely to elicit a general stress response in cells of this cell line under these conditions. However, statistically significant increases (approximately 2-fold, P = 0.001) in Fos mRNA levels were detected in exponential cells in transit to the plateau phase and in plateau-phase cells exposed to 835.62 MHz FMCW microwaves. For 847.74 MHz CDMA exposure, the increase was 1.4-fold (P = 0.04). This increase in Fos expression suggests that expression of specific genes could be affected by radiofrequency exposure.

(E) Gulati S, Yadav A, Kumar N, Kanupriya, Aggarwal NK, Kumar R, Gupta R. Effect of GSTM1 and GSTT1 Polymorphisms on Genetic Damage in Humans Populations Exposed to Radiation From Mobile Towers. Arch Environ Contam Toxicol. 70(3): 615-625, 2016. (HU, LE, GT)

All over the world, people have been debating about associated health risks due to radiation from mobile phones and mobile towers. The carcinogenicity of this nonionizing radiation has been the greatest health concern associated with mobile towers exposure until recently. The objective of our study was to evaluate the genetic damage caused by radiation from mobile towers and to find an association between genetic polymorphism of GSTM1 and GSTT1 genes and DNA damage. In our study, 116 persons exposed to radiation from mobile towers and 106 control subjects were genotyped for polymorphisms in the GSTM1 and GSTT1 genes by multiplex polymerase chain reaction method. DNA damage in peripheral blood lymphocytes was determined using alkaline comet assay in terms of tail moment (TM) value and micronucleus assay in buccal cells (BMN). There was a significant increase in BMN frequency and TM value in exposed subjects (3.65 ± 2.44 and 6.63 ± 2.32) compared with control subjects (1.23 ± 0.97 and 0.26 ± 0.27). However, there was no association of GSTM1 and GSTT1 polymorphisms with the level of DNA damage in both exposed and control groups.

(E) Gulati, S, Kosik, P, Durdik, M, Skorvaga, M., Jakl, L., Markova, E., Belyaev, I. Effects of different mobile phone UMTS signals on DNA, apoptosis and oxidative stress in human lymphocytes. Environ Pollut. 267:115632, 2020. (VT, AE, GT,WS)

Different scientific reports suggested a link between exposure to radiofrequency radiation (RF) from mobile communications and induction of reactive oxygen species (ROS) and DNA damage while other studies have not found such a link. However, the available studies are not directly comparable because they were performed at different parameters of exposure, including carrier frequency of RF signal, which was shown to be critical for appearance of the RF effects. For the first time, we comparatively analyzed genotoxic effects of UMTS signals at different frequency channels used by 3G mobile phones (1923, 1947.47, and 1977 MHz). Genotoxicity was examined in human lymphocytes exposed to RF for 1 h and 3 h using complimentary endpoints such as induction of ROS by imaging flow cytometry, DNA damage by alkaline comet assay, mutations in TP53 gene by RSM assay, preleukemic fusion genes (PFG) by RT-qPCR, and apoptosis by flow cytometry. No effects of RF exposure on ROS, apoptosis, PFG, and mutations in TP53 gene were revealed regardless of the UMTS frequency while inhibition of a bulk RNA expression was found. On the other hand, we found relatively small but statistically significant induction of DNA damage in dependence on UMTS frequency channel with maximal effect at 1977.0 MHz. Our data support a notion that each specific signal used in mobile communication should be tested in specially designed experiments to rule out that prolonged exposure to RF from mobile communication would induce genotoxic effects and affect the health of the human population

(E) Guler G, Tomruk A, Ozgur E, Seyhan N. The effect of radiofrequency radiation on DNA and lipid damage in non-pregnant and pregnant rabbits and their newborns. Gen Physiol Biophys 29:59-66, 2010. (VO, LE, LI, GT, OX, DE)

The concerns of people on possible adverse health effects of radiofrequency radiation (RFR) generated from mobile phones as well as their supporting transmitters (base stations) have increased markedly. RFR effect on oversensitive people, such as pregnant women and their developing fetuses, and older people is another source of concern that should be considered. In this study, oxidative DNA damage and lipid peroxidation levels in the brain tissue of pregnant and non-pregnant New Zealand White rabbits and their newborns exposed to RFR were investigated. Thirteen-month-old rabbits were studied in four groups as non-pregnant-control, non-pregnant-RFR exposed, pregnant-control and pregnant-RFR exposed. They were exposed to RFR (1800 MHz GSM; 14 V/m as reference level) for 15 min/day during 7 days. Malondialdehyde (MDA) and 8-hydroxy-2'-deoxyguanosine (8-OHdG) levels were analyzed. MDA and 8-OHdG levels of non-pregnant and pregnant-RFR exposed animals significantly increased with respect to controls ($p < 0.001$, Mann-Whitney test). No difference was found in the newborns ($p > 0.05$, Mann-Whitney). There exist very few experimental studies on the effects of RFR during pregnancy. It would be beneficial to increase the number of these studies in order to establish international standards for the protection of pregnant women from RFR.

(E) Güler G, Tomruk A, Ozgur E, Sahin D, Sepici A, Altan N, Seyhan N. The effect of radiofrequency radiation on DNA and lipid damage in female and male infant rabbits. Int J Radiat Biol. 88(4):367-373, 2012. (VO, LE, GT, OX, DE)

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.

(E) Gunes M, Ates K, Yalcin B, Akkurt S, Ozen S, Kaya B. An Evaluation of the Genotoxic Effects of Electromagnetic Radiation at 900 MHz, 1800 MHz, and 2100 MHz Frequencies with a SMART Assay in *Drosophila melanogaster.* Electromagn Biol Med 40(2):254-263, 2021. (VO, CE, GT)

With the development of today's technology, the electromagnetic radiation spread by mobile phones and base stations is also rapidly increasing, and this causes serious concerns about the environment and human health. The *Drosophila* model organism is widely used in genetic toxicology studies because its genome is highly similar to the genes identified in human diseases. In this study, the genotoxic effects of radiofrequency electromagnetic radiation were evaluated by the wing Somatic Mutation and Recombination Test (SMART) in *Drosophila melanogaster* at 900 MHz, 1800 MHz, and 2100 MHz. The SMART method is based on the observation of genetic changes occurring in the trichomes of the *Drosophila* wings appearing as mutant clones under the microscope. Throughout the study, total clone parameters were evaluated by exposing the *Drosophila* larvae to electromagnetic fields for two, four, and six hours per day for two days. As a result of the study, it was observed that the number of mutant clones was statistically increased according to the negative control group in all applications except for the six-hour application at 1800 MHz.

(NE) Gurbuz N, Sirav B, Yuvaci HU, Turhan N, Coskun ZK, Seyhan N. Is there any possible genotoxic effect in exfoliated bladder cells of rat under the exposure of 1800 MHz

GSM-like modulated radio frequency radiation (RFR)? Electromagn Biol Med. 29(3):98-104, 2010. (VO, LE, GT)

People are exposed to many carcinogenic and mutagenic chemicals in their everyday lives. These include antineoplastic drugs, Polycyclic aromatic hydrocarbons (PAH)s, aromatic amines, nitrosamines, metals, and electromagnetic radiation. Based on the state of knowledge acquired during the last 50 years of research on possible biological effects of electromagnetic fields (EMF), the majority of the scientific community is convinced that exposure to EMF below the existing security limits does not cause a risk to the health of the general public. However, this position is questioned by others, who are of the opinion that the available research data are contradictory or inconsistent and, therefore, unreliable. In this study, we aimed to investigate if there is any effect of 1800 MHz GSM modulated radio frequency radiation (RFR) on the number of micronucleus in exfoliated bladder cells of rat which will be informative about the genotoxic damage. Exposure period was 20 min/day, 5 days/week during a month. Six female Wistar rats were used for two groups: Group I (n=6): controls; Group II (n=6): 1.8 GHz exposed animals. 1800 MHz RFR did not showed a significant MN frequencies in rat bladder cells when compared with the control group ($p>0.05$). 1800 MHz RFR-exposed animals did not produce any genotoxic effect when compared with the control group ($p>0.05$). Kinetic studies are important for any biomarker, especially those in which tissue differentiation and maturation processes will heavily influence the time between induction of damage and collection of damaged cells for micronucleus analysis.

(NE) Gurbuz N, Sirav B, Colbay M, Yetkin I, Seyhan N. No genotoxic effect in exfoliated bladder cells of rat under the exposure of 1800 and 2100-MHz radio frequency radiation. Electromagn Biol Med. 33(4):296-301, 2014. (VO, LE, GT)

In this study, we aimed to investigate the effects of 1800 and 2100 MHz Radio Frequency (RF) radiation on the number of micronucleus (MN) in exfoliated bladder cells of rat which shows the genotoxic damage. Exposure period was 30 min/day, 6 days/week for a month and two months exposure periods. Thirty male wistar albino rats were used for five groups: Group I ($n = 6$): 1800 MHz RF exposed animals for one month, Group II ($n = 6$): 2100 MHz RF exposed animals for one month, Group III ($n = 6$): 2100 MHz RF exposed for two months, Group IV ($n = 6$): control group for one month, Group V ($n = 6$): control group for two months. Rats of the control groups were housed in their home cages during the entire experimental period without subjecting to any experimental manipulation. 1800 and 2100 MHz RF exposures did not result in any significant MN frequencies in rat bladder cells with respect to the control groups ($p > 0.05$). There was no statistically significant difference between 2100 MHz RF exposed groups, either. Further studies are needed to demonstrate if there is any genotoxic effect, micronucleus formation in other tissues of rats.

(NE) Gurbuz N, Sirav B, Kuzay D, Ozer C, Seyhan N. Does radio frequency radiation induce micronuclei frequency in exfoliated bladder cells of diabetic rats? Endocr Regul 49(3):126-130, 2015. (VT, LE, GT)

Objective: For many years there has been a discussion among both experts and the general public regarding the effects of radio frequency (RF) radiation on the human organism. The purpose of the present study was to evaluate the relationship of micronucleui (MN) frequency and RF radiation in exfoliated bladder cells of non-diabetic and diabetic rats. **Methods:** Three groups were used in the experiment: Group I (n=6): diabetic group without RF exposure; Group II (n=6): diabetic group exposed 2100 MHz RF radiation and Group III (n=6): control animals (non-diabetic group, no RF exposure). RF exposure in the experiment resulted in a whole body average SAR of 0.24 W/kg with an ERMS field of 17.5 V/m in non-thermal levels. **Result:** Results showed that there was no statistically important differences between non-RF exposed diabetes group and control group; Group I and Group III ($p>0.05$). There was no statistically important differences between diabetes group and diabetes+RF exposed group (Group I and Group II) ($p>0.05$). RF exposure did not result in increased MN frequencies in exfoliated bladder cells of diabetic rats with respect to control animals (Group II and Group III), either and this result found no statistically important ($p>0.05$). **Conclusions:** This study suggested no possible genotoxic effects of RF radiation among human beings especially with chronic disorders, such as diabetes.

(NE) Gurisik E, Warton K, Martin DK, Valenzuela SM. An in vitro study of the effects of exposure to a GSM signal in two human cell lines: monocytic U937 and neuroblastoma SK-N-SH. Cell Biol Int 30(10):793-759, 2006. (VT, AE, GE)

The use of mobile phones is increasing, which also increases the population's exposure to global system of mobile communications (GSM) signals. Questions of safety and possible biological effects are of concern and to date, remain largely unanswered. In order to examine possible biological effects of a GSM-like signal at a cellular level, we exposed two human cell lines (one of neuronal (SK-N-SH) and the other of monocytoid (U937) origin) to a 900 MHz RF signal, pulsed at 217 Hz, producing a specific absorption rate (SAR) of 0.2 W/kg. Putative effects were assessed by comparing radiofrequency-exposed cells to sham-exposed cells using a variety of assay techniques. For the cell line SK-N-SH, effects were specifically assessed by gene microarray, followed by real-time PCR of the genes of interest, Western blot analysis was used to measure heat shock protein levels, and flow cytometry to measure cell cycle distributions and apoptosis. Effects of radiofrequency on the cell line U937 were assessed by cell viability and cell cycle analysis. From our study of these two cell lines, we found no significant difference between sham-exposed versus radiofrequency-exposed cells in any of the assays or conditions examined.

(E) Gürler HS, Bilgici B, Akar AK, Tomak L, Bedir A. Increased DNA oxidation (8-OHdG) and protein oxidation (AOPP) by Low level electromagnetic field (2.45 GHz) in rat brain and protective effect of garlic. Int J Radiat Biol. 90(10):892-896, 2014. (VO, LE, LI, GT, OX, IX)

Purpose: To investigate the oxidative damage and protective effect of garlic on rats exposed to low level of electromagnetic fields (EMF) at 2.45 GHz Microwave radiation (MWR). Methods: Thirty six Wistar rats were divided into three groups. Group I was the control group and not exposed to EMF. Group II and III were exposed to low level EMF (3.68±0.36 V/m) at 2.45 GHz

MWR for 1 hour/day for 30 consecutive days. Daily 500 mg/kg garlic was given to Group III during the study period. At the end of the study, thiobarbituric acid reactive substances (TBARS), advanced oxidation protein products (AOPP) and 8-hydroxydeoxyguanosine (8-OHdG) levels were investigated in brain tissue and blood samples. Results: Exposure to low level of EMF increased 8-OHdG level in both plasma and brain tissue whereas it increased AOPP level only in plasma. Garlic prevented the increase of 8-OHdG level in brain tissue and plasma AOPP levels. Conclusions: It may be concluded that low level EMF at 2.45 GHz MWR increases the DNA damage in both brain tissues and plasma of the rats whereas it increases protein oxidation only in plasma. It may also be argued that the use of garlic decreases these effects.

(E) Gustavino B, Carboni G, Petrillo R, Paoluzzi G, Santovetti E, Rizzoni M. Exposure to 915 MHz radiation induces micronuclei in Vicia faba root tips. Mutagenesis. 31(2):187-192, 2016. (VO. AE, GT)

The increasing use of mobile phones and wireless networks raised a great debate about the real carcinogenic potential of radiofrequency-electromagnetic field (RF-EMF) exposure associated with these devices. Conflicting results are reported by the great majority of in vivo and in vitro studies on the capability of RF-EMF exposure to induce DNA damage and mutations in mammalian systems. Aimed at understanding whether less ambiguous responses to RF-EMF exposure might be evidenced in plant systems with respect to mammalian ones, in the present work the mutagenic effect of RF-EMF has been studied through the micronucleus (MN) test in secondary roots of Vicia faba seedlings exposed to mobile phone transmission in controlled conditions, inside a transverse electro magnetic (TEM) cell. Exposure of roots was carried out for 72h using a continuous wave (CW) of 915 MHz radiation at three values of equivalent plane wave power densities (23, 35 and 46W/m(2)). The specific absorption rate (SAR) was measured with a calorimetric method and the corresponding values were found to fall in the range of 0.4-1.5W/kg. Results of three independent experiments show the induction of a significant increase of MN frequency after exposure, ranging from a 2.3-fold increase above the sham value, at the lowest SAR level, up to a 7-fold increase at the highest SAR. These findings are in agreement with the limited number of data on cytogenetic effects detected in other plant systems exposed to mobile phone RF-EMF frequencies and clearly show the capability of radiofrequency exposure to induce DNA damage in this eukaryotic cell system.

(E) Habauzit D, Le Quément C, Zhadobov M, Martin C, Aubry M, Sauleau R, Le Dréan Y. Transcriptome analysis reveals the contribution of thermal and the specific effects in cellular response to millimeter wave exposure. PLoS One. 9(10):e109435, 2014. (VT, AE, GE)

Radiofrequency radiations constitute a new form of environmental pollution. Among them, millimeter waves (MMW) will be widely used in the near future for high speed communication systems. This study aimed therefore to evaluate the biocompatibility of MMW at 60 GHz. For this purpose, we used a whole gene expression approach to assess the effect of acute 60 GHz exposure on primary cultures of human keratinocytes. Controls were performed to dissociate the electromagnetic from the thermal effect of MMW. Microarray data were validated by RT-PCR, in order to ensure the reproducibility of the results. MMW exposure at 20 mW/cm2,

corresponding to the maximum incident power density authorized for public use (local exposure averaged over 1 cm2), led to an increase of temperature and to a strong modification of keratinocyte gene expression (665 genes differentially expressed). Nevertheless, when temperature is artificially maintained constant, no modification in gene expression was observed after MMW exposure. However, a heat shock control did not mimic exactly the MMW effect, suggesting a slight but specific electromagnetic effect under hyperthermia conditions (34 genes differentially expressed). By RT-PCR, we analyzed the time course of the transcriptomic response and 7 genes have been validated as differentially expressed: ADAMTS6, NOG, IL7R, FADD, JUNB, SNAI2 and HIST1H1A. Our data evidenced a specific electromagnetic effect of MMW, which is associated to the cellular response to hyperthermia. This study raises the question of co-exposures associating radiofrequencies and other environmental sources of cellular stress.

(NE) Habauzit D, Nugue G, Bourbon F, Martin C, Del Vecchio F, Maunoir-Regimbal S, Poyot T, Valente M, Jaoui R, Crouzier D, Le Dréan Y, Debouzy JC. Evaluation of the Effect of Chronic 94 GHz Exposure on Gene Expression in the Skin of Hairless Rats *In Vivo*. Radiat Res. 193(4):351-358, 2020. (VO. LE, GE)

Millimeter waves (MMW) are broadband frequencies that have recently been used in several applications in wireless communications, medical devices and nonlethal weapons [i.e., the nonlethal weapon, Active Denial Systems, (ADS) operating at 94-95 GHz, CW]. However, little information is available on their potential effects on humans. These radio-frequencies are absorbed and stopped by the first layer of the skin. In this study, we evaluated the effects of 94 GHz on the gene expression of skin cells. Two rat populations consisting of 17 young animals and 14 adults were subjected to chronic long-term 94 GHz MMW exposure. Each group of animals was divided into exposed and sham subgroups. The two independent exposure experiments were conducted for 5 months with rats exposed 3 h per day for 3 days per week to an incident power density of 10 mW/cm^2, which corresponded to twice the ICNIRP limit of occupational exposure for humans. At the end of the experiment, skin explants were collected and RNA was extracted. Then, the modifications to the whole gene expression profile were analyzed with a gene expression microarray. Without modification of the animal's temperature, long-term chronic 94 GHz-MMW exposure did not significantly modify the gene expression of the skin on either the young or adult rats.

(E) Haider T, Knasmueller S, Kundi M, Haider M. Clastogenic effects of radiofrequency Radiations on chromosomes of Tradescantia. Mutat Res 324(1-2):65-68, 1994. (VO, AE. GT)

The clastogenicity of electromagnetic fields (EMF) has so far been studied only under laboratory conditions. We used the Tradescantia-micronucleus (Trad-MCN) bioassay in an in situ experiment to find out whether short-wave electromagnetic fields used for broadcasting (10-21 MHz) may show genotoxic effects. Plant cuttings bearing young flower buds were exposed (30 h) on both sides of a slewable curtain antenna (300/500 kW, 40-170 V/m) and 15 m (90 V/m) and 30 m (70 V/m) distant from a vertical cage antenna (100 kW) as well as at the neighbors living near the broadcasting station (200 m, 1-3 V/m). The exposure at both sides of the slewable curtain antenna was performed simultaneously within cages, one of the Faraday type shielding

the field and one non-shielding mesh cage. Laboratory controls were maintained for comparison. Higher MCN frequencies than in laboratory controls were found for all exposure sites in the immediate vicinity of the antennae, where the exposure standards of the electric field strength of the International Radiation Protection Association (IRPA) were exceeded. The results at all exposure sites except one were statistically significant. Since the parallel exposure in a non-shielding and a shielding cage also revealed significant differences in MCN frequencies (the latter showing no significant differences from laboratory controls), the clastogenic effects are clearly attributable to the short-wave radiation from the antennae.

(E) Hamann W, Abou-Sherif S, Thompson S, Hall S. Pulsed radiofrequency applied to dorsal root ganglia causes a selective increase in ATF3 in small neurons. Eur J Pain. 10(2):171-176, 2006. (VO, AE, GE)

BACKGROUND: This is a "proof of concept study" to test the hypothesis that pulsed radiofrequency, PRF, produces cell stress at the primary afferent level without signs of overt thermal damage. We assumed that cell stress would result in impairment of normal function, and used the expression of activating transcription factor 3, ATF3, as an indicator of cellular "stress". METHODS: PRF (20ms of 500-kHz RF pulses, delivered at a rate of 2Hz; maximum temperature 42 degrees C) was delivered either to the sciatic nerve of adult rats in mid thigh, or to the L4 anterior primary ramus just distal to the intervertebral foramen. Controls were sham-operated or L4 axotomised. All tissues were examined 14 days after surgery. The percentage of CGRP- or ATF3-positive DRG neuronal somata was calculated using image analysis software (SigmaScan Pro 4). RESULTS: ATF3 expression was upregulated in L4 DRG neuronal cell bodies, irrespective of their size, after axotomy. It was also upregulated significantly ($p<0.002$) and selectively, in small and medium calibre L4 DRG neurons, when PRF was applied close to the DRG just distal to the intervertebral foramen. PRF did not produce any obvious cellular changes in the nerve or L4 DRG neurons when applied to the sciatic nerve in mid-thigh. CONCLUSION: PRF has a biological effect, unlikely to be related to overt thermal damage. It appears to be selective in that it targets the group of neurons whose axons are the small diameter C and A delta nociceptive fibres.

(E) Hancı H, Odacı E, Kaya H, Aliyazıcıoğlu Y, Turan İ, Demir S, Çolakoğlu S. The effect of prenatal exposure to 900-MHz electromagnetic field on the 21-old-day rat testicle. Reprod Toxicol. 42:203-209, 2013. (VO, LE, GT, OX, RP)

The aim of this study was to investigate the effect of exposure to a 900-MHz electromagnetic field (EMF) in the prenatal term on the 21-old-day rat testicle. Pregnant rats were divided into control (CG) and EMF (EMFG) groups. EMFG was exposed to 900-MHz EMF during days 13-21 of pregnancy. Newborn CG rats were obtained from the CG and newborn EMFG (NEMFG) rats from the EMFG. Testicles were extracted at postnatal day 21. Lipid peroxidation and DNA oxidation levels, apoptotic index and histopathological damage scores were compared. NEMFG rats exhibited irregularities in seminiferous tubule basal membrane and epithelium, immature germ cells in the lumen, and a decreased diameter in seminiferous tubules and thickness of epithelium. Apoptotic index, lipid peroxidation and DNA oxidation were higher in NEMFG rats

than in NCG. 21-day-old rat testicles exposed to 900-MHz EMF in the prenatal term may be adversely affected, and this effect persists after birth.

(NE) Hansteen IL, Lågeide L, Clausen KO, Haugan V, Svendsen M, Eriksen JG, Skiaker R, Hauger E, Vistnes AI, Kure EH. Cytogenetic effects of 18.0 and 16.5 GHz microwave radiation on human lymphocytes in vitro. Anticancer Res 29:2885-2892, 2009a. (VT, AE, GT, WS)

BACKGROUND: There are few cell studies on the direct genotoxic effects of microwave radiation. In this study, cytogenetic effects of microwave radiation alone or in combination with mitomycin C (MMC) were investigated. MATERIALS AND METHODS: Lymphocytes from two smoking and four non-smoking donors were exposed for 53 hours in vitro to 1.0 W/m continuous-wave radiation at 18.0 GHz or 10 W/m pulsed-wave at 16.5 GHz, alone or in combination with MMC. DNA synthesis and repair were inhibited in vitro in some cultures. RESULTS: No synergistic effect was observed in cells exposed to combinations of microwave radiation and in vitro exposure to MMC, or to cells pre-exposed in vivo to tobacco smoke. For the 16.5 GHz pulsed exposure, a non-significant trend consisting of an increase in aberration frequencies with microwave radiation was shown for the DNA synthesis and repair inhibited cultures both with and without MMC. CONCLUSION: Neither 18.0 GHz continuous-wave nor 16.5 GHz pulsed-wave exposure to human lymphocytes in vitro induced statistically significant increases in chromosomal aberration frequencies. 16.5 GHz pulsed-wave exposure requires further documentation before a true negative conclusion can be drawn.

(NE) Hansteen IL, Clausen KO, Haugan V, Svendsen M, Svendsen MV, Eriksen JG, Skiaker R, Hauger E, Lågeide L, Vistnes AI, Kure EH. Cytogenetic effects of exposure to 2.3 GHz radiofrequency radiation on human lymphocytes in vitro. Anticancer Res 29:4323-4330, 2009b. (VT, AE, GT, IX)

BACKGROUND: No previous in vitro studies have tested radio frequency radiation for at least one full cell cycle in culture. The aim was to test if exposure used in mobile phones and wireless network technologies would induce DNA damage in cultured human lymphocytes with and without a known clastogen. MATERIALS AND METHODS: Lymphocytes from six donors were exposed to 2.3 GHz, 10 W/m continuous waves, or 2.3 GHz, 10 W/m pulsed waves (200 Hz pulse frequency, 50% duty cycle). Mitomycin C was added to half of the cultures. DNA synthesis and repair were inhibited in one experiment. RESULTS: No statistically significant differences were observed between control and exposed cultures. A weak trend for more chromosomal damage with the interaction of pulsed fields with mitomycin C compared to a constant field was observed. CONCLUSION: Exposure during the whole cell cycle in inhibited cultures did not resulted in significant differences in chromosomal aberrations as compared to controls.

(E) Hao Y, Yang X, Chen C, Yuan-Wang, Wang X, Li M, Yu Z. STAT3 signalling pathway is involved in the activation of microglia induced by 2.45 GHz electromagnetic fields. . Int J Radiat Biol 86(1):27-36, 2010.(VT, AE, GE)

Purpose: Microglia activation plays a pivotal role in the initiation and progression of central nervous system (CNS) insult. The aim of the present work was to investigate the activation of microglia and involvement of signal transducer and activator of transcription 3 (STAT3) in microglia activation after 2.45 GHz electromagnetic fields (EMF) exposure. **Materials and methods:** In this study, murine N9 microglial cells were exposed to 2.45 GHz EMF, the protein expressions of STAT3, Janus Tyrosine kinase 1 and 2(JAK1 and JAK2), phosphor-(Try705) STAT3 and DNA binding activity of STAT3 were examined by Western blot analysis and electrophoresis mobility shift assay (EMSA). Levels of the nitric oxide (NO) derivative nitrite were determined in the culture medium by the Griess reaction. The mRNA expression of tumour necrosis factor alpha (TNF-alpha) and inducible nitric oxide synthase (iNOS) were detected by reverse transcription and polymerase chain reaction (RT-PCR). **Results:** A significant increase of STAT3 DNA-binding ability was noted after exposure. Consistent with this, EMF rapidly induced phosphorylation of STAT3 and activated JAK1 and JAK2. In addition, EMF exposure increased transcription levels of the inflammation-associated genes, iNOS and TNF-alpha, which are reported to contain STAT-binding elements in their promoter region. P6, a JAK inhibitor, reduced induction of iNOS and TNF-alpha, nuclear factor binding activity, and activation of STAT3 in EMF-stimulated microglia. **Conclusion:** These results provide evidence that EMF exposure can initiate the activation of microglia cells and STAT3 signalling involves in EMF-induced microglial activation.

(E) Hao Y, Li W, Wang H, Zhang J, Wang H, Dong J, Yao B, Xu X, Zhao L, Peng R. Microwave radiation induces neuronal autophagy through miR-30a-5p/AMPKα2 signal pathway. Biosci Rep 42(4):BSR20212584, 2022.(VO, VT, AE, GE)

The potential health hazards of microwaves have attracted much more attention. Our previous study found that 2856 MHz microwave radiation damaged synaptic plasticity and activated autophagy in neurons. However, the mechanisms underlying microwave-induced autophagy were still unclear. In the present study, we established neuronal damage models by exposing rat hippocampal neurons and rat adrenal pheochromocytoma (PC12) cell-derived neuron-like cells to 30 mW/cm2 microwaves, which resulted in miR-30a-5p ('miR-30a' for short) down-regulation and autophagy activation in vivo and in vitro. Bioinformatics analysis was conducted, and Beclin1, Prkaa2, Irs1, Pik3r2, Rras2, Ddit4, Gabarapl2 and autophagy-related gene 12 (Atg12) were identified as potential downstream genes of miR-30a involved in regulating autophagy. Based on our previous findings that microwave radiation could lead to abnormal energy metabolism in neurons, Prkaa2, encoding adenosine 5'-monophosphate-activated protein kinase (AMPK) α2 (AMPKα2, an important catalytic subunit of energy sensor AMPK), was selected for further analysis. Dual-luciferase reporter assay results showed that Prkaa2 was a downstream gene of miR-30a. Moreover, microwave radiation increased the expression of AMPKα2 and the phosphorylation of AMPKα (Thr172) both in vivo and in vitro. The transfection of PC12 cells with miR-30a mimics increased miR-30a levels, reduced AMPKα2 expression, suppressed AMPKα (Thr172) phosphorylation, and inhibited autophagy occurrence in neuron-like cells. Importantly, miR-30a overexpression abolished microwave-activated autophagy and inhibited microwave-induced AMPKα2 up-regulation and AMPKα (Thr172) phosphorylation. In conclusion, microwave radiation promoted the occurrence of autophagy in neurons through the miR-30a/AMPKα2 signal pathway.

(E) Harvey C, French PW, Effects on protein kinase C and gene expression in a human mast cell line, HMC-1, following microwave exposure. Cell Biol Int **23(11):739-748, 2000. (VT, LE, GE)**

We used a resonant cavity which delivered a continuous wave exposure at 864.3 MHz at an average specific absorption rate (SAR) of 7 W/kg to determine non-thermal biological effects of microwave exposure. A human mast cell line, HMC-1, was used as the biological target. Cells were given three exposures each of 20-min duration daily for 7 days. The temperature of the cell culture medium during the exposure fell to 26.5 degrees C. Effects were seen on localization of protein kinase C, and expression of three genes of 588 screened. The affected genes included the proto-oncogene c-kit, the transcription factor Nucleoside diphosphate kinase B and the apoptosis-associated gene DAD-1. Stress response genes were variably upregulated. No significant effect on morphology or on F-actin distribution was detected. We conclude that low-power microwave exposure may act on HMC-1 cells by altering gene expression via a mechanism involving activation of protein kinase C, and at temperatures well below those known to induce a heat shock response.

(E) Hassanzadeh-Taheri M, Khalili MA, Mohebati AH, Zardast G, Doostabadi MR. The detrimental effect of cell phone radiation on sperm biological characteristics in normozoospermic. Andrologia 54(1):e14257, 2022. (VT, AE, GT, RP)

Radiofrequency electromagnetic radiation emitted from cell phone has harmful effects on some organs of the body, such as the brain, heart, and testes. This study aimed to assess the effects of cell phones on sperm parameters, DNA fragmentation, and apoptosis in normozoospermic. Normal sperm samples were divided into two groups of control and case. The samples from the case were placed for 60 min at a distance of approximately 2.5 cm from the cell phone set in the active antenna position. Control samples were exposed to cell phones without active antennas. All specimens were analysed by World Health Organization criteria. Sperm viability, sperm with chromatin abnormality and maturity, DNA fragmentation, and apoptosis were examined. Viability and motility in the case were significantly lower than the control ($p < .001$, $p = .004$ respectively). The percentage of apoptotic sperms and DNA fragmentation were significantly higher in the case when compared with the control ($p = .031$, $p < .001$ respectively). The other parameters studied such as morphology, chromatin abnormality, and maturity showed no significant difference between the case and control groups. Cell phone waves had a detrimental effect on human sperm's biological features. Therefore, it is recommended to keep the cell phone away from the pelvis as much as possible.

(E) He Q, Sun Y, Zong L, Tong J, Cao Y. Induction of Poly(ADP-ribose) Polymerase in Mouse Bone Marrow Stromal Cells Exposed to 900 MHz Radiofrequency Fields: Preliminary Observations. Biomed Res Int 2016:4918691, 2016. (VT, LE, GE, LI)

Background. Several investigators have reported increased levels of poly(ADP-ribose) polymerase-1 (PARP-1), a nuclear enzyme which plays an important role in the repair of damaged DNA, in cells exposed to extremely low dose ionizing radiation which does not cause measurable DNA damage. Objective. To examine whether exposure of the cells to nonionizing radiofrequency fields (RF) is capable of increasing messenger RNA of PARP-1 and its protein

levels in mouse bone marrow stromal cells (BMSCs). Methods. BMSCs were exposed to 900 MHz RF at 120 μW/cm(2) power intensity for 3 hours/day for 5 days. PARP-1 mRNA and its protein levels were examined at 0, 0.5, 1, 2, 4, 6, 8, and 10 hours after exposure using RT-PCR and Western blot analyses. Sham-exposed (SH) cells and those exposed to ionizing radiation were used as unexposed and positive control cells. Results. BMSCs exposed to RF showed significantly increased expression of PARP-1 mRNA and its protein levels after exposure to RF while such changes were not observed in SH-exposed cells. Conclusion. Nonionizing RF exposure is capable of inducing PARP-1.

(E) He Q, Zong L, Sun Y, Vijayalaxmi, PrihodaTJ, Tong J, Cao Y. Adaptive response in mouse bone marrow stromal cells exposed to 900 MHz radiofrequency fields: Impact of poly (ADP-ribose) polymerase (PARP). Mutat Res 820:19-25, 2017. (VT, LE, GE, LI, IX)

This study examined whether non-ionizing radiofrequency fields (RF) exposure is capable of inducing poly (ADP-ribose) polymerase-1 (PARP-1) in bone marrow stromal cells (BMSCs) and whether it plays a role in RF-induced adaptive response (AR). Bone marrow stromal cells (BMSCs) were exposed to 900MHz RF at 120μW/cm^2 power flux density for 3h/day for 5days and then challenged with a genotoxic dose of 1.5Gy gamma-radiation (GR). Some cells were also treated with 3-aminobenzamide (3-AB, 2mM final concentration), a potent inhibitor of PARP-1. Un-exposed and sham (SH)-exposed control cells as well as positive control cells exposed to gamma radiation (GR) were included in the experiments. The expression of PARP-1 mRNA and its protein levels as well as single strand breaks in the DNA and the kinetics of their repair were evaluated at several times after exposures. The results indicated the following. (a) Cells exposed to RF alone showed significantly increased PARP-1 mRNA expression and its protein levels compared with those exposed to SH- and GR alone. (b) Treatment of RF-exposed cells with 3-AB had diminished such increase in PARP-1. (c) Cells exposed to RF+GR showed significantly decreased genetic damage as well as faster kinetics of repair compared with those exposed to GR alone. (d) Cells exposed to RF+3-AB+GR showed no such decrease in genetic damage. Thus, the overall date suggested that non-ionizing RF exposure was capable of inducing PARP-1 which has a role in RF-induced AR.

(E) Hekmat A, Saboury AA, Moosavi-Movahedi AA. The toxic effects of mobile phone radiofrequency (940MHz) on the structure of calf thymus DNA. Ecotoxicol Environ Saf. 88:35-41, 2013. (VT, AE, GT, LI)

Currently, the biological effects of nonionizing electromagnetic fields (EMFs) including radiofrequency (RF) radiation have been the subject of numerous experimental and theoretical studies. The aim of this study is to evaluate the possible biological effects of mobile phone RF (940MHz, 15V/m and SAR=40mW/kg) on the structure of calf thymus DNA (ct DNA) immediately after exposure and 2h after 45min exposure via diverse range of spectroscopic instruments. The UV-vis and circular dichroism (CD) experiments depict that mobile phone EMFs can remarkably cause disturbance on ct DNA structure. In addition, the DNA samples, immediately after exposure and 2h after 45min exposure, are relatively thermally unstable compared to the DNA solution, which was placed in a small shielded box (unexposed ct DNA). Furthermore, the exposed DNA samples (the DNA samples that were exposed to 940MHz EMF)

have more fluorescence emission when compared with the unexposed DNA, which may have occurred attributable to expansion of the exposed DNA structure. The results of dynamic light scattering (DLS) and zeta potential experiments demonstrate that RF-EMFs lead to increment in the surface charge and size of DNA. The structure of DNA immediately after exposure is not significantly different from the DNA sample 2h after 45min exposure. In other words, the EMF-induced conformational changes are irreversible. Collectively, our results reveal that 940MHz can alter the structure of DNA. The displacement of electrons in DNA by EMFs may lead to conformational changes of DNA and DNA disaggregation. Results from this study could have an important implication on the health effects of RF-EMFs exposure. In addition, this finding could proffer a novel strategy for the development of next generation of mobile phone.

(NE) Hintzsche H, Stopper H. Micronucleus frequency in buccal mucosa cells of mobile phone users. Toxicol Lett. 193(1):124-130, 2010. (HU, LE, GT)

Mobile phones are being used extensively throughout the world, with more than four billion accounts existing in 2009. This technology applies electromagnetic radiation in the microwave range. Health effects of this radiation have been subject of debate for a long time, both within the scientific community and within the general public. This study investigated the effect of mobile phone use on genomic instability of the human oral cavity's mucosa cells. 131 Individuals donated buccal mucosa cells extracted by slightly scraping the oral cavity with a cotton swab. Every participant filled out a questionnaire about mobile phone use including duration of weekly use, overall period of exposure and headset usage. 13 Individuals did not use mobile phones at all, 85 reported using the mobile phone for three hours per week or less, and 33 reported use of more than three hours per week. Additionally, information on age, gender, body weight, smoking status, medication and nutrition was retrieved. For staining of the cells a procedure using alpha-tubulin-antibody and chromomycin A(3) was applied. Micronuclei and other markers were evaluated in 1000 cells per individual at the microscope. A second scorer counted another 1000 cells, resulting in 2000 analyzed cells per individual. Mobile phone use did not lead to a significantly increased frequency of micronuclei.

(NE) Hintzsche H, Jastrow C, Kleine-Ostmann T, Schrader T, Stopper H. 900 MHz radiation does not induce micronucleus formation in different cell types. Mutagenesis. 27(4):477-483, 2012a. (VT, AE, GT)

The exposure of the population to non-ionising electromagnetic radiation is still increasing, mainly due to mobile communication. Whether low-intensity electromagnetic fields can cause other effects apart from heating has been a subject of debate. One of the effects, which were proposed to be caused by mobile phone radiation, is the occurrence of mitotic disturbances. The aim of this study was to investigate possible consequences of these mitotic disturbances as manifest genomic damage, i.e. micronucleus induction. Cells were irradiated at a frequency of 900 MHz, which is located in one of the main frequency bands applied for mobile communication. Two cell types were used, HaCaT cells as human cells and A(L) cells (human-hamster hybrid cells), in which mitotic disturbances had been reported to occur. After different post-exposure incubation periods, cells were fixed and micronucleus frequencies were evaluated. Both cell types did not show any genomic damage after exposure. To adapt the protocol for the micronucleus test into the direction of the protocol for mitotic disturbances, the post-exposure

incubation period was reduced and exposure time was extended to one cell cycle length. This did not result in any increase of the genomic damage. In conclusion, micronucleus induction was not observed as a consequence of exposure to non-ionising radiation, even though this agent was reported to cause mitotic disturbances under similar experimental conditions.

(NE) Hintzsche H, Jastrow C, Kleine-Ostmann T, Kärst U, Schrader T, Stopper H. Terahertz electromagnetic fields (0.106 THz) do not induce manifest genomic damage in vitro. PLoS One 7(9):e46397, 2012b. (VT, AE, GT)

Terahertz electromagnetic fields are non-ionizing electromagnetic fields in the frequency range from 0.1 to 10 THz. Potential applications of these electromagnetic fields include the whole body scanners, which currently apply millimeter waves just below the terahertz range, but future scanners will use higher frequencies in the terahertz range. These and other applications will bring along human exposure to these fields. Up to now, only a limited number of investigations on biological effects of terahertz electromagnetic fields have been performed. Therefore, research is strongly needed to enable reliable risk assessment.Cells were exposed for 2 h, 8 h, and 24 h with different power intensities ranging from 0.04 mW/cm(2) to 2 mW/cm(2), representing levels below, at, and above current safety limits. Genomic damage on the chromosomal level was measured as micronucleus formation. DNA strand breaks and alkali-labile sites were quantified with the comet assay. No DNA strand breaks or alkali-labile sites were observed as a consequence of exposure to terahertz electromagnetic fields in the comet assay. The fields did not cause chromosomal damage in the form of micronucleus induction.

(NE) Hirose H, Sakuma N, Kaji N, Suhara T, Sekijima M, Nojima T, Miyakoshi J. Phosphorylation and gene expression of p53 are not affected in human cells exposed to 2.1425 GHz band CW or W-CDMA modulated radiation allocated to mobile radio base stations. Bioelectromagnetics 27:494-504, 2006. (VT, AE, GE)

A large-scale in vitro study focusing on low-level radiofrequency (RF) fields from mobile radio base stations employing the International Mobile Telecommunication 2000 (IMT-2000) cellular system was conducted to test the hypothesis that modulated RF fields induce apoptosis or other cellular stress response that activate p53 or the p53-signaling pathway. First, we evaluated the response of human cells to microwave exposure at a specific absorption rate (SAR) of 80 mW/kg, which corresponds to the limit of the average whole-body SAR for general public exposure defined as a basic restriction by the International Commission on Non-Ionizing Radiation Protection (ICNIRP) guidelines. Second, we investigated whether continuous wave (CW) and wideband code division multiple access (W-CDMA) modulated signal RF fields at 2.1425 GHz induced apoptosis or any signs of stress. Human glioblastoma A172 cells were exposed to W-CDMA radiation at SARs of 80, 250, and 800 mW/kg, and CW radiation at 80 mW/kg for 24 or 48 h. Human IMR-90 fibroblasts from fetal lungs were exposed to both W-CDMA and CW radiation at a SAR of 80 mW/kg for 28 h. Under the RF field exposure conditions described above, no significant differences in the percentage of apoptotic cells were observed between the test groups exposed to RF signals and the sham-exposed negative controls, as evaluated by the Annexin V affinity assay. No significant differences in expression levels of phosphorylated p53 at serine 15 or total p53 were observed between the test groups and the negative controls by the bead-based multiplex assay. Moreover, microarray hybridization and

real-time RT-PCR analysis showed no noticeable differences in gene expression of the subsequent downstream targets of p53 signaling involved in apoptosis between the test groups and the negative controls. Our results confirm that exposure to low-level RF signals up to 800 mW/kg does not induce p53-dependent apoptosis, DNA damage, or other stress response in human cells.

(NE) Hirose H, Sakuma N, Kaji N, Nakayama K, Inoue K, Sekijima M, Nojima T, Miyakoshi J. Mobile phone base station-emitted radiation does not induce phosphorylation of Hsp27. Bioelectromagnetics 28:99-108, 2007. (VT, AE, GE)

An in vitro study focusing on the effects of low-level radiofrequency (RF) fields from mobile radio base stations employing the International Mobile Telecommunication 2000 (IMT-2000) cellular system was conducted to test the hypothesis that modulated RF fields act to induce phosphorylation and overexpression of heat shock protein hsp27. First, we evaluated the responses of human cells to microwave exposure at a specific absorption rate (SAR) of 80 mW/kg, which corresponds to the limit of the average whole-body SAR for general public exposure defined as a basic restriction in the International Commission on Non-Ionizing Radiation Protection (ICNIRP) guidelines. Second, we investigated whether continuous wave (CW) and Wideband Code Division Multiple Access (W-CDMA) modulated signal RF fields at 2.1425 GHz induced activation or gene expression of hsp27 and other heat shock proteins (hsps). Human glioblastoma A172 cells were exposed to W-CDMA radiation at SARs of 80 and 800 mW/kg for 2-48 h, and CW radiation at 80 mW/kg for 24 h. Human IMR-90 fibroblasts from fetal lungs were exposed to W-CDMA at 80 and 800 mW/kg for 2 or 28 h, and CW at 80 mW/kg for 28 h. Under the RF field exposure conditions described above, no significant differences in the expression levels of phosphorylated hsp27 at serine 82 (hsp27[pS82]) were observed between the test groups exposed to W-CDMA or CW signal and the sham-exposed negative controls, as evaluated immediately after the exposure periods by bead-based multiplex assays. Moreover, no noticeable differences in the gene expression of hsps were observed between the test groups and the negative controls by DNA Chip analysis. Our results confirm that exposure to low-level RF field up to 800 mW/kg does not induce phosphorylation of hsp27 or expression of hsp gene family.

(NE) Hook GJ, Zhang P, Lagroye I, Li L, Higashikubo R, Moros EG, Straube WL, Pickard WF, Baty JD, Roti Roti JL. Measurement of DNA damage and apoptosis in molt-4 cells after in vitro exposure to radiofrequency radiation. Radiat Res. 161(2): 193-200, 2004. (VT, AE, LI, GT)

To determine whether exposure to radiofrequency (RF) radiation can induce DNA damage or apoptosis, Molt-4 T lymphoblastoid cells were exposed with RF fields at frequencies and modulations of the type used by wireless communication devices. Four types of frequency/modulation forms were studied: 847.74 MHz code-division multiple-access (CDMA), 835.62 MHz frequency-division multiple-access (FDMA), 813.56 MHz iDEN(R) (iDEN), and 836.55 MHz time-division multiple-access (TDMA). Exponentially growing cells were exposed to RF radiation for periods up to 24 h using a radial transmission line (RTL) exposure system. The specific absorption rates used were 3.2 W/kg for CDMA and FDMA, 2.4 or 24 mW/kg for iDEN, and 2.6 or 26 mW/kg for TDMA. The temperature in the RTLs was maintained at 37 degrees C

+/- 0.3 degrees C. DNA damage was measured using the single-cell gel electrophoresis assay. The annexin V affinity assay was used to detect apoptosis. No statistically significant difference in the level of DNA damage or apoptosis was observed between sham-treated cells and cells exposed to RF radiation for any frequency, modulation or exposure time. Our results show that exposure of Molt-4 cells to CDMA, FDMA, iDEN or TDMA modulated RF radiation does not induce alterations in level of DNA damage or induce apoptosis.

(NE) Hou Q, Wang M, Wu S, Ma X, An G, Liu H, Xie F. Oxidative changes and apoptosis induced by 1800-MHz electromagnetic radiation in NIH/3T3 cells. Electromagn Biol Med 34(1):85-92, 2015. (VT, AE, GT)

To investigate the potential adverse effects of mobile phone radiation, we studied reactive oxygen species (ROS), DNA damage and apoptosis in mouse embryonic fibroblasts (NIH/3T3) after intermittent exposure (5 min on/10 min off, for various durations from 0.5 to 8 h) to an 1800-MHz GSM-talk mode electromagnetic radiation (EMR) at an average specific absorption rate of 2 W/kg. A 2',7'-dichlorofluorescin diacetate fluorescence probe was used to detect intracellular ROS levels, immunofluorescence was used to detect γH2AX foci as a marker for DNA damage, and flow cytometry was used to measure apoptosis. Our results showed a significant increase in intracellular ROS levels after EMR exposure and it reached the highest level at an exposure time of 1 h ($p < 0.05$) followed by a slight decrease when the exposure continued for as long as 8 h. No significant effect on the number of γH2AX was detected after EMR exposure. The percentage of late-apoptotic cells in the EMR-exposed group was significantly higher than that in the sham-exposed groups ($p < 0.05$). These results indicate that an 1800-MHz EMR enhances ROS formation and promotes apoptosis in NIH/3T3 cells.

(E) Houston BJ, Nixon B, McEwan KE, Martin JH, King BV, Aitken RJ, De Iuliis GN. Whole-body exposures to radiofrequency-electromagnetic energy can cause DNA damage in mouse spermatozoa via an oxidative mechanism. Sci Rep. 9(1):17478, 2019. (VO, LE, GT, RP, OX)

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Artificially generated radiofrequency-electromagnetic energy (RF-EME) is now ubiquitous in our environment owing to the utilization of mobile phone and Wi-Fi based communication devices. While several studies have revealed that RF-EME is capable of eliciting biological stress, particularly in the context of the male reproductive system, the mechanistic basis of this biophysical interaction remains largely unresolved. To extend these studies, here we exposed unrestrained male mice to RF-EME generated via a dedicated waveguide (905 MHz, 2.2 W/kg) for 12 h per day for a period of 1, 3 or 5 weeks. The testes of exposed mice exhibited no evidence of gross histological change or elevated stress, irrespective of the RF-EME exposure regimen. By contrast, 5 weeks of RF-EME exposure adversely impacted the vitality and motility profiles of mature epididymal spermatozoa. These spermatozoa also experienced increased mitochondrial generation of reactive oxygen species after 1 week of exposure, with elevated DNA oxidation and fragmentation across all exposure periods. Notwithstanding these lesions, RF-EME exposure did not impair the fertilization competence of spermatozoa nor their ability to support early embryonic development. This study supports the utility of male

germ cells as sensitive tools with which to assess the biological impacts of whole-body RF-EME exposure.

(NE) Huang TQ, Lee MS, Oh E, Zhang BT, Seo JS, Park WY. Molecular responses of Jurkat T-cells to 1763 MHz radiofrequency radiation. Int J Radiat Biol 84:734-741, 2008a. (VT, AE, GT, GE)

PURPOSE: The biological effects of exposure to mobile phone emitted radiofrequency (RF) radiation are the subject of intense study, yet the hypothesis that RF exposure is a potential health hazard remains controversial. In this paper, we monitored cellular and molecular changes in Jurkat human T lymphoma cells after irradiating with 1763 MHz RF radiation to understand the effect on RF radiation in immune cells. MATERIALS AND METHODS: Jurkat T-cells were exposed to RF radiation to assess the effects on cell proliferation, cell cycle progression, DNA damage and gene expression. Jurkat cells were exposed to 1763 MHz RF radiation at 10 W/kg specific absorption rate (SAR) and compared to sham exposed cells. RESULTS: RF exposure did not produce significant changes in cell numbers, cell cycle distributions, or levels of DNA damage. In genome-wide analysis of gene expressions, there were no genes changed more than two-fold upon RF-radiation while ten genes change to 1.3 approximately 1.8-fold. Among ten genes, two cytokine receptor genes such as chemokine (C-X-C motif) receptor 3 (CXCR3) and interleukin 1 receptor, type II (IL1R2) were down-regulated upon RF radiation, but they were not directly related to cell proliferation or DNA damage responses. CONCLUSION: These results indicate that the alterations in cell proliferation, cell cycle progression, DNA integrity or global gene expression was not detected upon 1763 MHz RF radiation under 10 W/kg SAR for 24 h to Jurkat T cells.

(NE) Huang TQ, Lee MS, Oh EH, Kalinec F, Zhang BT, Seo JS, Park WY. Characterization of biological effect of 1763 MHz radiofrequency exposure on auditory hair cells.Int J Radiat Biol 84:909-915, 2008b. (VT, AE, GT, GE)

Purpose: Radiofrequency (RF) exposure at the frequency of mobile phones has been reported not to induce cellular damage in in vitro and in vivo models. We chose HEI-OC1 immortalized mouse auditory hair cells to characterize the cellular response to 1763 MHz RF exposure, because auditory cells could be exposed to mobile phone frequencies. Materials and methods: Cells were exposed to 1763 MHz RF at a 20 W/kg specific absorption rate (SAR) in a code division multiple access (CDMA) exposure chamber for 24 and 48 h to check for changes in cell cycle, DNA damage, stress response, and gene expression. Results: Neither of cell cycle changes nor DNA damage was detected in RF-exposed cells. The expression of heat shock proteins (HSP) and the phosphorylation of mitogen-activated protein kinases (MAPK) did not change, either. We tried to identify any alteration in gene expression using microarrays. Using the Applied Biosystems 1700 full genome expression mouse microarray, we found that only 29 genes (0.09% of total genes examined) were changed by more than 1.5-fold on RF exposure. Conclusion: From these results, we could not find any evidence of the induction of cellular responses, including cell cycle distribution, DNA damage, stress response and gene expression, after 1763 MHz RF exposure at an SAR of 20 W/kg in HEI-OC1 auditory hair cells.

(E) Ibitayo AO, Afolabi OB, Akinyemi AJ, Ojiezeh TI, Adekoya KO, Ojewunmi OO. RAPD Profiling, DNA Fragmentation, and Histomorphometric Examination in Brains of

Wistar Rats Exposed to Indoor 2.5 Ghz Wi-Fi Devices Radiation. Biomed Res Int. 2017:8653286, 2017. (VO, LE, GT)

The advent of Wi-Fi connected high technology devices in executing day-to-day activities is fast evolving especially in developing countries of the world and hence the need to assess its safety among others. The present study was conducted to investigate the injurious effect of radiofrequency emissions from installed Wi-Fi devices in brains of young male rats. Animals were divided into four equal groups; group 1 served as control while groups 2, 3, and 4 were exposed to 2.5 Ghz at intervals of 30, 45, and 60 consecutive days with free access to food and water ad libitum. Alterations in harvested brain tissues were confirmed by histopathological analyses which showed vascular congestion and DNA damage in the brain was assayed using agarose gel electrophoresis. Histomorphometry analyses of their brain tissues showed perivascular congestion and tissue damage as well.

(E) Jeong YJ, Kang GY, Kwon JH, Choi HD, Pack JK, Kim N, Lee YS, Lee HJ. 1950 MHz Electromagnetic Fields Ameliorate Aβ Pathology in Alzheimer's Disease Mice. Curr Alzheimer Res. 12(5):481-492, 2015. (VO, LE, GE)

The involvement of radiofrequency electromagnetic fields (RF-EMF) in the neurodegenerative disease, especially Alzheimer's disease (AD), has received wide consideration, however, outcomes from several researches have not shown consistency. In this study, we determined whether RF-EMF influenced AD pathology in vivo using Tg-5xFAD mice as a model of AD-like amyloid β (Aβ) pathology. The transgenic (Tg)-5xFAD and wild type (WT) mice were chronically exposed to RF-EMF for 8 months (1950 MHz, SAR 5W/kg, 2 hrs/day, 5 days/week). Notably, chronic RFEMF exposure significantly reduced not only Aβ plaques, APP, and APP carboxyl-terminal fragments (CTFs) in whole brain including hippocampus and entorhinal cortex but also the ratio of Aβ42 and Aβ40 peptide in the hippocampus of Tg-5xFAD mice. We also found that parenchymal expression of β-amyloid precursor protein cleaving enzyme 1(BACE1) and neuroinflammation were inhibited by RF-EMF exposure in Tg-5xFAD. In addition, RF-EMF was shown to rescue memory impairment in Tg-5xFAD. Moreover, gene profiling from microarray data using hippocampus of WT and Tg- 5xFAD following RF-EMF exposure revealed that 5 genes (Tshz2, Gm12695, St3gal1, Isx and Tll1), which are involved in Aβ, are significantly altered inTg-5xFAD mice, exhibiting different responses to RF-EMF in WT or Tg-5xFAD mice; RF-EMF exposure in WT mice showed similar patterns to control Tg-5xFAD mice, however, RF-EMF exposure in Tg- 5xFAD mice showed opposite expression patterns. These findings indicate that chronic RF-EMF exposure directly affects Aβ pathology in AD but not in normal brain. Therefore, RF-EMF has preventive effects against AD-like pathology in advanced AD mice with a high expression of Aβ, which suggests that RF-EMF can have a beneficial influence on AD.

(NE) Jeong YJ, Son Y, Han NK, Choi HD, Pack JK, Kim N, Lee YS, Lee HJ. Impact of Long-Term RF-EMF on Oxidative Stress and Neuroinflammation in Aging Brains of C57BL/6 Mice. Int J Mol Sci. 19(7): 2103, 2018. (VO, CE, GT, OX)

The expansion of mobile phone use has raised questions regarding the possible biological effects of radiofrequency electromagnetic field (RF-EMF) exposure on oxidative stress and brain inflammation. Despite accumulative exposure of humans to radiofrequency electromagnetic fields (RF-EMFs) from mobile phones, their long-term effects on oxidative stress and neuroinflammation in the aging brain have not been studied. In the present study, middle-aged C57BL/6 mice (aged 14 months) were exposed to 1950 MHz electromagnetic fields for 8 months (specific absorption rate (SAR) 5 W/kg, 2 h/day, 5 d/week). Compared with those in the young group, levels of protein (3-nitro-tyrosine) and lipid (4-hydroxy-2-nonenal) oxidative damage markers were significantly increased in the brains of aged mice. In addition, levels of markers for DNA damage (8-hydroxy-2'-deoxyguanosine, p53, p21, γH2AX, and Bax), apoptosis (cleaved caspase-3 and cleaved poly(ADP-ribose) polymerase 1 (PARP-1)), astrocyte (GFAP), and microglia (Iba-1) were significantly elevated in the brains of aged mice. However, long-term RF-EMF exposure did not change the levels of oxidative stress, DNA damage, apoptosis, astrocyte, or microglia markers in the aged mouse brains. Moreover, long-term RF-EMF exposure did not alter locomotor activity in aged mice. Therefore, these findings indicate that long-term exposure to RF-EMF did not influence age-induced oxidative stress or neuroinflammation in C57BL/6 mice.

(E) Jeong, Y.J., Son,Y., Choi, H-D., Kim, N., Lee, Y-S., Kom, Y-G., Lee, H-J (2020). Behavioral changes and gene profile alterations after chronic 1,950-MHz radiofrequency exposure: An observation in C57BL/6 mice. Brain Behav. 10(11):e01815, 2020. (VO, LE, GE)

Introduction: Due to public concerns about deleterious biological consequences of radiofrequency electromagnetic fields (RF-EMF), the potential effects of RF-EMF on the central nervous system have received wide consideration. Methods: Here, two groups of C57BL/6 mice, aged 2 and 12 months, were exposed to 1,950-MHz RF-EMF at a specific absorption rate of 5.0 W/kg for chronic periods (2 hr/day and 5 days/week for 8 months). Behavioral changes were then assessed in the mice at 10 months (sham- or RF-10M) and 20 months (sham- or RF-20M), on the open-field test, the Y-maze test, and an object recognition memory task, while biological effects were analyzed via microarray gene profiling of the hippocampus. Results: Open-field test results showed a decrease in the time duration spent at the center while there was a decrease in enhanced memory shown by the Y-maze test and the novel object recognition test in the RF-20M mice, compared to sham-exposed mice, but no significant changes in the RF-10M group. Based on a 2-fold change cutoff, the microarray data revealed that 15 genes, which are listed as being involved in neurogenesis on Gene Ontology, were altered in both groups. Quantitative real-time PCR for validation showed increased expression of Epha8 and Wnt6 in the hippocampi of RF-20M group mice, although 13 additional genes showed no significant changes following RF-EMF exposure. Conclusion: Therefore, cognitive enhancement following chronic exposure for 8 months to RF-EMF from middle age may be associated with increases in neurogenesis-related signals in the hippocampus of C57BL/6 mice.

(E) Ji S, Oh E, Sul D, Choi JW, Park H, Lee E. DNA Damage of lymphocytes in volunteers after 4 hours use of mobile phone. J Prev Med Public Health. 37(4):373-380, 2004. (HU, AE, GT)

OBJECTIVES: There has been gradually increasing concern about the adverse health effects of electromagnetic radiation originating from cell phones which are widely used in modern life. Cell phone radiation may affect human health by increasing free radicals of human blood cells. This study has been designed to identify DNA damage of blood cells by electromagnetic radiation caused by cell phone use. **METHODS:** This study investigated the health effect of acute exposure to commercially available cell phones on certain parameters such as an indicator of DNA damage for 14 healthy adult volunteers. Each volunteer during the experiment talked over the cell phone with the keypad facing the right side of the face for 4 hours. The single cell gel electrophoresis assay (Comet assay), which is very sensitive in detecting the presence of DNA strand-breaks and alkali-labile damage in individual cells, was used to assess peripheral blood cells (T-cells, B-cells, granulocytes) from volunteers before and after exposure to cell phone radiation. The parameters of Comet assay measured were Olive Tail Moment and Tail DNA %. **RESULTS:** The Olive Tail Moment of B-cells and granulocytes and Tail DNA % of B-cells and granulocytes were increased by a statistically significant extent after 4- hour use of a cell phone compared with controls. **CONCLUSIONS:** It is concluded that cell phone radiation caused the DNA damage during the 4 hours of experimental condition. Nonetheless, this study suggested that cell phone use may increase DNA damage by electromagnetic radiation and other contributing factors.

(E) Ji Y, He Q, Sun Y, Tong J, Cao Y. Adaptive response in mouse bone-marrow stromal cells exposed to 900-MHz radiofrequency fields: Gamma-radiation-induced DNA strand breaks and repair. J Toxicol Environ Health A. 79(9-10):419-426, 2016. (VT, LE, LI, GT, IX) (protective effect on DN damage)

The aim of this study was to examine whether radiofrequency field (RF) preexposure induced adaptive responses (AR) in mouse bone-marrow stromal cells (BMSC) and the mechanisms underlying the observed findings. Cells were preexposed to 900-MHz radiofrequency fields (RF) at 120 μW/cm(2) power intensity for 4 h/d for 5 d. Some cells were subjected to 1.5 Gy γ-radiation (GR) 4 h following the last RF exposure. The intensity of strand breaks in the DNA was assessed immediately at 4 h. Subsequently, some BMSC were examined at 30, 60, 90, or 120 min utilizing the alkaline comet assay and γ-H2AX foci technique. Data showed no significant differences in number and intensity of strand breaks in DNA between RF-exposed and control cells. A significant increase in number and intensity of DNA strand breaks was noted in cells exposed to GR exposure alone. RF followed by GR exposure significantly decreased number of strand breaks and resulted in faster kinetics of repair of DNA strand breaks compared to GR alone. Thus, data suggest that RF preexposure protected cells from damage induced by GR. Evidence indicates that in RF-mediated AR more rapid repair kinetics occurs under conditions of GR-induced damage, which may be attributed to diminished DNA strand breakage.

(E) Jiang B, Nie J, Zhou Z, Zhang J, Tong J, Cao Y. Adaptive response in mice exposed to 900 MHz radiofrequency fields: primary DNA damage. PLoS One. 7(2):e32040, 2012. (VO, LE, GT, IA)

The phenomenon of adaptive response (AR) in animal and human cells exposed to ionizing radiation is well documented in scientific literature. We have examined whether such AR could be induced in mice exposed to non-ionizing radiofrequency fields (RF) used for wireless communications. Mice were pre-exposed to 900 MHz RF at 120 μW/cm(2) power density for 4 hours/day for 1, 3, 5, 7 and 14 days and then subjected to an acute dose of 3 Gy γ-radiation. The primary DNA damage in the form of alkali labile base damage and single strand breaks in the DNA of peripheral blood leukocytes was determined using the alkaline comet assay. The results indicated that the extent of damage in mice which were pre-exposed to RF for 1 day and then subjected to γ-radiation was similar and not significantly different from those exposed to γ-radiation alone. However, mice which were pre-exposed to RF for 3, 5, 7 and 14 days showed progressively decreased damage and was significantly different from those exposed to γ-radiation alone. Thus, the data indicated that RF pre-exposure is capable of inducing AR and suggested that the pre-exposure for more than 4 hours for 1 day is necessary to elicit such AR.

(E) Jiang B, Zong C, Zhao H, Ji Y, Tong J, Cao Y. Induction of adaptive response in mice exposed to 900MHz radiofrequency fields: application of micronucleus assay. Mutat Res 751(2):127-129, 2013. (VO, LE, GT, IX)

Adult male ICR mice were pre-exposed to non-ionizing radiofrequency fields (RF), 900MHz at 120μW/cm(2) power density for 4h/day for 7 days (adaptation dose, AD) and then subjected to an acute whole body dose of 3Gy γ-radiation (challenge dose, CD). The classical micronucleus (MN) assay was used to determine the extent of genotoxicity in immature erythrocytes in peripheral blood and bone marrow. The data obtained in mice exposed to AD+CD were compared with those exposed to CD alone. The results indicated that in both tissues, the MN indices were similar in un-exposed controls and those exposed to AD alone while a significantly increased MN frequency was observed in mice exposed to CD alone. Exposure of mice to AD+CD resulted in a significant decrease in MN indices compared to those exposed to CD alone. Thus, the data suggested that pre-exposure of mice to non-ionizing RF is capable of 'protecting' the erythrocytes in the blood and bone marrow from genotoxic effects of subsequent γ-radiation. Such protective phenomenon is generally described as 'adaptive response' (AR) and is well documented in human and animal cells which were pre-exposed to very low doses of ionizing radiation. It is interesting to observe AR being induced by non-ionizing RF.

(E) Jin H, Kim K, Park GY, Kim M, Lee H-J, Jeon S, Kim JH, Kim HR, Lim KM, Lee YS. The Protective Effects of EMF-LTE against DNA Double-Strand Break Damage In Vitro and In Vivo. Int J Mol Sci 22(10):5134, 2021. (VT, VO, LE, AE, GT, IX)

With the rapid growth of the wireless communication industry, humans are extensively exposed to electromagnetic fields (EMF) comprised of radiofrequency (RF). The skin is considered the

primary target of EMFs given its outermost location. Recent evidence suggests that extremely low frequency (ELF)-EMF can improve the efficacy of DNA repair in human cell-lines. However, the effects of EMF-RF on DNA damage remain unknown. Here, we investigated the impact of EMF-long term evolution (LTE, 1.762 GHz, 8 W/kg) irradiation on DNA double-strand break (DSB) using the murine melanoma cell line B16 and the human keratinocyte cell line HaCaT. EMF-LTE exposure alone did not affect cell viability or induce apoptosis or necrosis. In addition, DNA DSB damage, as determined by the neutral comet assay, was not induced by EMF-LTE irradiation. Of note, EMF-LTE exposure can attenuate the DNA DSB damage induced by physical and chemical DNA damaging agents (such as ionizing radiation (IR, 10 Gy) in HaCaT and B16 cells and bleomycin (BLM, 3 μM) in HaCaT cells and a human melanoma cell line MNT-1), suggesting that EMF-LTE promotes the repair of DNA DSB damage. The protective effect of EMF-LTE against DNA damage was further confirmed by attenuation of the DNA damage marker γ-H2AX after exposure to EMF-LTE in HaCaT and B16 cells. Most importantly, irradiation of EMF-LTE (1.76 GHz, 6 W/kg, 8 h/day) on mice in vivo for 4 weeks reduced the γ-H2AX level in the skin tissue, further supporting the protective effects of EMF-LTE against DNA DSB damage. Furthermore, p53, the master tumor-suppressor gene, was commonly upregulated by EMF-LTE irradiation in B16 and HaCaT cells. This finding suggests that p53 plays a role in the protective effect of EMF-LTE against DNA DSBs. Collectively, these results demonstrated that EMF-LTE might have a protective effect against DNA DSB damage in the skin, although further studies are necessary to understand its impact on human health.

(NE) Juutilainen J, Heikkinen P, Soikkeli H, Mäki-Paakkanen J. Micronucleus frequency in erythrocytes of mice after long-term exposure to radiofrequency radiation. Int J Radiat Biol. 83(4):213-220, 2007. (VO, LE, GT)

PURPOSE: The aim of the study was to investigate genotoxicity of long-term exposure to radiofrequency (RF) electromagnetic fields by measuring micronuclei in erythrocytes. The blood samples were collected in two animal studies evaluating possible cocarcinogenic effects of RF fields. METHODS: In study A, female CBA/S mice were exposed for 78 weeks (1.5 h/d, 5 d/week) to either a continuous 902.5 MHz signal similar to that emitted by analog NMT (Nordic Mobile Telephone) phones at a whole-body specific absorption rate (SAR) of 1.5 W/kg, or to a pulsed 902.4 MHz signal similar to that of digital GSM (Global System for Mobile Communications) phones at 0.35 W/kg. A third group was sham-exposed, and a fourth group served as cage controls. All but the cage control animals were exposed to 4 Gy of x-rays during three first weeks of the experiment. In study B, female transgenic mice (line K2) and their nontransgenic littermates were exposed for 52 weeks (1.5 h/d, 5 d/week). Two digital mobile phone signals, GSM and DAMPS (Digital Advanced Mobile Phone System), were used at 0.5 W/kg. All but the cage-control animals were exposed 3 times per week to an ultraviolet radiation dose of 1.2 MED (minimum erythema dose). RESULTS AND CONCLUSIONS: The results did not show any effects of RF fields on micronucleus frequency in polychromatic or normochromatic erythrocytes. The results were consistent in two mouse strains (and in a transgenic variant of the second strain), after 52 or 78 weeks of exposure, at three SAR levels relevant to human exposure from mobile phones, and for three different mobile signals.

(NE) Kadeh H, Saravani S, Moradi M, Alimanesh N. A Comparative Evaluation of the Genotoxic Effects of Mobile Phone Radiation Using Buccal Micronucleus Assay. J Dent (Shiraz) 24(1 Suppl):118-124, 2023. (HU, LE, GT)

Statement of the problem: Mobile usage has increased worldwide over the past two decades. There are conflicting reports about the carcinogenic effects of cell phone radiation on the oral mucosa. Micronucleus (MN) is considered a reliable marker for genotoxic damage. **Purpose:** This study aimed to identify the impact of mobile phone radiation on the MN frequency in oral mucosal cells. **Materials and method:** In this descriptive-analytical study, 50 mobile phone users between the age group of 20-38 years were included. Samples were obtained from the right and left cheek mucosa of each subject (a total 100 cell samples). Every participant filled out a questionnaire about his or her cell phone usage habits. Additionally, personal information such as age, gender, and body mass index (BMI) were assessed. The Feulgen and Papanicolaou staining methods were used for staining of the cell samples. A total of 1000 cells in each sample were evaluated for MNs. **Results:** The mean number of MN in exposed and non-exposed mucosa by Feulgen method was 0.71±1.13 and 0.57±1.36, respectively. Also in Papanicolaou staining, the mean number of MN in the exposed mucosa and non-exposed mucosa was 6.94±6.61 and 6.54±6.88, respectively, but these differences were not significant ($p > 0.05$). The frequency of MN in non-specific DNA staining was significantly (5- to 6-fold) higher than DNA-specific staining. We observed no statically significant differences between MN frequency according to age, gender, BMI, and other cell phone usage habits ($p > 0.05$). **Conclusion:** This study showed that cell phone use does not cause genotoxic effects in the buccal mucosa in the oral cavity. Moreover, using non-specific DNA staining methods can increase the frequency of MN by more than 5- to 6-fold.

(NE) Kao HK, Li Q, Flynn B, Qiao X, Ruberti JW, Murphy GF, Guo L. Collagen synthesis modulated in wounds treated by pulsed radiofrequency energy. Plast Reconstr Surg. 131(4):490e-498e, 2013.(VT, AE, GE)

BACKGROUND: Chronic wounds are biochemically complex and are associated with insufficient cell proliferation, angiogenesis, and extracellular matrix remodeling. The mechanisms by which pulsed radiofrequency energy modulates wound healing are still unclear. METHODS: Db/db mice were wounded and exposed to pulsed radiofrequency energy. Gross closure, cell proliferation, and morphometric analysis of CD31-stained wound cross-sections were assessed. The mRNA expression of profibrotic factors (transforming growth factor-β and platelet-derived growth factor-A), angiogenetic factors (vascular endothelial growth factor and basic fibroblast growth factor), and extracellular matrix components (collagen I and α-smooth muscle actin) were evaluated by quantitative reverse-transcriptase polymerase chain reaction. Collagen protein level of the wound was determined by Western blot analysis. To test the effect of pulsed radiofrequency energy on cell movement in wound healing, cell migration was monitored in monolayer dermal fibroblast cultures. The degree of collagen alignment and gelation time was quantitatively assessed using image analysis techniques. RESULTS: Pulsed radiofrequency energy-treated wounds were characterized by dermal cell proliferation and increased collagen synthesis. By contrast, the CD31 density and the mRNA expression of vascular endothelial growth factor and basic fibroblast growth factor showed no significant

difference between the pulsed radiofrequency energy-treated wounds and the sham group. The pulsed radiofrequency energy-treated dermal fibroblast cultures expressed a significantly longer gelation time compared with the sham-exposed cultures. CONCLUSIONS: Exposing wounds to pulsed radiofrequency accelerated wound healing in this diabetic mouse model by means of significantly increasing dermal cell proliferation and collagen synthesis. A cellular mechanism behind these observations has been proposed.

(E) Karaca E, Durmaz B, Altug H, Yildiz T, Guducu C, Irgi M, Koksal MG, Ozkinay F, Gunduz C, Cogulu O. The genotoxic effect of radiofrequency waves on mouse brain. J Neurooncol 106:53-58, 2012. (VT, LE, GT, GE)

Concerns about the health effects of radiofrequency (RF) waves have been raised because of the gradual increase in usage of cell phones, and there are scientific questions and debates about the safety of those instruments in daily life. The aim of this study is to evaluate the genotoxic effects of RF waves in an experimental brain cell culture model. Brain cell cultures of the mice were exposed to 10.715 GHz with specific absorbtion rate (SAR) 0.725 W/kG signals for 6 h in 3 days at 25°C to check for the changes in the micronucleus (MNi) assay and in the expression of 11 proapoptotic and antiapoptotic genes. It was found that MNi rate increased 11-fold and STAT3 expression decreased 7-fold in the cell cultures which were exposed to RF. Cell phones which spread RF may damage DNA and change gene expression in brain cells.

(E) Keleş AI, Süt BB Histopathological and epigenetic alterations in the spinal cord due to prenatal electromagnetic field exposure: An H3K27me3-related mechanism. Toxicol Ind Health 37(4):189-197, 2021. (VO, LE, DE, EP)

Neural system development is one of the most important stages of embryogenesis. Perturbations in this crucial process due to genetic and environmental risk factors cause neural tube defects and other central nervous system diseases. We investigated the effects of prenatal exposure to 900-MHz electromagnetic field (EMF) on the spinal cord. Pregnant rats were exposed to 900-MHz EMF for 1 h/day from E13.5 until birth. Six pups from the control and EMF groups were sacrificed at postnatal day 32, and the upper thoracic region of the spine was removed and processed for histological procedures. For histopathological analyses, hematoxylineosin staining and, for stereological analyses and the quantitation of motor neurons, cresyl violet staining was performed. H3K27me3 levels were determined via immunofluorescence staining. Histopathological analysis identified structural alterations of ependymal cells, enlarged central canals, as well as degenerated and shrunken motor neurons in the EMF group, while the control group tissues had normal appearances. We also observed enrichment of H3K27me3 in the ependymal cells and the motor neurons in the spinal cord of the control group rats, while the EMF group had low levels of H3K27me3 staining. Our results suggest that the loss of H3K27me3 signals might correlate with reduced neuronal stem cell potential in the EMF group and result in anatomical and structural differences in the spinal cord. This study provided a comprehensive histopathological analysis of the spinal cord after prenatal EMF exposure and offered an H3K27me3-dependent molecular explanation for the detrimental effects of EMF exposure on the spine.

(NE) Kerbacher JJ, Meltz ML, Erwin DN, Influence of radiofrequency radiation on chromosome aberrations in CHO cells and its interaction with DNA-damaging agents. Radiat Res 123(3):311-319, 1990. (VT, AE, GT, IX)

A limited number of contradictory reports have appeared in the literature about the ability of radiofrequency (rf) radiation to induce chromosome aberrations in different biological systems. The technical documentation associated with such reports is often absent or deficient. In addition, no information is available as to whether any additional genotoxic hazard would result from a simultaneous exposure of mammalian cells to rf radiation and a chemical which (by itself) induces chromosome aberrations. In the work described, we have therefore tested two hypotheses. The first is that rf radiation by itself, at power densities and exposure conditions which are higher than is consistent with accepted safety guidelines, can induce chromosome aberrations in mammalian cells. The second is that, during a simultaneous exposure to a chemical known to be genotoxic, rf radiation can affect molecules, biochemical processes, or cellular organelles, and thus result in an increase or decrease in chromosome aberrations. Mitomycin C (MMC) and Adriamycin (ADR) were selected because they act by different mechanisms, and because they might put normal cells at risk during combined-modality rf radiation (hyperthermia)-chemotherapy treatment of cancer. The studies were performed with suitable 37 degrees C and equivalent convection heating-temperature controls in a manner designed to discriminate between any thermal and possible nonthermal action. Radiofrequency exposures were conducted for 2 h under conditions resulting in measurable heating (a maximum increase of 3.2 degrees C), with pulsed-wave rf radiation at a frequency of 2450 MHz and an average net forward power of 600 W, resulting in an SAR of 33.8 W/kg. Treatments with MMC or ADR were for a total of 2.5 h and encompassed the 2-h rf radiation exposure period. The CHO cells from each of the conditions were subsequently analyzed for chromosome aberrations. In cells exposed to rf radiation alone, and where a maximum temperature of approximately 40 degrees C was achieved in the tissue culture medium, no alteration in the frequency from 37 degrees C control levels was observed. Relative to the chemical treatment with MMC alone at 37 degrees C, for two different concentrations, no alteration was observed in the extent of chromosome aberrations induced by either simultaneous rf radiation exposure or convection heating to equivalent temperatures. At the ADR concentration that was used, most of the indices of chromosome aberrations which were scored indicated a similar result.

(E) Kesari KK, Behari J. Fifty-gigahertz Microwave exposure effect of radiations on rat brain. Appl Biochem Biotechnol 158:126-139, 2009. (VO, LE, GT, OX)

The object of this study is to investigate the effects of 50-GHz microwave radiation on the brain of Wistar rats. Male rats of the Wistar strain were used in the study. Animals of 60-day age were divided into two groups-group 1, sham-exposed, and group 2, experimental (microwave-exposed). The rats were housed in a temperature-controlled room (25 degrees C) with constant humidity (40-50%) and received food and water ad libitum. During exposure, rats were placed in Plexiglas cages with drilled ventilation holes and kept in an anechoic chamber. The animals were exposed for 2 h a day for 45 days continuously at a power level of 0.86 muW/cm with nominal

specific absorption rate 8.0 x 10(-4) w/kg. After the exposure period, the rats were killed and homogenized, and protein kinase C (PKC), DNA double-strand break, and antioxidant enzyme activity [superoxides dismutase (SOD), catalase, and glutathione peroxidase (GPx)] were estimated in the whole brain. Result shows that the chronic exposure to these radiations causes DNA double-strand break (head and tail length, intensity and tail migration) and a significant decrease in GPx and SOD activity ($p = <0.05$) in brain cells, whereas catalase activity shows significant increase in the exposed group of brain samples as compared with control ($p = <0.001$). In addition to these, PKC decreased significantly in whole brain and hippocampus ($p < 0.05$). All data are expressed as mean +/- standard deviation. We conclude that these radiations can have a significant effect on the whole brain.

(E) Kesari KK, Behari J, Kumar S. Mutagenic response of 2.45 GHz radiation exposure on rat brain. Int J Radiat Biol 86:334-343, 2010. (VO, LE, GT, OX)

Purpose: To investigate the effect of 2.45 GHz microwave radiation on rat brain of male wistar strain. Material and methods: Male rats of wistar strain (35 days old with 130 +/- 10 g body weight) were selected for this study. Animals were divided into two groups: Sham exposed and experimental. Animals were exposed for 2 h a day for 35 days to 2.45 GHz frequency at 0.34 mW/cm power density. The whole body specific absorption rate (SAR) was estimated to be 0.11 W/Kg. Exposure took place in a ventilated Plexiglas cage and kept in anechoic chamber in a far field configuration from the horn antenna. After the completion of exposure period, rats were sacrificed and the whole brain tissue was dissected and used for study of double strand DNA (Deoxyribonucleic acid) breaks by micro gel electrophoresis and the statistical analysis was carried out using comet assay (IV-2 version software). Thereafter, antioxidant enzymes and histone kinase estimation was also performed. Results: A significant increase was observed in comet head ($P < 0.002$), tail length ($P < 0.0002$) and in tail movement ($P < 0.0001$) in exposed brain cells. An analysis of antioxidant enzymes glutathione peroxidase ($P < 0.005$), and superoxide dismutase ($P < 0.006$) showed a decrease while an increase in catalase ($P < 0.006$) was observed. A significant decrease ($P < 0.023$) in histone kinase was also recorded in the exposed group as compared to the control (sham-exposed) ones. One-way analysis of variance (ANOVA) method was adopted for statistical analysis. Conclusion: The study concludes that the chronic exposure to these radiations may cause significant damage to brain, which may be an indication of possible tumour promotion (Behari and Paulraj 2007).

(E) Kesari KK, Kumar S, Behari J. Effects of radiofrequency electromagnetic wave exposure from cellular phones on the reproductive pattern in male Wistar rats. Appl Biochem Biotechnol. 164(4):546-559, 2011. (VO, LE, GT, OX)

The present study investigates the effect of free radical formation due to mobile phone exposure and effect on fertility pattern in 70-day-old male Wistar rats (sham exposed and exposed). Exposure took place in Plexiglas cages for 2 h a day for 35 days to mobile phone frequency. The specific absorption rate was estimated to be 0.9 W/kg. An analysis of antioxidant enzymes glutathione peroxidase ($P < 0.001$) and superoxide dismutase ($P < 0.007$) showed a decrease, while an increase in catalase ($P < 0.005$) was observed. Malondialdehyde ($P < 0.003$) showed an increase and histone kinase ($P = 0.006$) showed a significant decrease in the exposed group. Micronuclei also show a significant decrease ($P < 0.002$) in the exposed group. A significant

change in sperm cell cycle of G(0)-G(1) (P = 0.042) and G(2)/M (P = 0.022) were recorded. Generation of free radicals was recorded to be significantly increased (P = 0.035). Our findings on antioxidant, malondialdehyde, histone kinase, micronuclei, and sperm cell cycle are clear indications of an infertility pattern, initiated due to an overproduction of reactive oxygen species. It is concluded that radiofrequency electromagnetic wave from commercially available cell phones might affect the fertilizing potential of spermatozoa.

(E) Kesari KK, Meena R, Nirala J, Kumar J, Verma HN. Effect of 3G cell phone exposure with computer controlled 2-D stepper motor on non-thermal activation of the hsp27/p38MAPK stress pathway in rat brain. Cell Biochem Biophys. 68(2):347-358, 2014. (VO, LE, GT, GE)

Cell phone radiation exposure and its biological interaction is the present concern of debate. Present study aimed to investigate the effect of 3G cell phone exposure with computer controlled 2-D stepper motor on 45-day-old male Wistar rat brain. Animals were exposed for 2 h a day for 60 days by using mobile phone with angular movement up to zero to 30°. The variation of the motor is restricted to 90° with respect to the horizontal plane, moving at a pre-determined rate of 2° per minute. Immediately after 60 days of exposure, animals were scarified and numbers of parameters (DNA double-strand break, micronuclei, caspase 3, apoptosis, DNA fragmentation, expression of stress-responsive genes) were performed. Result shows that microwave radiation emitted from 3G mobile phone significantly induced DNA strand breaks in brain. Meanwhile a significant increase in micronuclei, caspase 3 and apoptosis were also observed in exposed group (P < 0.05). Western blotting result shows that 3G mobile phone exposure causes a transient increase in phosphorylation of hsp27, hsp70, and p38 mitogen-activated protein kinase (p38MAPK), which leads to mitochondrial dysfunction-mediated cytochrome c release and subsequent activation of caspases, involved in the process of radiation-induced apoptotic cell death. Study shows that the oxidative stress is the main factor which activates a variety of cellular signal transduction pathways, among them the hsp27/p38MAPK is the pathway of principle stress response. Results conclude that 3G mobile phone radiations affect the brain function and cause several neurological disorders.

(NE) Khalil, A.M., Ashamali, A.M., Gagaa, M. Detection of oxidative stress induced by mobile phone radiation in tissues of mice using 8-0xo-7, 8-dihydro-2'-deoxyguanosine as a biomarker. Int. J. Biol. Biomol. Agricul. Food Biotech. Eng. 5: 240-245, 2011. (VO, LE, GT, OX)

We investigated oxidative DNA damage caused by radio frequency radiation using 8-oxo-7, 8-dihydro-2'- deoxyguanosine (8-oxodG) generated in mice tissues after exposure to 900 MHz mobile phone radio frequency in three independent experiments. The RF was generated by a Global System for Mobile Communication (GSM) signal generator. The radio frequency field was adjusted to 25 V/m. The whole body specific absorption rate (SAR) was 1.0 W/kg. Animals were exposed to this field for 30 min daily for 30 days. 24 h post-exposure, blood serum, brain and spleen were removed and DNA was isolated. Enzyme-linked immunosorbent assay (ELISA) was used to measure 8-oxodG concentration. All animals survived the whole experimental period. The body weight of animals did not change significantly at the end of the experiment. No

statistically significant differences observed in the levels of oxidative stress. Our results are not in favor of the hypothesis that 900 MHz RF induces oxidative damage.

(E) Khalil AM, Gagaa M, Alshamali A. 8-Oxo-7, 8-dihydro-2'-deoxyguanosine as a biomarker of DNA damage by mobile phone radiation. Hum ExpToxicol 31(7):734-740, 2012. (VO, AE, GT, OX)

We examined the effect of exposure to mobile phone 1800 MHz radio frequency radiation (RFR) upon the urinary excretion of 8-oxo-7, 8-dihydro-2'-deoxyguanosine (8-oxodG), one major form of oxidative DNA damage, in adult male Sprague-Dawley rats. Twenty-four rats were used in three independent experiments (RFR exposed and control, 12 rats, each). The animals were exposed to RFR for 2 h from Global System for Mobile Communications (GSM) signal generator with whole-body-specific absorption rate of 1.0 W/kg. Urine samples were collected from the rat while housed in a metabolic cage during the exposure period over a 4-h period at 0.5, 1.0, 2.0 and 4.0 h from the beginning of exposure. In the control group, the signal generator was left in the turn-off position. The creatinine-standardized concentrations of 8-oxodG were measured. With the exception of the urine collected in the last half an hour of exposure, significant elevations were noticed in the levels of 8-oxodG in urine samples from rats exposed to RFR when compared to control animals. Significant differences were seen overall across time points of urine collection with a maximum at 1 h after exposure, suggesting repair of the DNA lesions leading to 8-oxodG formation.

(NE) Khalil AM, Khadra KMA, Aljaberi AM, Gagaa MH, Issa HS. Assessment of oxidant/antioxidant status in saliva of cell phone users. Electromagn Biol Med 33(2):92-97, 2014. (HU, AE, oxidative DNA damage)

Hazardous health effects resulting from exposure to radiofrequency electromagnetic radiation (RF-EMR) emitted from cell phones have been reported in the literature. However, the cellular and molecular targets of RF-EMR are still controversial. The aim of this study was to examine the oxidant/antioxidant status in saliva of cell phone users. Saliva samples collected before using a cell phone as well as at the end of 15 and 30 min calls were tested for two commonly used oxidative stress biomarkers: malondialdehyde (MDA) and 8-oxo-7,8-dihydro-2'-deoxyguanosine (8-Oxo-dG). The 8-oxo-dG levels were determined by enzyme-linked immunosorbent (ELISA) competitive assay, while the MDA levels were measured using the OxiSelect MDA adduct ELISA Kit. The antioxidant capacity of the saliva was evaluated using the oxygen radical absorption capacity (ORAC) and the hydroxyl radical averting capacity (HORAC) assays according to the manufacture instructions. The mean 8-oxo-dG and the Bradford protein concentrations (ng/ml and mg/ml, respectively) peaked at 15 min. The levels of HORAC, ORAC and MDA progressively increased with time and reached maximum at 30 min. However, there was no significant effect of talking time on the levels of 8-OxodG and MDA. Similarly, there was no statistically significant effect of talking time on the oxygen and hydroxyl radicals averting capacities, (ORAC) and (HORAC), respectively. These findings suggest that there is no relationship between exposure to radio frequency radiation (RFR) and changes in the salivary oxidant/antioxidant profile.

(E) Khodamoradi E, Afrashi S, Khoshgard K, Fathi F, Shahasavari S, Azmoonfar R, Najafi M. Simultaneous effect of gamma and Wi-Fi radiation on gamma-H2Ax expression in peripheral blood of rat: A radio-protection note. Biochem Biophys Rep. 30, 101233, 2022. (VO, AE, GT, IX)

Introduction. Nuclear medicine patients are isolated in a room after the injection of a radiopharmaceutical. They may be active Wi-Fi option of its smartphone mobile or other environmental radiofrequency waves. The hypothesis of this study was the evaluation of increased biological effects of the simultaneous exposure to gamma-ray and the Wi-Fi waves by measuring the level of the increased double strand-breaks DNA in peripheral blood lymphocyte in the rat. Materials and methods. Fifty male Wistar rats were exposed for 2, 24, and 72 h only by Wi-Fi, 99m Tc, and simultaneously by Wi-Fi and 99m Tc. The power density levels of Wi-Fi emitter at 15 cm was 4.2nW/ cm2. An activity of 100 μCi of 99m Tc was injected intraperitoneally. Blood samples were taken by cardiac puncture following general anesthesia. Mononuclear cells are extraction by Ficoll-Hypaque density gradient centrifugation. The number of gamma-H2AX foci per nucleus was counted by flow cytometry. The statistical differences between experimental groups at 2, 24, and 72 h were determined with a repeated measure's analysis of variance. The significant difference between groups at the same time was analyzed with the Kruskal-Wallis Test. Results The manner of gamma-H2AX expression was not the same for three groups in time. The number of gamma-H2AX foci between the three groups was a significant difference after 72 h. Conclusion Simultaneous Wi-Fi and gamma-ray exposures can increase the number of double-strand break DNA in peripheral blood lymphocytes to exposure of gamma-ray to 72 h after technetium injection in the rat.

(E) Kim JY, Hong SY, Lee YM, Yu SA, Koh WS, Hong JR, Son T, Chang SK, Lee M.In vitro assessment of clastogenicity of mobile-phone radiation (835 MHz) using the alkaline comet assay and chromosomal aberration test. Environ Toxicol 23:319-327, 2008. (VT, AE, GT, IX)

Recently we demonstrated that 835-MHz radiofrequency radiation electromagnetic fields (RF-EMF) neither affected the reverse mutation frequency nor accelerated DNA degradation in vitro. Here, two kinds of cytogenetic endpoints were further investigated on mammalian cells exposed to 835-MHz RF-EMF (the most widely used communication frequency band in Korean CDMA mobile phone networks) alone and in combination with model clastogens: in vitro alkaline comet assay and in vitro chromosome aberration (CA) test. No direct cytogenetic effect of 835-MHz RF-EMF was found in the in vitro CA test. The combined exposure of the cells to RF-EMF in the presence of ethylmethanesulfonate (EMS) revealed a weak and insignificant cytogenetic effect when compared to cells exposed to EMS alone in CA test. Also, the comet assay results to evaluate the ability of RF-EMF alone to damage DNA were nearly negative, although showing a small increase in tail moment. However, the applied RF-EMF had potentiation effect in comet assay when administered in combination with model clastogens (cyclophosphamide or 4-nitroquinoline 1-oxide). Thus, our results imply that we cannot confidently exclude any possibility of an increased risk of genetic damage, with important implications for the possible health effects of exposure to 835-MHz electromagnetic fields.

(NE) Kim HS, H-D Choi , J-K Pack, N Kim, Y H Ahn. Biological Effects of Exposure to a Radiofrequency Electromagnetic Field on the Placental Barrier in Pregnant Rats. Bioelectromagnetics 42(3):191-199, 2021. (VO, LE, GE)

The placenta protects the fetus against excessive stress-associated maternal cortisol during pregnancy. We studied whether exposure to radiofrequency electromagnetic field (RF-EMF) radiation during pregnancy can cause changes in dams and their placentas. Pregnant Sprague-Dawley rats were divided into cage-control, sham-exposed, and RF-exposed groups. They were exposed to RF-EMF signals at a whole-body specific absorption rate of 4 W/kg for 8 h/day from gestational Day 1 to 19. Levels of cortisol in the blood, adrenal gland, and placenta were measured by enzyme-linked immunosorbent assay. Levels of adrenocorticotropic hormone and corticotropin-releasing hormone were monitored in maternal blood. Expression levels of placental 11β-hydroxysteroid dehydrogenase type 2 (11β-HSD2) messenger RNA (mRNA) were measured by reverse transcription polymerase chain reaction. Morphological changes in the placenta were analyzed using hematoxylin and eosin staining. Fetal parts of the placenta were measured using Zen 2.3 blue edition software. Maternal cortisol in circulating blood (RF: 230 ± 24.6 ng/ml and Sham: 156 ± 8.3 ng/ml) and the adrenal gland (RF: 58.3 ± 4.5 ng/ml and Sham: 30 ± 3.8 ng/ml) was significantly increased in the RF-exposed group ($P < 0.05$). Placental cortisol was stably maintained, and the level of placental 11β-HSD2 mRNA expression was not changed in the RF-exposed group. RF-EMF exposure during pregnancy caused a significant elevation of cortisol levels in circulating blood; however, no changes in the placental barrier were observed in pregnant rats.

(E) Kim JH, Jeon S, Choi H-D, Lee J-H, Bae J-S, Kim N, Kim H-G, Kim K-B, Kim HR. Exposure to long-term evolution radiofrequency electromagnetic fields decreases neuroblastoma cell proliferation via Akt/mTOR-mediated cellular senescence. J Toxicol Environ Health A 84(20):846-857, 2021. (VT, LE, GE)

The aim of this study was to examine the potential effects of long-term evolution (LTE) radiofrequency electromagnetic fields (RF-EMF) on cell proliferation using SH-SY5Y neuronal cells. The growth rate and proliferation of SH-SY5Y cells were significantly decreased upon exposure to 1760 MHz RF-EMF at 4 W/kg specific absorption rate (SAR) for 4 hr/day for 4 days. Cell cycle analysis indicated that the cell cycle was delayed in the G0/G1 phase after RF-EMF exposure. However, DNA damage or apoptosis was not involved in the reduced cellular proliferation following RF-EMF exposure because the expression levels of histone H2A.X at Ser139 (γH2AX) were not markedly altered and the apoptotic pathway was not activated. However, SH-SY5Y cells exposed to RF-EMF exhibited a significant elevation in Akt and mTOR phosphorylation levels. In addition, the total amount of p53 and phosphorylated-p53 was significantly increased. Data suggested that Akt/mTOR-mediated cellular senescence led to p53 activation via stimulation of the mTOR pathway in SH-SY5Y cells. The transcriptional activation of p53 led to a rise in expression of cyclin-dependent kinase (CDK) inhibitors p21 and p27. Further, subsequent inhibition of CDK2 and CDK4 produced a fall in phosphorylated retinoblastoma (pRb at Ser807/811), which decreased cell proliferation. Taken together, these data suggest that exposure to RF-EMF might induce Akt/mTOR-mediated cellular senescence, which may delay the cell cycle without triggering DNA damage in SH-SY5Y neuroblastoma cells.

Kıvrak EG, Yurt KK, Kaplan AA, Alkan I, Altun G. Effects of electromagnetic fields exposure on the antioxidant defense system. J Microsc Ultrastruct. 5(4):167-176, 2017. (Review)

Technological devices have become essential components of daily life. However, their deleterious effects on the body, particularly on the nervous system, are well known. Electromagnetic fields (EMF) have various chemical effects, including causing deterioration in large molecules in cells and imbalance in ionic equilibrium. Despite being essential for life, oxygen molecules can lead to the generation of hazardous by-products, known as reactive oxygen species (ROS), during biological reactions. These reactive oxygen species can damage cellular components such as proteins, lipids and DNA. Antioxidant defense systems exist in order to keep free radical formation under control and to prevent their harmful effects on the biological system. Free radical formation can take place in various ways, including ultraviolet light, drugs, lipid oxidation, immunological reactions, radiation, stress, smoking, alcohol and biochemical redox reactions. Oxidative stress occurs if the antioxidant defense system is unable to prevent the harmful effects of free radicals. Several studies have reported that exposure to EMF results in oxidative stress in many tissues of the body. Exposure to EMF is known to increase free radical concentrations and traceability and can affect the radical couple recombination. The purpose of this review was to highlight the impact of oxidative stress on antioxidant systems.

Kocaman A, Altun G, Kaplan AA, Deniz ÖG, Yurt KK, Kaplan S. Genotoxic and carcinogenic effects of non-ionizing electromagnetic fields. Environ Res. 163:71-79, 2018. (review)

New technologies in electronics and communications are continually emerging. An increasing use of these electronic devices such as mobile phone, computer, wireless fidelity connectors or cellular towers is raising questions concerning whether they have an adverse effect on the body. Exposure to electromagnetic fields (EMF) is frequently suggested to have adverse health effects on humans and other organisms. This idea has been reported in many studies. In contrast, the therapeutic effects of EMF on different organs have also been reported. Research findings are inconsistent. This has given rise to very profound discrepancies. The duration and frequency of mobile phone calls and the association observed with various health effects has raised serious concerns due to the frequency with which these devices are used and the way they are held close to the head. The present review assesses the results of in vitro, in vivo, experimental, and epidemiological studies. The purpose of the study is to assess data concerning the carcinogenic and genotoxic effects of non-ionizing EMF. The major genotoxic and carcinogenic effects of EMF, divided into subsections as low frequency effects and radiofrequency effects, were reviewed. The inconsistent results between similar studies and the same research groups have made it very difficult to make any comprehensive interpretation. However, evaluation of current studies suggests that EMF may represent a serious source of concern and may be hazardous to living organisms.

(NE) Komatsubara Y, Hirose H, Sakurai T, Koyama S, Suzuki Y, Taki M, Miyakoshi J. Effect of high-frequency electromagnetic fields with a wide range of SARs on chromosomal

aberrations in murine m5S cells. Mutat Res. 587(1-2):114-119, 2005. (VT, AE, GT) (very high SAR)

To investigate the induction of chromosomal aberrations in mouse m5S cells after exposure to high-frequency electromagnetic fields (HFEMFs) at 2.45GHz, cells were exposed for 2h at average specific absorption rates (SARs) of 5, 10, 20, 50 and 100W/kg with continuous wave-form (CW), or at a mean SAR of 100W/kg (with a maximum of 900W/kg) with pulse wave-form (PW). The effects of HFEMF exposure were compared with those in sham-exposed controls and with mitomycin C (MMC) or X-ray treatment as positive controls. We examined all structural, chromatid-type and chromosome-type changes after HFEMF exposures and treatments with MMC and X-rays. No significant differences were observed following exposure to HFEMFs at SARs from 5 to 100W/kg CW and at a mean SAR of 100W/kg PW (a maximum SAR of 900W/kg) compared with sham-exposed controls, whereas treatments with MMC and X-rays increased the frequency of chromatid-type and chromosome-type aberrations. In summary, HFEMF exposures at 2.45GHz for 2h with up to 100W/kg SAR CW and an average 100W/kg PW (a maximum SAR of 900W/kg) do not induce chromosomal aberrations in m5S cells. Furthermore, there was no difference between exposures to CW and PW HFEMFs.

(E) Korenstein-Ilan A, Barbul A, Hasin P, Eliran A, Gover A, Korenstein R. Terahertz radiation increases genomic instability in human lymphocytes. Radiat Res 170(2):224-34, 2008. (VT, AE, GT)

Terahertz radiation is increasingly being applied in new and evolving technologies applied in areas such as homeland security and medical imaging. Thus a timely assessment of the potential hazards and health effects of occupational and general population exposure to THz radiation is required. We applied continuous-wave (CW) 0.1 THz radiation (0.031 mW/ cm(2)) to dividing lymphocytes for 1, 2 and 24 h and examined the changes in chromosome number of chromosomes 1, 10, 11 and 17 and changes in the replication timing of their centromeres using interphase fluorescence in situ hybridization (FISH). Chromosomes 11 and 17 were most vulnerable (about 30% increase in aneuploidy after 2 and 24 h of exposure), while chromosomes 1 and 10 were not affected. We observed changes in the asynchronous mode of replication of centromeres 11, 17 and 1 (by 40%) after 2 h of exposure and of all four centromeres after 24 h of exposure (by 50%). It is speculated that these effects are caused by radiation-induced low-frequency collective vibrational modes of proteins and DNA. Our results demonstrate that exposure of lymphocytes in vitro to a low power density of 0.1 THz radiation induces genomic instability. These findings, if verified, may suggest that such exposure may result in an increased risk of cancer.

(E) Koyama S, Nakahara T, Wake K, Taki M, IsozumiY, Miyakoshi J. Effects of high frequency electromagnetic fields on micronucleus formation in CHO-K1 cells. Mutat Res. 541: 81-89, 2003. (VT, AE, GT)

To investigate the effects of high frequency electromagnetic fields (HFEMFs), we assessed the frequency of micronucleus (MN) formation induced by chromosomal breakage or inhibition of spindles during cell division in Chinese hamster ovary (CHO)-K1 cells, using the cytokinesis

block micronucleus method. The MN frequency in cells in the inner, middle and outer wells of an annular culture plate was determined for the following four conditions: (1) CHO-K1 cells were exposed to a HFEMF for 18 h at average specific absorption rates (SARs) of 13, 39 and 50 W/kg with input power 7.8 W, and were compared with a sham-exposed control; (2) the cells were also exposed to a HFEMF at SARs of 78 and 100 W/kg with input power 13 W, and were compared with a sham-exposed control; (3) the cells were treated with bleomycin alone or with bleomycin followed by exposure to a HFEMF for 18 h at SARs of 25, 78 and 100 W/kg, and were compared with a bleomycin-treated positive control. The cells treated with bleomycin alone were compared with sham-exposed controls; and (4) As a high temperature control, CHO-K1 cells were incubated at 39 degrees C for 18 h. In study (1), the MN frequency of cells exposed to a HFEMF at a SAR of up to 50 W/kg was not different to that in sham-exposed cells. In study (2), there were statistically significant increases in the MN frequencies of cells in the middle and outer wells of the annular culture plate caused by exposure to a HFEMF at 100 and 78 W/kg, respectively. In study (3), the MN frequencies of cells in the middle (100 W/kg) and outer wells (78 W/kg) of the annular culture plate were statistically higher than that caused by bleomycin-treatment alone. In study (4), there was a statistically significant increase of MN frequency in the cells treated by heat at 39 degrees C. These results indicate that cells exposed to a HFEMF at a SAR of 78 W/kg and higher form MN more frequently than sham-exposed cells, while exposure to a HFEMF at up to 50 W/kg does not induce MN formation. In addition, a HFEMF at a SAR of 78 W/kg and higher may potentiate MN formation induced by bleomycin-treatment.

(E) Koyama S, Isozumi Y, Suzuki Y, Taki M, Miyakoshi J. Effects of 2.45-GHz electromagnetic fields with a wide range of SARs on micronucleus formation in CHO-K1 cells. ScientificWorldJournal 4 Suppl 2:29-40, 2004. (VT, AE, GT) (very high SAR)

There has been considerable discussion about the influence of high-frequency electromagnetic fields (HFEMF) on the human body. In particular, HFEMF used for mobile phones may be of great concern for human health. In order to investigate the properties of HFEMF, we have examined the effects of 2.45-GHz EMF on micronucleus (MN) formation in Chinese hamster ovary (CHO)-K1 cells. MN formation is induced by chromosomal breakage or inhibition of spindles during cell division and leads to cell damage. We also examined the influence of heat on MN formation, since HFEMF exposure causes a rise in temperature. CHO-K1 cells were exposed to HFEMF for 2 h at average specific absorption rates (SARs) of 5, 10, 20, 50, 100, and 200 W/kg, and the effects on these cells were compared with those in sham-exposed control cells. The cells were also treated with bleomycin alone as a positive control or with combined treatment of HFEMF exposure and bleomycin. Heat treatment was performed at temperatures of 37, 38, 39, 40, 41, and 42 degrees C. The MN frequency in cells exposed to HFEMF at a SAR of lower than 50 W/kg did not differ from the sham-exposed controls, while those at SARs of 100 and 200 W/kg were significantly higher when compared with the sham-exposed controls. There was no apparent combined effect of HFEMF exposure and bleomycin treatment. On heat treatment at temperatures from 38-42 degrees C, the MN frequency increased in a temperature-dependent manner. We also showed that an increase in SAR causes a rise in temperature and this may be connected to the increase in MN formation generated by exposure to HFEMF.

(NE) Koyama S, Narita E, Shimizu Y, Shiina T, Taki M, Shinohara N, et al. Twenty four-hour exposure to a 0.12 THz electromagnetic field does not affect the genotoxicity, morphological changes, or expression of heat shock protein in HCE-T cells. Int. J. Envrion. Health Res. Pub. Health. 13: 793, 1-9, 2016a. (VT, AE, GT)

To investigate the cellular effects of terahertz (THz) exposure, human corneal epithelial (HCE-T) cells derived from human eye were exposed to 0.12 THz radiation at 5 mW/cm² for 24 h, then the genotoxicity, morphological changes, and heat shock protein (Hsp) expression of the cells were examined. There was no statistically significant increase in the micronucleus (MN) frequency of cells exposed to 0.12 THz radiation compared with sham-exposed controls and incubator controls, whereas the MN frequency of cells treated with bleomycin for 1 h (positive control) did increase significantly. Similarly, there were no significant morphological changes in cells exposed to 0.12 THz radiation compared to sham-exposed controls and incubator controls, and Hsp expression (Hsp27, Hsp70, and Hsp90α) was also not significantly different between the three treatments. These results indicate that exposure to 0.12 THz radiation using the present conditions appears to have no or very little effect on MN formation, morphological changes, and Hsp expression in cells derived from human eye.

(NE) Koyama S, Narita E, Shimizu Y, Suzuki Y, Shiina T, Taki M, et al. Effects of long-term exposure to 60 GHz millileter-wavelength radiation on the genotoxicity and heat shock protein (Hsp) expression of cells derived from human eye. Int. J. Envrion. Health Res. Pub. Health. 13: 802, 1-9, 2016b. (VT, AE, GT)

Human corneal epithelial (HCE-T) and human lens epithelial (SRA01/04) cells derived from the human eye were exposed to 60 gigahertz (GHz) millimeter-wavelength radiation for 24 h. There was no statistically significant increase in the micronucleus (MN) frequency in cells exposed to 60 GHz millimeter-wavelength radiation at 1 mW/cm² compared with sham-exposed controls and incubator controls. The MN frequency of cells treated with bleomycin for 1 h provided positive controls. The comet assay, used to detect DNA strand breaks, and heat shock protein (Hsp) expression also showed no statistically significant effects of exposure. These results indicate that exposure to millimeter-wavelength radiation has no effect on genotoxicity in human eye cells.

(E) Kucukbagriacik Y, Dastouri M, Ozgur-Buyukatalay E, Akarca Dizakar OA, Yegin K. Investigation of oxidative damage, antioxidant balance, DNA repair genes, and apoptosis due to radiofrequency-induced adaptive response in mice. Electromagn Biol Med 41(4):389-401, 2022.(VO, LE, GE)

This study aims to determine whether exposure to non-ionizing radiofrequency fields could induce an adaptive response (AR) in adult mice and to reveal potential molecular mechanisms triggered by RF-induced AR. The study was performed on 24 adult male Swiss-Albino mice. The average mass of the mice was 37 g. Four groups of adult mice, each consisting of 6, were formed. The radiofrequency group (R) and the adaptive response group (RB) were exposed to 900 MHz of global system for mobile communications (GSM) signal at 0.339 W/kg (1 g average

specific absorption rate) 4 h/day for 7 days, while the control group (C) and the bleomycin group (B) were not exposed. 20 minutes after the last radiofrequency field (RF) exposure, the mice in the B and RB groups were injected intraperitoneal (ip) bleomycin (BLM), 37.5 mg/kg. All the animals were sacrificed 30 minutes after the BLM injection. Oxidative damage and antioxidant mechanism were subsequently investigated in the blood samples. Changes in the expression of the genes involved in DNA repair were detected in the liver tissue. TUNEL method was used to determine the apoptosis developed by DNA fragmentation in the liver tissue. The RB group, which produced an adaptive response, was compared with the control group. According to the results, the increase of reactive oxygen species (ROS) in the RB group may have played an important role in triggering the adaptive response and producing the required minimum stress level. Furthermore, tumor suppressor 53(p53), oxo guanine DNA glycosylase (OGG-1) levels responsible for DNA repair mechanism genes expression were increased in conjunction with the increase in ROS. The change in the poly (ADP-ribose) polymerase 1 (PARP-1) and glutathione peroxidase 1 (GPx-1) gene expression were not statistically significant. The antioxidant enzyme levels of superoxide dismutase (SOD), catalase (CAT), and total antioxidant capacity (TAC) were decreased in the group with adaptive response. According to the data obtained from terminal deoxynucleotidyl transferase dUTP nick end labeling (TUNEL) analysis, apoptosis was decreased in the RB group due to the decrease in cell death, which might have resulted from an increase in gene expression responsible for DNA repair mechanisms. The results of our study show that exposure to RF radiation may create a protective reaction against the bleomycin. The minimal oxidative stress due to the RF exposure leads to an adaptive response in the genes that play a role in the DNA repair mechanism and enzymes, enabling the survival of the cell.

(E) Kumar A, Kaur S, Chandel S, Singh HP, Batish DR, Kohli RK. Comparative cyto- and genotoxicity of 900 MHz and 1800 MHz electromagnetic field radiations in root meristems of Allium cepa. Ecotoxicol Environ Saf. 188:109786, 2020. (VO, AE, LI, GT, WS)

In the last few decades, tremendous increase in the use of wireless electronic gadgets, particularly the cell phones, has significantly enhanced the levels of electromagnetic field radiations (EMF-r) in the environment. Therefore, it is pertinent to study the effect of these radiations on biological systems including plants. We investigated comparative cytotoxic and DNA damaging effects of 900 and 1800 MHz EMF-r in Allium cepa (onion) root meristematic cells in terms of mitotic index (MI), chromosomal aberrations (CAs) and single cell gel electrophoresis (comet assay). Onion bulbs were subjected to 900 and 1800 MHz (at power densities 261 ± 8.50 mW m^{-2} and 332 ± 10.36 mW m^{-2}, respectively) of EMF-r for 0.5 h, 1 h, 2 h, and 4 h. Root length declined by 13.2% and 12.3%, whereas root thickness was increased by 46.7% and 48.3% after 4 h exposure to 900 MHz and 1800 MHz, respectively. Cytogenetic studies exhibited clastogenic effect of EMF-r as depicted by increased CAs and MI. MI increased by 36% and 53% after 2 and 4 h exposure to 900 MHz EMF-r, whereas it increased by 41% and 67% in response to 1800 MHz EMF-r. Aberration index was increased by 41%-266% and 14%-257% during 0.5-4 h of exposure to 900 MHz and 1800 MHz, respectively, over the control. EMF-r exposure decreased % head DNA (DNAH) and increased % tail DNA (DNAT) and olive tail moment (OTM) at both 900 and 1800 EMF-r. In 4 h exposure treatments, head DNA (%) declined by 19% and 23% at 900 MHz and 1800 MHz, respectively. DNAT and OTM

were increased by 2.3 and 3.7 fold upon exposure to 900 MHz EMF-r over that in the control, whereas 2.8 and 5.8 fold increase was observed in response to 1800 MHz EMF-r exposure for 4 h and the difference was statistically significant. The study concludes that EMF-r in the communication range (900 and 1800 MHz) adversely affect root meristems in plants and induce cytotoxic and DNA damage. EMF-r induced DNA damage was more pronounced at 1800 MHz than that at 900 MHz.

(NE) Kumar G, Wood AW, Anderson V, McIntosh RL, Chen YY, McKenzie RJ. Evaluation of Hematopoietic System Effects After in Vitro Radiofrequency Radiation Exposure in Rats. Int J Radiat Biol 87(2):231-240, 2011. (VT, AE, GT)

Purpose: This study was designed to investigate the effect of a 900-MHz continuous-wave (CW) radiofrequency radiation (RFR) exposure on the hematopoietic system in the rat. **Materials and methods:** Rat long bones (femur and tibia) were divided into two groups: Sham-exposed and radiofrequency (RF)-exposed. The mean Specific energy Absorption Rate (SAR) at 900-MHz averaged over the bone marrow (calculated by the finite-difference-time-domain (fdtD) method) was 2 W/kg at 16.7 W root mean square (rms) forward power into a Transverse Electromagnetic (TEM) cell. The bones, placed in a Petri dish containing media, were kept in the TEM cell for 30 min duration of sham or RF exposure. After exposure, the bone marrow cells were extracted and the following end points were tested: (a) Proliferation rate of whole bone marrow cells, (b) maturation rate of erythrocytes, (c) proliferation rate of lymphocytes, and (d) DNA damage (strand breaks/alkali labile sites) of lymphocytes. **Results:** Our data did not indicate any significant change in the proliferation rate of bone marrow cells and lymphocytes, erythrocyte maturation rate and DNA damage of lymphocytes. **Conclusion:** Our findings revealed no effect on the hematopoietic system in rats for 900 MHz CW RF exposure at the 2 W/kg localised SAR limit value recommended by the International Commission for Non-Ionising Radiation Protection (ICNIRP) for public exposures.

(NE) Kumar G, McIntosh RL, Anderson V, McKenzie RJ, Wood AW. A genotoxic analysis of the hematopoietic system after mobile phone type radiation exposure in rats. Int J Radiat Biol. 91(8):664-672, 2015. (VT, AE, GT)

PURPOSE: In our earlier study we reported that 900 MHz continuous wave (CW) radiofrequency radiation (RFR) exposure (2 W/kg specific absorption rate [SAR]) had no significant effect on the hematopoietic system of rats. In this paper we extend the scope of the previous study by testing for possible effects at: (i) different SAR levels; (ii) both 900 and 1800 MHz, and; (iii) both CW and pulse modulated (PM) RFR. MATERIALS AND METHODS: Excised long bones from rats were placed in medium and RFR exposed in (i) a Transverse Electromagnetic (TEM) cell or (ii) a waveguide. Finite-difference time-domain (FDTD) numerical analyses were used to estimate forward power needed to produce nominal SAR levels of 2/10 and 2.5/12.4 W/kg in the bone marrow. After exposure, the lymphoblasts were extracted and assayed for proliferation rate, and genotoxicity. RESULTS: Our data did not indicate any significant change in these end points for any combination of CW/PM exposure at 900/1800 MHz at SAR levels of nominally 2/10 W/kg or 2.5/12.4 W/kg. CONCLUSIONS: No significant changes were observed in the

hematopoietic system of rats after the exposure of CW/PM wave 900 MHz/1800 MHz RF radiations at different SAR values.

(E) Kumar, R , Deshmukh, P.S. , Sharma, S., Banerjee, B.D. Effect of mobile phone signal radiation on epigenetic modulation in the hippocampus of Wistar rat. Environ Res 192:110297, 2021. (VO, CE, GE, LI, EP)

Exponential increase in mobile phone uses, given rise to public concern regarding the alleged deleterious health hazards as a consequence of prolonged exposure. In 2018, the U.S. National toxicology program reported, two year toxicological studies for potential health hazards from exposure to cell phone radiations. Epigenetic modulations play a critical regulatory role in many cellular functions and pathological conditions. In this study, we assessed the dose-dependent and frequency-dependent epigenetic modulation (DNA and Histone methylation) in the hippocampus of Wistar rats. A Total of 96 male Wistar rats were segregated into 12 groups exposed to 900 MHz, 1800 MHz and 2450 MHz RF-MW at a specific absorption rate (SAR) of 5.84×10^{-4} W/kg, 5.94×10^{-4} W/kg and 6.4×10^{-4} W/kg respectively for 2 h per day for 1-month, 3-month and 6-month periods. At the end of the exposure duration, animals were sacrificed to collect the hippocampus. Global hippocampal DNA methylation and histone methylation were estimated by ELISA. However, DNA methylating enzymes, DNA methyltransferase1 (DNMT1) and histone methylating enzymes euchromatic histone methylthransferase1 (EHMT1) expression was evaluated by real-time PCR, as well as further validated with Western blot. Alteration in epigenetic modulation was observed in the hippocampus. Global DNA methylation was decreased and histone methylation was increased in the hippocampus. We observed that microwave exposure led to significant epigenetic modulations in the hippocampus with increasing frequency and duration of exposure. Microwave exposure with increasing frequency and exposure duration brings significant ($p < 0.05$) epigenetic modulations which alters gene expression in the hippocampus.

(E) Kumar S, Kesari KK, Behari J. Evaluation of genotoxic effects in male Wistar rats following microwave exposure. Indian J Exp Biol 48:586-592, 2010. (VO, LE, LI, GT, OX)

Wistar rats (70 days old) were exposed for 2 h a day for 45 days continuously at 10 GHz [power density 0.214 mW/cm2, specific absorption rate (SAR) 0.014 W/kg] and 50 GHz (power density 0.86 microW/cm2, SAR 8.0 x10(-4) W/kg). Micronuclei (MN), reactive oxygen species (ROS), and antioxidant enzymes activity were estimated in the blood cells and serum. These radiations induce micronuclei formation and significant increase in ROS production. Significant changes in the level of serum glutathione peroxidase, superoxide dismutase and catalase were observed in exposed group as compared with control group. It is concluded that microwave exposure can be affective at genetic level. This may be an indication of tumor promotion, which comes through the overproduction of reactive oxygen species.

(E) Kumar S, Behari J, Sisodia R. Influence of electromagnetic fields on reproductive system of male rats. Int J Radiat Biol. 89: 147-154, 2013. (VO, LE, GT, RP, LI)

Purpose: Reports of declining male fertility have renewed interest in the role of environmental and occupational exposures in the etiology of human infertility. The aim of the present work was

to investigate the effect of 10 GHz exposure on the male Wistar rat's reproductive system and to find out the possible causative factors. **Materials and methods:** The study was divided into sham-exposed and exposed groups. Seventy day-old rats were exposed to 10 GHz microwave radiation for 2 h per day for 45 days at power density 0.21 mW/cm(2) and specific absorption rate (SAR) of 0.014 W/kg. After the end of the experiment, blood samples were collected for the estimation of in vivo chromosomal aberration damage and micronucleus test. Spermatozoa were taken out for estimation of Caspase-3, comet assay, testosterone and electron microscopy and compared with sham-exposed. **Results:** The study of scanning electron microscopic revealed shrinkage of the lumen of the seminiferous tubules. Apoptotic bodies were found in exposed group. A flow cytometry examination showed formation of micronuclei body in lymphocytes of exposed group. Comet assay confirmed DNA (deoxyribonucleic acid) strand break. Testosterone level was found significantly decreased with the shrinkage of testicular size. **Conclusions:** 10 GHz field has an injurious effect on fertility potential of male-exposed animals.

(E) Kumar S, Nirala JP, Behari J, Paulraj R. Effect of electromagnetic irradiation produced by 3G mobile phone on male rat reproductive system in a simulated scenario. Indian J Exp Biol. 52(9):890-897, 2014. (VO, LE, LI, GT, OX, RP)

Reports of declining male fertility have renewed interest in assessing the role of electromagnetic fields (EMFs). Testicular function is particularly susceptible to the radiation emitted by EMFs. Significant decrease in sperm count, increase in the lipid peroxidation damage in sperm cells, reduction in seminiferous tubules and testicular weight and DNA damage were observed following exposure to EMF in male albino rats. The results suggest that mobile phone exposure adversely affects male fertility.

(E) Kundu A, Vangaru S, Bhowmick S, Bhattacharyya S, Mallick AI, Gupta B. One-time Electromagnetic Irradiation Modifies Stress-sensitive Gene Expressions in Rice Plant. Bioelectromagnetics 42(8):649-658, 2021.(VO, AE, GE)

Electromagnetic energy is utilized over multiple frequency bands to provide seamless wireless communication services. Plants can well perceive electromagnetic energy present in open environment due to reasonably high permittivity and electrical conductivity of constituent tissues. Moreover, higher surface-to-volume ratio of plant structure facilitates increased interaction with the incident electromagnetic waves. To date, a few well-designed studies have been conducted inside controlled electromagnetic reverberation chambers to investigate either short duration-low amplitude or long duration-periodic electromagnetic irradiation-induced molecular responses in plants. However, as far as is known, studies investigating molecular responses particularly at the mid-vegetative stage in plants following one-time (hours-long) electromagnetic irradiation have not been reported earlier. Hence, the present study aimed at investigating molecular responses in 40-day-old Swarnaprabha rice plants following one-time 1837.50 MHz, 2.75 mW/m2 electromagnetic irradiation of 2 h 30 min duration. Controlled electromagnetic irradiation inside a simple reverberation chamber was ensured to achieve pure electromagnetic environment at 1837.50 MHz with deterministic electromagnetic power density at selected position. Swarnaprabha rice plant was chosen for this investigation since the rice variety is widely cultivated and consumed in the Indian subcontinent. Subsequent alterations in some selected stress-sensitive gene expressions were assayed using real-time quantitative

polymerase chain reaction technique-significant upregulation in calmodulin and phytochrome B gene expressions were noted. This investigation was purposefully focused on subsequent molecular responses immediately following electromagnetic irradiation so that the possible effects of secondary stimulations could be avoided. Observed molecular responses strongly suggested that plants perceive 1837.50 MHz, 2.75 mW/m2 electromagnetic irradiation similar to other injurious stimuli.

(NE) Kuribayashi M, Wang J, Fujiwara O, Doi Y, Nabae K, Tamano S, Ogiso T, Asamoto M, Shirai T. Lack of effects of 1439 MHz electromagnetic near field exposure on the blood-brain barrier in immature and young rats. Bioelectromagnetics. 26(7):578-88, 2005. (VO, LE, GE)

Possible effects of 1439 MHz electromagnetic near field (EMF) exposure on the blood-brain barrier (BBB) were investigated using immature (4 weeks old) and young (10 weeks old) rats, equivalent in age to the time when the BBB development is completed and the young adult, respectively. Alteration of BBB related genes, such as those encoding p-glycoprotein, aquaporin-4, and claudin-5, was assessed at the protein and mRNA levels in the brain after local exposure of the head to EMF at 0, 2, and 6 W/kg specific energy absorption rates (SARs) for 90 min/day for 1 or 2 weeks. Although expression of the 3 genes was clearly decreased after administration of 1,3-dinitrobenzene (DNB) as a positive control, when compared with the control values, there were no pathologically relevant differences with the EMF at any exposure levels at either age. Vascular permeability, monitored with reference to transfer of FITC-dextran, FD20, was not affected by EMF exposure. Thus, these findings suggest that local exposure of the head to 1439 MHz EMF exerts no adverse effects on the BBB in immature and young rats.

(NE) Lagroye I, Anane R, Wettring BA, Moros EG, Straube WL, Laregina M, Niehoff M, Pickard WF, Baty J, Roti JL. Measurement of DNA damage after acute exposure to pulsed-wave 2450 MHz microwaves in rat brain cells by two alkaline comet assay methods. Int J Radiat Biol. 80(1):11-20, 2004a. (VO, AE, GT)

Purpose: To investigate the effect of 2450 MHz pulsed-wave microwaves on the induction of DNA damage in brain cells of exposed rats and to discover whether proteinase K is needed to detect DNA damage in the brain cells of rats exposed to 2450 MHz microwaves. Materials and methods: Sprague-Dawley rats were exposed to 2450 MHz pulsed-wave microwaves and sacrificed 4 h after a 2-h exposure. Rats irradiated whole-body with 1 Gy (137)Cs were included as positive controls. DNA damage was assayed by two variants of the alkaline comet assay on separate aliquots of the same cell preparation. Results: Significant DNA damage was observed in the rat brain cells of rats exposed to gamma-rays using both versions of the alkaline comet assay independent of the presence or absence of proteinase K. However, neither version of the assay could detect any difference in comet length and/or normalized comet moment between sham- and 2450 MHz pulsed-wave microwave-exposed rats, regardless of the inclusion or omission of proteinase K in the comet assay. Conclusions: No DNA damage in brain cells was detected following exposure of rats to 2450 MHz microwaves pulsed-wave at a specific

absorption rate of 1.2 W kg(-1) regardless of whether or not proteinase K was included in the assay. Thus, the results support the conclusion that low-level 2450 MHz pulsed-wave microwave exposures do not induce DNA damage detectable by the alkaline comet assay.

(NE) Lagroye I, Hook GJ, Wettring BA, Baty JD, Moros EG, Straube WL, Roti Roti JL. Measurements of alkali-labile DNA damage and protein-DNA crosslinks after 2450 MHz microwave and low-dose gamma irradiation In vitro. Radiat Res. 161(2): 201-214, 2004b. (VT, AE. GT)

In vitro experiments were performed to determine whether 2450 MHz microwave radiation induces alkali-labile DNA damage and/or DNA-protein or DNA-DNA crosslinks in C3H 10T(1/2) cells. After a 2-h exposure to either 2450 MHz continuous-wave (CW) microwaves at an SAR of 1.9 W/kg or 1 mM cisplatinum (CDDP, a positive control for DNA crosslinks), C3H 10T(1/2) cells were irradiated with 4 Gy of gamma rays ((137)Cs). Immediately after gamma irradiation, the single-cell gel electrophoresis assay was performed to detect DNA damage. For each exposure condition, one set of samples was treated with proteinase K (1 mg/ml) to remove any possible DNA-protein crosslinks. To measure DNA-protein crosslinks independent of DNA-DNA crosslinks, we quantified the proteins that were recovered with DNA after microwave exposure, using CDDP and gamma irradiation, positive controls for DNA-protein crosslinks. Ionizing radiation (4 Gy) induced significant DNA damage. However, no DNA damage could be detected after exposure to 2450 MHz CW microwaves alone. The crosslinking agent CDDP significantly reduced both the comet length and the normalized comet moment in C3H 10T(1/2) cells irradiated with 4 Gy gamma rays. In contrast, 2450 MHz microwaves did not impede the DNA migration induced by gamma rays. When control cells were treated with proteinase K, both parameters increased in the absence of any DNA damage. However, no additional effect of proteinase K was seen in samples exposed to 2450 MHz microwaves or in samples treated with the combination of microwaves and radiation. On the other hand, proteinase K treatment was ineffective in restoring any migration of the DNA in cells pretreated with CDDP and irradiated with gamma rays. When DNA-protein crosslinks were specifically measured, we found no evidence for the induction of DNA-protein crosslinks or changes in amount of the protein associated with DNA by 2450 MHz CW microwave exposure. Thus 2-h exposures to 1.9 W/ kg of 2450 MHz CW microwaves did not induce measurable alkali-labile DNA damage or DNA-DNA or DNA-protein crosslinks.

(E) Lai H, Singh NP, Acute low-intensity microwave exposure increases DNA single-strand breaks in rat brain cells. Bioelectromagnetics 16(3):207-210, 1995. (VO, AE, GT)

Levels of DNA single-strand break were assayed in brain cells from rats acutely exposed to low-intensity 2450 MHz microwaves using an alkaline microgel electrophoresis method. Immediately after 2 h of exposure to pulsed (2 microseconds width, 500 pulses/s) microwaves, no significant effect was observed, whereas a dose rate-dependent [0.6 and 1.2 W/kg whole body specific absorption rate (SAR)] increase in DNA single-strand breaks was found in brain cells of rats at 4 h postexposure. Furthermore, in rats exposed for 2 h to continuous-wave 2450 MHz microwaves (SAR 1.2 W/kg), increases in brain cell DNA single-strand breaks were

observed immediately as well as at 4 h postexposure.

(E) Lai H, Singh NP, Single- and double-strand DNA breaks in rat brain cells after acute exposure to radiofrequency electromagnetic radiation. Int J Radiat Biol 69(4):513-521, 1996. (VO, AE, GT)

We investigated the effects of acute (2-h) exposure to pulsed (2-micros pulse width, 500 pulses s(-1)) and continuous wave 2450-MHz radiofrequency electromagnetic radiation on DNA strand breaks in brain cells of rat. The spatial averaged power density of the radiation was 2mW/cm2, which produced a whole-body average-specific absorption rate of 1.2W/kg. Single- and double-strand DNA breaks in individual brain cells were measured at 4h post-exposure using a microgel electrophoresis assay. An increase in both types of DNA strand breaks was observed after exposure to either the pulsed or continuous-wave radiation, No significant difference was observed between the effects of the two forms of radiation. We speculate that these effects could result from a direct effect of radiofrequency electromagnetic energy on DNA molecules and/or impairment of DNA-damage repair mechanisms in brain cells. Our data further support the results of earlier in vitro and in vivo studies showing effects of radiofrequency electromagnetic radiation on DNA.

(E) Lai, H, Singh, NP, Melatonin and a spin-trap compound block radiofrequency electromagnetic radiation-induced DNA strand breaks in rat brain cells. Bioelectromagnetics 18(6):446-454, 1997. (VO, AE, GT, OX)

Effects of in vivo microwave exposure on DNA strand breaks, a form of DNA damage, were investigated in rat brain cells. In previous research, we have found that acute (2 hours) exposure to pulsed (2 microseconds pulses, 500 pps) 2450-MHz radiofrequency electromagnetic radiation (RFR) (power density 2 mW/cm2, average whole body specific absorption rate 1.2 W/kg) caused an increase in DNA single- and double-strand breaks in brain cells of the rat when assayed 4 hours post exposure using a microgel electrophoresis assay. In the present study, we found that treatment of rats immediately before and after RFR exposure with either melatonin (1 mg/kg/injection, SC) or the spin-trap compound N-tert-butyl-alpha-phenylnitrone (PBN) (100 mg/kg/injection, i.p.) blocks this effects of RFR. Since both melatonin and PBN are efficient free radical scavengers it is hypothesized that free radicals are involved in RFR-induced DNA damage in the brain cells of rats. Since cumulated DNA strand breaks in brain cells can lead to neurodegenerative diseases and cancer and an excess of free radicals in cells has been suggested to be the cause of various human diseases, data from this study could have important implications for the health effects of RFR exposure.

(E) Lai H, Singh NP, Interaction of microwaves and a temporally incoherent magnetic field on single and double DNA strand breaks in rat brain cells. Electromag Biol Med 24:23-29, 2005. (VO, AE, GT, IX)

The effect of a temporally incoherent magnetic field ('noise') on microwave-induced DNA single and double strand breaks in rat brain cells was investigated. Four treatment groups of rats were studied: microwave-exposure (continuous-wave 2450-MHz microwaves, power density 1 mW/cm^2, average whole body specific absorption rate of 0.6 W/kg), 'noise'-exposure (45 mG),

'microwave + noise'-exposure, and sham-exposure. Animals were exposed to these conditions for 2 hrs. DNA single and double strand breaks in brain cells of these animals were assayed 4 hrs later using a microgel electrophoresis assay. Results show that brain cells of microwave-exposed rats had significantly higher levels of DNA single and double strand breaks when compared with sham-exposed animals. Exposure to 'noise' alone did not significantly affect the levels (i.e., they were similar to those of the sham-exposed rats). However, simultaneous 'noise' exposure blocked microwave-induced increases in DNA strand breaks. These data indicate that simultaneous exposure to a temporally incoherent magnetic field could block microwave-induced DNA damage in brain cells of the rat.

(E) Lai, H, Carino, MA, Singh, NP, Naltrexone blocks RFR-induced DNA double strand breaks in rat brain cells. Wireless Networks 3:471-476, 1997. (VO, AE, GT)

Previous research in our laboratory has shown that various effects of radiofrequency electromagnetic radiation (RFR) exposure on the nervous system are mediated by endogenous opioids in the brain. We have also found that acute exposure to RFR induced DNA strand breaks in brain cells of the rat. The present experiment was carried out to investigate whether endogenous opioids are also involved in RFR-induced DNA strand breaks. Rats were treated with the opioid antagonist naltrexone (1 mg/kg, IP) immediately before and after exposure to 2450-MHz pulsed (2 s pulses, 500 pps) RFR at a power density of 2 mW/cm2 (average whole body specific absorption rate of 1.2 W/kg) for 2 hours. DNA double strand breaks were assayed in brain cells at 4 hours after exposure using a microgel electrophoresis assay. Results showed that the RFR exposure significantly increased DNA double strand breaks in brain cells of the rat, and the effect was partially blocked by treatment with naltrexone. Thus, these data indicate that endogenous opioids play a mediating role in RFR-induced DNA strand breaks in brain cells of the rat.

(E) Lakshmi NK, Tiwari R, Bhargava SC, Ahuja YR. Investigations on DNA damage and frequency of micronuclei in occupational exposure to electromagnetic fields (EMFs) emitted from video display terminals (VDTs). Gen Mol Biol 33, 154-158, 2010. (HU, LE, GT)

The potential effect of electromagnetic fields (EMFs) emitted from video display terminals (VDTs) to elicit biological response is a major concern for the public. The software professionals are subjected to cumulative EMFs in their occupational environments. This study was undertaken to evaluate DNA damage and incidences of micronuclei in such professionals. To the best of our knowledge, the present study is the first attempt to carry out cytogenetic investigations on assessing bioeffects in personal computer users. The study subjects (n = 138) included software professionals using VDTs for more than 2 years with age, gender, socioeconomic status matched controls (n = 151). DNA damage and frequency of micronuclei were evaluated using alkaline comet assay and cytochalasin blocked micronucleus assay respectively. Overall DNA damage and incidence of micronuclei showed no significant differences between the exposed and control subjects. With exposure characteristics, such as total duration (years) and frequency of use (minutes/day) sub-groups were assessed for such parameters. Although cumulative frequency of use showed no significant changes in the DNA

integrity of the classified sub-groups, the long-term users (> 10 years) showed higher induction of DNA damage and increased frequency of micronuclei and micro nucleated cells.

(E) Lameth J, Arnaud-Cormos D, Lévêque P, Boillée S, Edeline JM, Mallat M. Effects of a single head exposure to GSM-1800 MHz signals on the transcriptome profile in the rat cerebral cortex: enhanced gene responses under proinflammatory conditions. Neurotox Res. 2020 Mar 21. doi: 10.1007/s12640-020-00191-3. [Epub ahead of print] (VO, AE, GE, IX)

Mobile communications are propagated by electromagnetic fields (EMFs), and since the 1990s, they operate with pulse-modulated signals such as the GSM-1800 MHz. The biological effects of GSM-EMF in humans affected by neuropathological processes remain seldom investigated. In this study, a 2-h head-only exposure to GSM-1800 MHz was applied to (i) rats undergoing an acute neuroinflammation triggered by a lipopolysaccharide (LPS) treatment, (ii) age-matched healthy rats, or (iii) transgenic hSOD1^{G93A} rats that modeled a presymptomatic phase of human amyotrophic lateral sclerosis (ALS). Gene responses were assessed 24 h after the GSM head-only exposure in a motor area of the cerebral cortex (mCx) where the mean specific absorption rate (SAR) was estimated to be 3.22 W/kg. In LPS-treated rats, a genome-wide mRNA profiling was performed by RNA-seq analysis and revealed significant (adjusted p value < 0.05) but moderate (fold changes < 2) upregulations or downregulations affecting 2.7% of the expressed genes, including genes expressed predominantly in neuronal or in glial cell types and groups of genes involved in protein ubiquitination or dephosphorylation. Reverse transcription-quantitative PCR analyses confirmed gene modulations uncovered by RNA-seq data and showed that in a set of 15 PCR-assessed genes, significant gene responses to GSM-1800 MHz depended upon the acute neuroinflammatory state triggered in LPS-treated rats, because they were not observed in healthy or in hSOD1^{G93A} rats. Together, our data specify the extent of cortical gene modulations triggered by GSM-EMF in the course of an acute neuroinflammation and indicate that GSM-induced gene responses can differ according to pathologies affecting the CNS.

(NE) Lamkowski A, Kreitlow M, Radunz J, Willenbockel M, Sabath F, Schuhn W, Stiemer M, Fichte LO, Dudzinski M, Böhmelt S, Ullmann R, Majewski M, Franchini V, Eder S, Rump A, Port M, Abend M. Gene expression analysis in human peripheral blood cells after 900 MHz RF-EMF short-term exposure. Radiat Res. 189(5):529-540, 2018. (VT, AE, GE)

Radiofrequency electromagnetic fields (RF-EMF) are a basic requirement of modern wireless communication technology. Statutory thresholds of RF-EMF are established to limit relevant additional heat supply in human tissue. Nevertheless, to date, questions concerning nonthermal biological effects have yet to be fully addressed. New versions of microarrays (8 × 60K v2) provide a higher resolution of whole genome gene expression to display adaptive processes in cells after irradiation. In this ex vivo/ in vitro study, we irradiated peripheral blood cells from five donors with a continuous wave of 900 MHz RF-EMF for 0, 30, 60 and 90 min. Gene expression changes ($P \leq 0.05$ and ≥twofold differences above or below the room temperature control exposed samples) were evaluated with microarray analysis. The results were compared with data from

room temperature + 2°C samples. Verification of microarray results was performed using bioinformatic analyses and qRT-PCR. We registered a lack of an EMF-specific gene expression response after applying the false discovery rate adjustment (FDR), using a high-stringency approach. Low-stringency analysis revealed 483 statistically significant deregulated transcripts in all RF-EMF groups relative to the room temperature exposed samples without an association with their corresponding room temperature + 2°C controls. Nevertheless, these transcripts must be regarded as statistical artefacts due to the absence of a targeted biological response, including enrichment and network analyses administered to microarray expressed gene subset profiles. Correspondingly, 14 most promising candidate transcripts examined by qRT-PCR displayed an absence of correlation with respect to the microarray results. In conclusion, these findings indicate that 900 MHz EMF exposure establishing an average specific absorption rate of 9.3 W/kg to whole blood cells is insufficient to induce nonthermal effects in gene expression during short-time exposure up to 90 min.

(NE) Lamkowski A, Kreitlow M, Radunz J, Willenbockel M, Stiemer M, Fichte LO, Rädel CF, Majewski M, Ostheim P, Port M, Abend M. Analyzing the impact of 900 MHz EMF short-term exposure to the expression of 667 miRNAs in human peripheral blood cells. Sci Rep. 11(1):4444, 2021. (VT, AE, GE)

More than ever before, people around the world are frequently exposed to different sections of the electromagnetic spectrum, mainly emitted from wireless modern communication technologies. Especially, the level of knowledge on non-thermal biological EMF effects remains controversial. New technologies allow for a more detailed detection of non-coding RNAs which affect the post-transcriptional control. Such method shall be applied in this work to investigate the response of human blood cells to electromagnetic irradiation. In this ex vivo in vitro study, we exposed peripheral blood cells from 5 male donors to a continuous wave of 900 MHz EMF for 0, 30, 60 and 90 min. Significant micro RNA (miRNA) expression changes ($p \leq 0.05$) above or below the SHAM exposed samples were evaluated using a quantitative real time PCR platform for simultaneous detection of 667 miRNAs called low density array. Only significant miRNA expression changes which were detectable in at least 60% of the samples per exposure group were analyzed. The results were compared with data from room temperature + 2 °C (RT + 2 °C) samples (here referred to as hyperthermia) to exclude miRNA expression altered by hyperthermia. The validation study by using the same donors and study design was performed after an interval of 2 years. When analyzing a total of 667 miRNAs during the screening study, 2 promising candidate miRNAs were identified, which were down regulated almost twice and showed a complete separation from the unexposed control group (miR-194 at 30 min and miR-939 at 60 min). The p-values even survived the Bonferroni correction for multiple comparisons ($p = 0.0007$ and $p = 0.004$, respectively). None of these miRNAs were expressed at a second time point after EMF exposure. Following an alternative analysis approach, we examined for miRNAs revealing an expected significant association of differential miRNA expression with the dose-time EMF exposure product, separately for each donor. Donors 2 and 3 revealed 11 and 10 miRNA species being significantly associated with EMF exposure which differed significantly from the other donors showing a minor number of differentially expressed miRNAs and could identify donors 2 and 3 as particularly EMF-responsive. The measurements were repeated after 2

years. The number of expressed/non-expressed miRNAs was almost similar (97.4%), but neither the number nor the previously differentially expressed miRNAs could be reproduced. Our data neither support evidence of early changes at miRNA expression level in human whole blood cells after 900 MHz EMF exposure nor the identification of EMF-responsive individuals.

(E) Lawler NB, Evans CW, Romanenko S, Chaudhari N, Fear M, Wood F, Smith NM, Wallace VP, Swaminathan Iyer K. Millimeter waves alter DNA secondary structures and modulate the transcriptome in human fibroblasts. Biomed Opt Express. 2022 Apr 28;13(5):3131-3144, 2022. (VT, LE, GT)

As millimetre wave (MMW) frequencies of the electromagnetic spectrum are increasingly adopted in modern technologies such as mobile communications and networking, characterising the biological effects is critical in determining safe exposure levels. We study the exposure of primary human dermal fibroblasts to MMWs, finding MMWs trigger genomic and transcriptomic alterations. In particular, repeated 60 GHz, 2.6 mW cm^{-2}, 46.8 J cm^{-2} d^{-1} MMW doses induce a unique physiological response after 2 and 4 days exposure. We show that high dose MMWs induce simultaneous non-thermal alterations to the transcriptome and DNA structural dynamics, including formation of G-quadruplex and i-motif secondary structures, but not DNA damage.

(E) Le Quément C, Nicolas Nicolaz C, Zhadobov M, Desmots F, Sauleau R, Aubry M, Michel D, Le Dréan Y. Whole-genome expression analysis in primary human keratinocyte cell cultures exposed to 60 GHz radiation. Bioelectromagnetics 33(2):147-158, 2012.(VT, AE, GE)

The main purpose of this study is to investigate potential responses of skin cells to millimeter wave (MMW) radiation increasingly used in the wireless technologies. Primary human skin cells were exposed for 1, 6, or 24 h to 60.4 GHz with an average incident power density of 1.8 mW/cm(2) and an average specific absorption rate of 42.4 W/kg. A large-scale analysis was performed to determine whether these exposures could affect the gene expression. Gene expression microarrays containing over 41,000 unique human transcript probe sets were used, and data obtained for sham and exposed cells were compared. No significant difference in gene expression was observed when gene expression values were subjected to a stringent statistical analysis such as the Benjamini-Hochberg procedure. However, when a t-test was employed to analyze microarray data, 130 transcripts were found to be potentially modulated after exposure. To further quantitatively analyze these preselected transcripts, real-time PCR was performed on 24 genes with the best combination of high fold change and low P-value. Five of them, namely CRIP2, PLXND1, PTX3, SERPINF1, and TRPV2, were confirmed as differentially expressed after 6 h of exposure. To the best of our knowledge, this is the first large-scale study reporting on potential gene expression modification associated with MMW radiation used in wireless communication applications.

(E) Lee K-S, Choi J-S, Hong S-Y, Son T-H, Yu K. Mobile phone electromagnetic radiation activates MAPK signaling and regulates viability in Drosophila. Bioelectromagnetics 29(5):371-379, 2008. (VO, AE, GE)

Mobile phones are widely used in the modern world. However, biological effects of electromagnetic radiation produced by mobile phones are largely unknown. In this report, we show biological effects of the mobile phone 835 MHz electromagnetic field (EMF) in the Drosophila model system. When flies were exposed to the specific absorption rate (SAR) 1.6 W/kg, which is the proposed exposure limit by the American National Standards Institute (ANSI), more than 90% of the flies were viable even after the 30 h exposure. However, in the SAR 4.0 W/kg strong EMF exposure, viability dropped from the 12 h exposure. These EMF exposures triggered stress response and increased the production of reactive oxygen species. The EMF exposures also activated extracellular signal regulated kinase (ERK) and c-Jun N-terminal kinase (JNK) signaling, but not p38 kinase signaling. Interestingly, SAR 1.6 W/kg activated mainly ERK signaling and expression of an anti-apoptotic gene, whereas SAR 4.0 W/kg strongly activated JNK signaling and expression of apoptotic genes. In addition, SAR 4.0 W/kg amplified the number of apoptotic cells in the fly brain. These findings demonstrate that the exposure limit on electromagnetic radiation proposed by ANSI triggered ERK-survival signaling but the strong electromagnetic radiation activated JNK-apoptotic signaling in Drosophila.

(E) Lee S, Johnson D, Dunbar K, Hui Dong, Xijin Ge, Yeong C Kim, Claudia Wing, Nimanthi Jayathilaka, Nimmi Emmanuel, Chenn Q Zhou, Howard L Gerber, Charles C Tseng, San Ming Wang. 2.45 GHz radiofrequency fields alter gene expression in cultured human cells. FEBS Lett 579(21):4829-4836, 2005. (VT, AE, GE)

The biological effect of radiofrequency (RF) fields remains controversial. We address this issue by examining whether RF fields can cause changes in gene expression. We used the pulsed RF fields at a frequency of 2.45 GHz that is commonly used in telecommunication to expose cultured human HL-60 cells. We used the serial analysis of gene expression (SAGE) method to measure the RF effect on gene expression at the genome level. We observed that 221 genes altered their expression after a 2-h exposure. The number of affected genes increased to 759 after a 6-h exposure. Functional classification of the affected genes reveals that apoptosis-related genes were among the upregulated ones and the cell cycle genes among the downregulated ones. We observed no significant increase in the expression of heat shock genes. These results indicate that the RF fields at 2.45 GHz can alter gene expression in cultured human cells through non-thermal mechanism.

(NE) Lerchl A, Klose M, Drees K. No Increased DNA Damage Observed in the Brain, Liver, and Lung of Fetal Mice Treated With Ethylnitrosourea and Exposed to UMTS Radiofrequency Electromagnetic Fields. Bioelectromagnetics 41(8):611-616, 2020. (VO, LE, GT, IX)

The widespread use of mobile phones and Wi-Fi-based communication devices makes exposure to radiofrequency electromagnetic fields (RF-EMF) unavoidable. Previous experiments have revealed the tumor-promoting effects of non-ionizing RF-EMF in adult carcinogen-treated mice in utero. To extend these investigations, we tested whether these effects are due to the co-carcinogenicity of RF-EMF which would manifest as elevated DNA damage. Similar to previous experiments, pregnant mice were exposed to RF-EMF (Universal Mobile Telecommunication System [UMTS] standard, approximately 1,960 MHz) from day 7 post-conception (p.c.) at 0 (sham), 0.04, and 0.4 W/kg SAR. At day 14 p.c., the mice were injected with the carcinogen

ethylnitrosourea (ENU, 40 mg/kg). At three time-points specifically 24, 36, and 72 h later, the pregnant females were sacrificed and the fetuses (n = 24-57) were removed. A dye (cy3) specific for adenyl adducts was used to detect DNA damage by fluorescence microscopy in the brain, liver, and lung of each fetus. Compared to control (0 W/kg SAR), exposure to RF-EMF had no effect on the formation of DNA adducts in the inspected tissues. We conclude that increased adenyl formation of DNA by RF-EMF exposure is not a valid explanation for the previously reported tumor-promoting effects of RF-RMF. Our findings may help to gain a deeper insight into the biological effects of RF-EMF exposure in the context of malignancy.

(E) Li H, Yu G, Yong Z, Qiao S, Zhi W, Ma L, Xu X, Zhao X, Zhang J, Wang L, Hu X. Associations Between a Polymorphism in the Rat 5-HT1A Receptor Gene Promoter Region (rs198585630) and Cognitive Alterations Induced by Microwave Exposure. Front Pub Health. 10:802386, 2022. (VT, VO, LE, GE)

The nervous system is a sensitive target of electromagnetic radiation (EMR). Chronic microwave exposure can induce cognitive deficits, and 5-HT system is involved in this effect. Genetic polymorphisms lead to individual differences. In this study, we evaluated whether the single-nucleotide polymorphism (SNP) rs198585630 of 5-HT_{1A} receptor is associated with cognitive alterations in rats after microwave exposure with a frequency of 2.856 GHz and an average power density of 30 mW/cm^2. Rats were exposed to microwaves for 6 min three times a week for up to 6 weeks. PC12 cells and 293T cells were exposed to microwaves for 5 min up to 3 times at 2 intervals of 5 min. Transcriptional activity of 5-HT_{1A} receptor promoter containing rs198585630 C/T allele was determined in vitro. Electroencephalograms (EEGs), spatial learning and memory, and mRNA and protein expression of 5-HT_{1A} receptor were evaluated in vivo. We demonstrated that transcriptional activity of 5-HT_{1A} receptor promoter containing rs198585630 C allele was higher than that of 5-HT_{1A} receptor promoter containing T allele. The transcriptional activity of 5-HT_{1A} receptor promoter was stimulated by 30 mW/cm^2 microwave exposure, and rs198585630 C allele was more sensitive to microwave exposure, as it showed stronger transcriptional activation. Rats carrying rs198585630 C allele exhibited increased mRNA and protein expression of 5-HT_{1A} receptor and were more susceptible to 30 mW/cm^2 microwave exposure, showing cognitive deficits and inhibition of brain electrical activity. These findings suggest SNP rs198585630 of the 5-HT_{1A} receptor is an important target for further research exploring the mechanisms of hypersensitivity to microwave exposure.

(NE) Li L, Bisht KS, LaGroye I, Zhang P, Straube WL, Moros EG, Roti Roti JL.Measurement of DNA damage in mammalian cells exposed in vitro to radiofrequency fields at sars of 3-5 w/kg. Radiat Res 156:328-332, 2001. (VT, AE, GT)

In the present study, we determined whether exposure of mammalian cells to 3.2-5.1 W/kg specific absorption rate (SAR) radiofrequency fields could induce DNA damage in murine C3H 10T(1/2) fibroblasts. Cell cultures were exposed to 847.74 MHz code-division multiple access (CDMA) and 835.62 frequency-division multiple access (FDMA) modulated radiations in radial transmission line (RTL) irradiators in which the temperature was regulated to 37.0 +/- 0.3 degrees C. Using the alkaline comet assay to measure DNA damage, we found no statistically significant differences in either comet moment or comet length between sham-

exposed cells and those exposed for 2, 4 or 24 h to CDMA or FDMA radiations in either exponentially growing or plateau-phase cells. Further, a 4-h incubation after the 2-h exposure resulted in no significant changes in comet moment or comet length. Our results show that exposure of cultured C3H 10T(1/2) cells at 37 degrees C CDMA or FDMA at SAR values of up to 5.1 W/kg did not induce measurable DNA damage.

(E) Li M, Hao B, Zhang M, Reiter RJ, Lin S, Zheng T, Chen X, Ren Y, Yue L, Abay B, Chen G, Xu X, Shi Y, Fan L. Melatonin enhances radiofrequency-induced NK antitumor immunity, causing cancer metabolism reprogramming and inhibition of multiple pulmonary tumor development. Signal Transduct Target Ther 2021 Sep 1;6(1):330. (VO, AE, GE, IX)

Surgery is the common treatment for early lung cancer with multiple pulmonary nodules, but it is often accompanied by the problem of significant malignancy of other nodules in non-therapeutic areas. In this study, we found that a combined treatment of local radiofrequency ablation (RFA) and melatonin (MLT) greatly improved clinical outcomes for early lung cancer patients with multiple pulmonary nodules by minimizing lung function injury and reducing the probability of malignant transformation or enlargement of nodules in non-ablated areas. Mechanically, as demonstrated in an associated mouse lung tumor model, RFA not only effectively remove treated tumors but also stimulate antitumor immunity, which could inhibit tumor growth in non-ablated areas. MLT enhanced RFA-stimulated NK activity and exerted synergistic antitumor effects with RFA. Transcriptomics and proteomics analyses of residual tumor tissues revealed enhanced oxidative phosphorylation and reduced acidification as well as hypoxia in the tumor microenvironment, which suggests reprogrammed tumor metabolism after combined treatment with RFA and MLT. Analysis of residual tumor further revealed the depressed activity of MAPK, NF-kappa B, Wnt, and Hedgehog pathways and upregulated P53 pathway in tumors, which was in line with the inhibited tumor growth. Combined RFA and MLT treatment also reversed the Warburg effect and decreased tumor malignancy. These findings thus demonstrated that combined treatment of RFA and MLT effectively inhibited the malignancy of non-ablated nodules and provided an innovative non-invasive strategy for treating early lung tumors with multiple pulmonary nodules.

(E) Li R, Ma M, Li L, Zhao L, Zhang T, Gao X, Zhang D, Zhu Y, Peng Q, Luo X, Wang M. The Protective Effect of Autophagy on DNA Damage in Mouse Spermatocyte-Derived Cells Exposed to 1800 MHz Radiofrequency Electromagnetic Fields. Cell Physiol Biochem. 48(1):29-41, 2018. (VT, AE, GT, OX, RP)

Background/aims: The effects of exposure to radiofrequency electromagnetic fields (RF-EMFs) on the male reproductive system have raised public concern and studies have shown that exposure to RF-EMFs can induce DNA damage and autophagy. However, there are no related reports on the role of autophagy in DNA damage in spermatocytes, especially after exposure to RF-EMFs. The aim of the present study was to determine the mechanism and role of autophagy induced by RF-EMFs in spermatozoa cells. **Methods:** Mouse spermatocyte-derived cells (GC-2) were exposed to RF-EMFs 4 W/kg for 24 h. The level of reactive oxygen species (ROS) was determined by ROS assay kit. Comet assay was utilized to detect DNA damage. Autophagy was

detected by three indicators: LC3II/LC3I, autophagic vacuoles, and GFP-LC3 dots, which were measured by western blot, transmission electron microscopy, and transfection with GFP-LC3, respectively. The expression of the molecular signaling pathway AMP-activated protein kinase (AMPK)/mTOR was determined by western blot. **Results:** The results showed that RF-EMFs induced autophagy and DNA damage in GC-2 cells via ROS generation, and the autophagy signaling pathway AMPK/mTOR was activated by ROS generation. Furthermore, following inhibition of autophagy by knockdown of AMPKα, increased DNA damage was observed in GC-2 cells following RF-EMFs exposure, and overexpression of AMPKα promoted autophagy and attenuated DNA damage. **Conclusions:** These findings demonstrated that the autophagy which was induced by RF-EMFs via the AMPK/mTOR signaling pathway could prevent DNA damage in spermatozoa cells.

(NE) Li Y, Deng P, Chen C, Ma Q, Pi H, He M, Lu Y, Gao P, Zhou C, He Z, Zhang Y, Yu Z, Zhang L. 1,800 MHz Radiofrequency Electromagnetic Irradiation Impairs Neurite Outgrowth With a Decrease in Rap1-GTP in Primary Mouse Hippocampal Neurons and Neuro2a Cells. Front Public Health 9:771508, 2021. (VT, AE, GE)

Background: With the global popularity of communication devices such as mobile phones, there are increasing concerns regarding the effect of radiofrequency electromagnetic radiation (RF-EMR) on the brain, one of the most important organs sensitive to RF-EMR exposure at 1,800 MHz. However, the effects of RF-EMR exposure on neuronal cells are unclear. Neurite outgrowth plays a critical role in brain development, therefore, determining the effects of 1,800 MHz RF-EMR exposure on neurite outgrowth is important for exploring its effects on brain development. **Objectives:** We aimed to investigate the effects of 1,800 MHz RF-EMR exposure for 48 h on neurite outgrowth in neuronal cells and to explore the associated role of the Rap1 signaling pathway. **Material and Methods:** Primary hippocampal neurons from C57BL/6 mice and Neuro2a cells were exposed to 1,800 MHz RF-EMR at a specific absorption rate (SAR) value of 4 W/kg for 48 h. CCK-8 assays were used to determine the cell viability after 24, 48, and 72 h of irradiation. Neurite outgrowth of primary hippocampal neurons (DIV 2) and Neuro2a cells was observed with a 20 × optical microscope and recognized by ImageJ software. Rap1a and Rap1b gene expressions were detected by real-time quantitative PCR. Rap1, Rap1a, Rap1b, Rap1GAP, and p-MEK1/2 protein expressions were detected by western blot. Rap1-GTP expression was detected by immunoprecipitation. The role of Rap1-GTP was assessed by transfecting a constitutively active mutant plasmid (Rap1-Gly_Val-GFP) into Neuro2a cells. **Results:** Exposure to 1,800 MHz RF-EMR for 24, 48, and 72 h at 4 W/kg did not influence cell viability. The neurite length, primary and secondary neurite numbers, and branch points of primary mouse hippocampal neurons were significantly impaired by 48-h RF-EMR exposure. The neurite-bearing cell percentage and neurite length of Neuro2a cells were also inhibited by 48-h RF-EMR exposure. Rap1 activity was inhibited by 48-h RF-EMR with no detectable alteration in either gene or protein expression of Rap1. The protein expression of Rap1GAP increased after 48-h RF-EMR exposure, while the expression of p-MEK1/2 protein decreased. Overexpression of constitutively active Rap1 reversed the decrease in Rap1-GTP and the neurite outgrowth impairment in Neuro2a cells induced by 1,800 MHz RF-EMR exposure for 48 h. **Conclusion:** Rap1 activity and related signaling pathways are involved in the disturbance of

neurite outgrowth induced by 48-h 1,800 MHz RF-EMR exposure. The effects of RF-EMR exposure on neuronal development in infants and children deserve greater focus.

(E) Li ZQ, Zhang Y, Wan YM, Zhou Q, Liu C, Wu HX, Mu YZ, He YF, Rauniyar R, Wu XN. Testing of behavioral and cognitive development in rats after prenatal exposure to 1800 and 2400 MHz radiofrequency fields. J Radiat Res. 61:197-206, 2020. (VO, CE, GE, DE)

The objective of the study was to explore the effects of behavioral and cognitive development in rats after prenatal exposure to 1800 and 2400 MHz radiofrequency fields. Pregnant female rats were exposed to radiofrequency fields beginning on the 21st day of pregnancy. The indicators of physiological and behavioral development were observed and measured in the offspring rats: Y maze measured at 3-weeks postnatal, open field at 7-weeks postnatal, and the expression of N-methyl-D-aspartate receptors (NMDARs) measured by reverse transcription-PCR in the hippocampus at 9-weeks postnatal. The body weight of the 1800 MHz group and the 1800 MHz + WiFi group showed a downward trend. The eye opening time of newborn rats was much earlier in the WiFi group than in the control group. Compared to the control group, the overall path length of the 1800 MHz + WiFi group was shortened and the stationary time was delayed. The path length of the WiFi group was shortened and the average velocity was increased in the error arm. The 1800 MHz + WiFi group displayed an increased trend in path length, duration, entry times and stationary time in the central area. In both the 1800 MHz + WiFi and WiFi groups, NR2A and NR2B expression was down-regulated, while NR2D, NR3A and NR3B were up-regulated. Moreover, NR1 and NR2C in the WiFi group were also up-regulated. Prenatal exposure to 1800 MHz and WiFi radiofrequency may affect the behavioral and cognitive development of offspring rats, which may be associated with altered mRNA expression of NMDARs in the hippocampus.

(E)Lin KW, Yang CJ, Lian HY, Cai P. Exposure of ELF-EMF and RF-EMF increase the rate of glucose transport and TCA cycle in budding yeast. Front Microbiol. 7:1378, 2016 (VO, AE, GE)

In this study, we investigated the transcriptional response to 50 Hz extremely low frequency electromagnetic field (ELF-EMF) and 2.0 GHz radio frequency electromagnetic field (RF-EMF) exposure by Illumina sequencing technology using budding yeast as the model organism. The transcription levels of 28 genes were upregulated and those of four genes were downregulated under ELF-EMF exposure, while the transcription levels of 29 genes were upregulated and those of 24 genes were downregulated under RF-EMF exposure. After validation by reverse transcription quantitative polymerase chain reaction (RT-qPCR), a concordant direction of change both in differential gene expression (DGE) and RT-qPCR was demonstrated for nine genes under ELF-EMF exposure and for 10 genes under RF-EMF exposure. The RT-qPCR results revealed that ELF-EMF and RF-EMF exposure can upregulate the expression of genes involved in glucose transportation and the tricarboxylic acid (TCA) cycle, but not the glycolysis pathway. Energy metabolism is closely related with the cell response to environmental stress including EMF exposure. Our findings may throw light on the mechanism underlying the biological effects of EMF.

(E) Lin Y-Y, Wu T, Liu J-Y, Gao P, Li K-C, Guo Q-Y, Yuan M, Lang H-Y, Zeng L-H, Guo G-Z. 1950 MHz Radio Frequency Electromagnetic Radiation Inhibits Testosterone Secretion of Mouse Leydig Cells. Int J Environ Res Public Health 15(1):17, 2017. (VT, AE, GE)

More studies that are focused on the bioeffects of radio-frequency (RF) electromagnetic radiation that is generated from the communication devices, but there were few reports with confirmed results about the bioeffects of RF radiation on reproductive cells. To explore the effects of 1950 MHz RF electromagnetic radiation (EMR) on mouse Leydig (TM3) cells. TM3 cells were irradiated or sham-irradiated continuously for 24 h by the specific absorption rate (SAR) 3 W/kg radiation. At 0, 1, 2, 3, 4, and 5 days after irradiation, cell proliferation was detected by cell counting kit-8 (CCK-8) method, cell cycle distribution, percentage of apoptosis, and cellular reactive oxygen species (ROS) were examined by flow cytometry, Testosterone level was measured using enzyme-linked immunosorbent assay (ELISA) assay, messenger ribonucleic acid (mRNA) expression level of steroidogenic acute regulatory protein (StAR) and P450scc in TM3 cells was detected by real-time polymerase chain reaction (PCR). After being irradiated for 24 h, cell proliferation obviously decreased and cell cycle distribution, secretion capacity of Testosterone, and P450scc mRNA level were reduced. While cell apoptosis, ROS, and StAR mRNA level did not change significantly. The current results indicated that 24 h of exposure at 1950 MHz 3 W/kg radiation could cause some adverse effects on TM3 cells proliferation and Testosterone secretion, further studies about the biological effects in the reproductive system that are induced by RF radiation are also needed.

(E) Liu C, Duan W, Xu S, Chen C, He M, Zhang L, Yu Z, Zhou Z. Exposure to 1800 MHz radiofrequency electromagnetic radiation induces oxidative DNA base damage in a mouse spermatocyte-derived cell line. Toxicol Lett 218(1): 2-9, 2013a. (VT, AE, GT, OX, RP)

Whether exposure to radiofrequency electromagnetic radiation (RF-EMR) emitted from mobile phones can induce DNA damage in male germ cells remains unclear. In this study, we conducted a 24 h intermittent exposure (5 min on and 10 min off) of a mouse spermatocyte-derived GC-2 cell line to 1800 MHz Global System for Mobile Communication (GSM) signals in GSM-Talk mode at specific absorption rates (SAR) of 1 W/kg, 2 W/kg or 4 W/kg. Subsequently, through the use of formamidopyrimidine DNA glycosylase (FPG) in a modified comet assay, we determined that the extent of DNA migration was significantly increased at a SAR of 4 W/kg. Flow cytometry analysis demonstrated that levels of the DNA adduct 8-oxoguanine (8-oxoG) were also increased at a SAR of 4 W/kg. These increases were concomitant with similar increases in the generation of reactive oxygen species (ROS); these phenomena were mitigated by co-treatment with the antioxidant α-tocopherol. However, no detectable DNA strand breakage was observed by the alkaline comet assay. Taking together, these findings may imply the novel possibility that RF-EMR with insufficient energy for the direct induction of DNA strand breaks may produce genotoxicity through oxidative DNA base damage in male germ cells.

(E) Liu C, Gao P, Xu SC, Wang Y, Chen CH, He MD, Yu ZP, Zhang L, Zhou Z. Mobile phone radiation induces mode-dependent DNA damage in a mouse spermatocyte-derived

cell line: a protective role of melatonin. Int J Radiat Biol. 89(11):993-1001, 2013b. (VT, AE, GT, OX, RP)

Purpose: To evaluate whether exposure to mobile phone radiation (MPR) can induce DNA damage in male germ cells. Materials and methods: A mouse spermatocyte-derived GC-2 cell line was exposed to a commercial mobile phone handset once every 20 minutes in standby, listen, dialed or dialing modes for 24 h. DNA damage was determined using an alkaline comet assay. Results: The levels of DNA damage were significantly increased following exposure to MPR in the listen, dialed and dialing modes. Moreover, there were significantly higher increases in the dialed and dialing modes than in the listen mode. Interestingly, these results were consistent with the radiation intensities of these modes. However, the DNA damage effects of MPR in the dialing mode were efficiently attenuated by melatonin pretreatment. Conclusions: These results regarding mode-dependent DNA damage have important implications for the safety of inappropriate mobile phone use by males of reproductive age and also suggest a simple preventive measure, keeping our body from mobile phones as far away as possible, not only during conversations but during "dialed" and "dialing" operation modes as well. Since the "dialed" mode is actually part of the standby mode, mobile phones should be kept at a safe distance from our body even during standby operation. Furthermore, the protective role of melatonin suggests that it may be a promising pharmacological candidate for preventing mobile phone use-related reproductive impairments.

(E) Lixia S, Yao K, Kaijun W, Deqiang L, Huajun H, Xiangwei G, Baohong W, Wei Z, Jianling L, Wei W. Effects of 1.8GHz radiofrequency field on DNA damage and expression of heat shock protein 70 in human lens epithelial cells. Mutat Res 602(1-2):135-42, 2006. (VT, AE, GT, GE)

To investigate the DNA damage, expression of heat shock protein 70 (Hsp70) and cell proliferation of human lens epithelial cells (hLEC) after exposure to the 1.8GHz radiofrequency field (RF) of a global system for mobile communications (GSM). An Xc-1800 RF exposure system was used to employ a GSM signal at 1.8GHz (217Hz amplitude-modulated) with the output power in the specific absorption rate (SAR) of 1, 2 and 3W/kg. After 2h exposure to RF, the DNA damage of hLEC was accessed by comet assay at five different incubation times: 0, 30, 60, 120 and 240min, respectively. Western blot and RT-PCR were used to determine the expression of Hsp70 in hLECs after RF exposure. The proliferation rate of cells was evaluated by bromodeoxyuridine incorporation on days 0, 1 and 4 after exposure. The results show that the difference of DNA-breaks between the exposed and sham-exposed (control) groups induced by 1 and 2W/kg irradiation were not significant at any incubation time point ($P>0.05$). The DNA damage caused by 3W/kg irradiation was significantly increased at the times of 0 and 30min after exposure ($P<0.05$), a phenomenon that could not be seen at the time points of 60, 120 or 240min ($P>0.05$). Detectable mRNA as well as protein expression of Hsp70 was found in all groups. Exposure at SARs of 2 and 3W/kg for 2h exhibited significantly increased Hsp70 protein expression ($P<0.05$), while no change in Hsp70 mRNA expression could be found in any of the groups ($P>0.05$). No difference of the cell proliferation rate between the sham-exposed and exposed cells was found at any exposure dose tested ($P>0.05$). The results indicate that exposure to non-thermal dosages of RF for wireless communications can induce no or repairable DNA

damage and the increased Hsp70 protein expression in hLECs occurred without change in the cell proliferation rate. The non-thermal stress response of Hsp70 protein increase to RF exposure might be involved in protecting hLEC from DNA damage and maintaining the cellular capacity for proliferation.

(E) López-Martín E, Bregains J, Relova-Quinteiro JL, Cadarso-Suárez C, Jorge-Barreiro FJ, Ares-Pena FJ. The action of pulse-modulated GSM radiation increases regional changes in brain activity and c-Fos expression in cortical and subcortical areas in a rat model of picrotoxin-induced seizure proneness. J Neurosci Res. 87(6):1484-1499, 2009. (VO, AE, GE, WS)

The action of the pulse-modulated GSM radiofrequency of mobile phones has been suggested as a physical phenomenon that might have biological effects on the mammalian central nervous system. In the present study, GSM-exposed picrotoxin-pretreated rats showed differences in clinical and EEG signs, and in c-Fos expression in the brain, with respect to picrotoxin-treated rats exposed to an equivalent dose of unmodulated radiation. Neither radiation treatment caused tissue heating, so thermal effects can be ruled out. The most marked effects of GSM radiation on c-Fos expression in picrotoxin-treated rats were observed in limbic structures, olfactory cortex areas and subcortical areas, the dentate gyrus, and the central lateral nucleus of the thalamic intralaminar nucleus group. Nonpicrotoxin-treated animals exposed to unmodulated radiation showed the highest levels of neuronal c-Fos expression in cortical areas. These results suggest a specific effect of the pulse modulation of GSM radiation on brain activity of a picrotoxin-induced seizure-proneness rat model and indicate that this mobile-phone-type radiation might induce regional changes in previous preexcitability conditions of neuronal activation.

(E) López-Martín E, Jorge-Barreirob FJ, Relova-Quinteroc JL, .Salas-Sánchezde AA, Ares-Penad FJ. Exposure to 2.45 GHz radiofrequency modulates calcitonin-dependent activity and HSP-90 protein in parafollicular cells of rat thyroid gland. Tissue and Cell 68: 101478, 2021.(VO, AE, GE)

In this study we analyzed the response of parafollicular cells in rat thyroid gland after exposure to radiofrequency at 2.45 GHz using a subthermal experimental diathermy model. Forty-two Sprague Dawley rats, divided into two groups of 21 rats each, were individually exposed at 0 (control), 3 or 12 W in a Gigahertz Transverse Electro-Magnetic (GTEM) chamber for 30 min. After radiation, we used simple or fluorescence immunohistochemistry to measure calcitonin cells or cellular stress levels, indicated by the presence hyperplasia of parafollicular cells, heat shock protein (HSP) 90. Immunomarking of calcitonin-positive cells was statistically significant higher in the thyroid tissue of rats exposed to 2.45 GHz radiofrequency and cell hyperplasia appeared 90 min after radiation at the SAR levels studied. At the same time, co-localized expression of HSP-90 and calcitonin in parafollicular cells was statistically significant attenuated 90 min after radiation and remained statistically significantly low 24 h after radiation, even though parafollicular cell levels normalized. These facts indicate that subthermal radiofrequency (RF) at 2.45 GHz constitutes a negative external stress stimulus that alters the activity and

homeostasis of parafollicular cells in the rat thyroid gland. However, further research is needed to determine if there is toxic action in human C cells.

(E) Luukkonen J, Hakulinen P, Mäki-Paakkanen J, Juutilainen J, Naarala J. Enhancement of chemically induced reactive oxygen species production and DNA damage in human SH-SY5Y neuroblastoma cells by 872MHz radiofrequency radiation. Mutat Res 662:54-58, 2009. (VT, AE, GT, OX, WS)

The objective of the study was to investigate effects of 872 MHz radiofrequency (RF) radiation on intracellular reactive oxygen species (ROS) production and DNA damage at a relatively high SAR value (5W/kg). The experiments also involved combined exposure to RF radiation and menadione, a chemical inducing intracellular ROS production and DNA damage. The production of ROS was measured using the fluorescent probe dichlorofluorescein and DNA damage was evaluated by the Comet assay. Human SH-SY5Y neuroblastoma cells were exposed to RF radiation for 1h with or without menadione. Control cultures were sham exposed. Both continuous waves (CW) and a pulsed signal similar to that used in global system for mobile communications (GSM) mobile phones were used. Exposure to the CW RF radiation increased DNA breakage ($p<0.01$) in comparison to the cells exposed only to menadione. Comparison of the same groups also showed that ROS level was higher in cells exposed to CW RF radiation at 30 and 60 min after the end of exposure ($p<0.05$ and $p<0.01$, respectively). No effects of the GSM signal were seen on either ROS production or DNA damage. The results of the present study suggest that 872MHz CW RF radiation at 5W/kg might enhance chemically induced ROS production and thus cause secondary DNA damage. However, there is no known mechanism that would explain such effects from CW RF radiation but not from GSM modulated RF radiation at identical SAR.

(NE) Luukkonen J, Juutilainen J, Naarala J. Combined effects of 872 MHz radiofrequency radiation and ferrous chloride on reactive oxygen species production and DNA damage in human SH-SY5Y neuroblastoma cells. Bioelectromagnetics 31:417-424, 2010. (VT, AE, GT, OX)

The aim of the present study was to investigate possible cooperative effects of radiofrequency (RF) radiation and ferrous chloride (FeCl) on reactive oxygen species (ROS) production and DNA damage. In order to test intracellular ROS production as a possible underlying mechanism of DNA damage, we applied the fluorescent probe DCFH-DA. Integrity of DNA was quantified by alkaline comet assay. The exposures to 872 MHz RF radiation were conducted at a specific absorption rate (SAR) of 5 W/kg using continuous waves (CW) or a modulated signal similar to that used in Global System for Mobile Communications (GSM) phones. Four groups were included: Sham exposure (control), RF radiation, Chemical treatment, Chemical treatment, and RF radiation. In the ROS production experiments, human neuroblastoma (SH-SY5Y) cells were exposed to RF radiation and 10 microg/ml FeCl for 1 h. In the comet assay experiments, the exposure time was 3 h and an additional chemical (0.015% diethyl maleate) was used to make DNA damage level observable. The chemical treatments resulted in statistically significant responses, but no effects from either CW or modulated RF radiation were observed on ROS production, DNA damage or cell viability.

(E) Maes A, Verschaeve L, Arroyo A, De Wagter C, Vercruyssen L, In vitro cytogenetic effects

of 2450 MHz waves on human peripheral blood lymphocytes. *Bioelectromagnetics* 14(6):495-501, 1993. (VT, AE, GT)

Cytogenetic analyses were performed on human peripheral blood lymphocytes exposed to 2450 MHz microwaves during 30 and 120 min at a constant temperature of 36.1 degrees C (body temperature). The temperature was kept constant by means of a temperature probe put in the blood sample which gives feedback to a microcomputer that controls the microwave supply. We found a marked increase in the frequency of chromosome aberrations (including dicentric chromosomes and acentric fragments) and micronuclei. On the other hand the microwave exposure did not influence the cell kinetics nor the sister chromatid exchange (SCE) frequency.

(E) Maes A, Collier M, Slaets D, Verschaeve L, 954 MHz microwaves enhance the mutagenic properties of mitomycin C. *Environ Mol Mutagen* 28(1):26-30, 1996. (VT, AE, GT, IX)

This paper focuses on the combined effects of microwaves from mobile communication frequencies and a chemical DNA damaging agent mitomycin C (MMC). The investigation was performed in vitro by exposing whole blood samples to a 954 MHz emitting antenna from a GSM (Global System for Mobile Communication) base station, followed by lymphocyte cultivation in the presence of MMC. A highly reproducible synergistic effect was observed as based on the frequencies of sister chromatid exchanges in metaphase figures.

(NE) Maes A, Collier M, Van Gorp U, Vandoninck S, Verschaeve L, Cytogenetic effects of 935.2-MHz (GSM) microwaves alone and in combination with mitomycin C. *Mutat Res* 393(1-2):151-156, 1997. (VT, AE, GT, IX)

This paper focuses on the genetic effects of microwaves from mobile communication frequencies (935.2 MHz) alone and in combination with a chemical DNA-damaging agent (mitomycin C). Three cytogenetic endpoints were investigated after in vitro exposure of human whole blood cells. These endpoints were the 'classical' chromosome aberration test, the sister chromatid exchange test and the alkaline comet assay. No direct cytogenetic effect was found. The combined exposure of the cells to the radiofrequency fields followed by their cultivation in the presence of mitomycin C revealed a very weak effect when compared to cells exposed to mitomycin C alone.

(NE) Maes A, Collier M, Verschaeve L Cytogenetic investigations on microwaves emitted by a 455.7 MHz car phone. Folia Biol (Praha) 46(5):175-180, 2000. (VT, AE, GT, IX)

The chromosome aberration or sister chromatid exchange frequency was determined in 455.7 MHz microwave-exposed human lymphocytes and in lymphocytes that were subsequently exposed to MMC or X-rays. The exposure was performed by placing the cells at 5 cm from the antenna of a car phone. In this way the specific absorption ratio was approximately 6.5 W/kg. The temperature and humidity was kept constant during the experiments. No statistically significant difference was found between microwave-exposed and unexposed control samples. When the microwave exposure was followed by exposure to MMC, some differences were found between the combined treatments and the MMC treatments alone. However, there was

no consistency in the results. Combined treatments with X-rays did not provide any indication of a synergistic action between the RF fields and X-rays, either. Our data therefore do not support the hypothesis that RF fields act synergistically with chemical or physical mutagens.

(NE) Maes A, Collier M, Verschaeve L Cytogenetic effects of 900 MHz (GSM) microwaves on human lymphocytes. Bioelectromagnetics 22(2):91-96, 2001. (VT, AE, GT, IX)

The cytogenetic effects of 900 MHz radiofrequency fields were investigated with the chromosome aberration and sister chromatid exchange frequency methods. Three different modes of exposure (continuous, pseudo-random and dummy burst) were studied for different power outputs (0, 2, 8, 15, 25, 50 W). The specific absorption rates varied between 0 and 10 W/kg. We investigated the possible effects of the 900 MHz radiation alone as well as of combined exposure to the chemical or physical mutagens mitomycin C and X-rays. Overall, no indication was found of a mutagenic, and/or co-mutagenic/synergistic effect of this kind of nonionizing radiation.

(NE) Maes A, Van Gorp U, Verschaeve L. Cytogenetic investigation of subjects professionally exposed to radiofrequency radiation. Mutagenesis 21:139-142, 2006. (HU, VT, LE, GT, IX)

Nowadays, virtually everybody is exposed to radiofrequency radiation (RFR) from mobile phone base station antennas or other sources. At least according to some scientists, this exposure can have detrimental health effects. We investigated cytogenetic effects in peripheral blood lymphocytes from subjects who were professionally exposed to mobile phone electromagnetic fields in an attempt to demonstrate possible RFR-induced genetic effects. These subjects can be considered well suited for this purpose as their RFR exposure is 'normal' though rather high, and definitely higher than that of the 'general population'. The alkaline comet assay, sister chromatid exchange (SCE) and chromosome aberration tests revealed no evidence of RFR-induced genetic effects. Blood cells were also exposed to the well known chemical mutagen mitomycin C in order to investigate possible combined effects of RFR and the chemical. No cooperative action was found between the electromagnetic field exposure and the mutagen using either the comet assay or SCE test.

(NE) Malini V, Resolving the enigma of effect of mobile phone usage on spermatogenetsis in humans in south Indian population. Asian J Pharmaceut Clin Res. 10: 233-237, 2017. (HU, LE, GT)

Objective: This study was aimed at to evaluate the possible risk of radiofrequency and electromagnetic waves of mobile phones on spermatogenic impairment and functional capacity of the spermatozoa along with oxidative stress, DNA damages, and hormone profile among mobile phone users. **Methods:** Mobile phone users were classified into three groups are 1-5, 6-10, and above 10 hrs/day, respectively, based on the exposure to electromagnetic radiation. Blood and semen samples are collected with informed consent letter. The semen samples used to carry out to the physical examination such as volume, liquefaction time, color, odor, pH, and viscosity, and functional status of the spermatozoa was carried out such as nuclear chromatin decondensation test, hypo-osmotic swelling test, and acrosomal intactness test. Seminal plasma

was used for to evaluate the oxidative stress markers superoxide dismutase (SOD) activity, reactive oxygen species (ROS) levels, and total antioxidant capacity (TAC). Blood serum was used to estimate the level of follicle stimulating hormone (FSH), luteinizing hormone (LH), and testosterone. DNA collected from blood used for DNA ladder assay. **Results:** In the present investigation, both physical and microscopic examinations were negatively correlated with mobile phone usage. No variation exists in functional status of spermatozoa. Oxidative stress markers such as the presence of ROS, enzymatic scavengers such as SOD and TAC showed no statistical variations between control group and mobile phone users and even no variations in hormone profile such as testosterone, FSH, and LH of users of mobile phone compared to normal reference values. **Conclusion:** In conclusion, though the literature has suggested that mobile phone use alters semen parameters, functional status of spermatozoa, increased oxidative stress, with subsequent sperm DNA damage in humans. The present study deviates from previous study stating nil impact of mobile phones on spermatogenetic impairment in humans.

(NE) Malyapa RS, Ahern EW, Straube WL, Moros EG, Pickard WF, Roti Roti JL, Measurement of DNA damage after exposure to 2450 MHz electromagnetic radiation. Radiat Res 148(6):608-617, 1997a. (VT, AE, GT)

Recent reports suggest that exposure to 2450 MHz electromagnetic radiation causes DNA single-strand breaks (SSBs) and double-strand breaks (DSBs) in cells of rat brain irradiated in vivo (Lai and Singh, Bioelectromagnetics 16, 207-210, 1995; Int. J. Radiat. Biol. 69, 513-521, 1996). Therefore, we endeavored to determine if exposure of cultured mammalian cells in vitro to 2450 MHz radiation causes DNA damage. The alkaline comet assay (single-cell gel electrophoresis), which is reportedly the most sensitive method to assay DNA damage in individual cells, was used to measure DNA damage after in vitro 2450 MHz irradiation. Exponentially growing U87MG and C3H 10T1/2 cells were exposed to 2450 MHz continuous-wave (CW) radiation in specially designed radial transmission lines (RTLs) that provided relatively uniform microwave exposure. Specific absorption rates (SARs) were calculated to be 0.7 and 1.9 W/kg. Temperatures in the RTLs were measured in real time and were maintained at 37 +/- 0.3 degrees C. Every experiment included sham exposure(s) in an RTL. Cells were irradiated for 2 h, 2 h followed by a 4-h incubation at 37 degrees C in an incubator, 4 h and 24 h. After these treatments samples were subjected to the alkaline comet assay as described by Olive et al. (Exp. Cell Res. 198, 259-267, 1992). Images of comets were digitized and analyzed using a PC-based image analysis system, and the "normalized comet moment" and "comet length" were determined. No significant differences were observed between the test group and the controls after exposure to 2450 MHz CW irradiation. Thus 2450 MHz irradiation does not appear to cause DNA damage in cultured mammalian cells under these exposure conditions as measured by this assay.

(NE) Malyapa RS, Ahern EW, Straube WL, Moros EG, Pickard WF, Roti Roti JL. Measurement of DNA damage after exposure to electromagnetic radiation in the cellular phone communication frequency band (835.62 and 847.74 MHz). Radiat Res 148(6):618-627, 1997b. (VT, AE, GT)

Mouse C3H 10T1/2 fibroblasts and human glioblastoma U87MG cells were exposed to cellular phone communication frequency radiations to investigate whether such exposure produces DNA damage in in vitro cultures. Two types of frequency modulations were studied: frequency-modulated continuous-wave (FMCW), with a carrier frequency of 835.62 MHz, and code-division multiple-access (CDMA) centered on 847.74 MHz. Exponentially growing (U87MG and C3H 10T1/2 cells) and plateau-phase (C3H 10T1/2 cells) cultures were exposed to either FMCW or CDMA radiation for varying periods up to 24 h in specially designed radial transmission lines (RTLs) that provided relatively uniform exposure with a specific absorption rate (SAR) of 0.6 W/kg. Temperatures in the RTLs were monitored continuously and maintained at 37 +/- 0.3 degrees C. Sham exposure of cultures in an RTL (negative control) and 137Cs gamma-irradiated samples (positive control) were included with every experiment. The alkaline comet assay as described by Olive et al. (Exp. Cell Res. 198, 259-269, 1992) was used to measure DNA damage. No significant differences were observed between the test group exposed to FMCW or CDMA radiation and the sham-treated negative controls. Our results indicate that exposure of cultured mammalian cells to cellular phone communication frequencies under these conditions at an SAR of 0.6 W/kg does not cause DNA damage as measured by the alkaline comet assay.

(NE) Malyapa RS, Ahern EW, Bi C, Straube WL, LaRegina M, Pickard WF, Roti RotiJL, DNA damage in rat brain cells after in vivo exposure to 2450 MHz electromagnetic radiation and various methods of euthanasia. Radiat Res 149(6):637-645, 1998. (VO, AE, GT)

The present study was done to confirm the reported observation that low-intensity acute exposure to 2450 MHz radiation causes DNA single-strand breaks (Lai and Singh, Bioelectromagnetics 16, 207-210, 1995). Male Sprague-Dawley rats weighing approximately 250 g were irradiated with 2450 MHz continuous-wave (CW) microwaves for 2 h at a specific absorption rate of 1.2 W/kg in a cylindrical waveguide system (Guy et al., Radio Sci. 14, 63-74, 1979). There was no associated rise in the core body temperature of the rats. After the irradiation or sham treatments, rats were euthanized by either CO2 asphyxia or decapitation by guillotine (eight pairs of animals per euthanasia group). After euthanasia the brains were removed and immediately immersed in cold Ames medium and the cells of the cerebral cortex and the hippocampus were dissociated separately and subjected to the alkaline comet assay. Irrespective of whether the rats were euthanized by CO2 asphyxia or decapitated by guillotine, no significant differences were observed between either the comet length or the normalized comet moment of cells from either the cerebral cortex or the hippocampus of sham-treated rats and those from the irradiated rats. However, the data for the rats asphyxiated with CO2 showed more intrinsic DNA damage and more experiment-to-experiment variation than did the data for rats euthanized by guillotine. Therefore, the guillotine method of euthanasia is the most appropriate in studies relating to DNA damage. Furthermore, we did not confirm the observation that DNA damage is produced in cells of the rat cerebral cortex or the hippocampus after a 2-h exposure to 2450 MHz CW microwaves or at 4 h after the exposure.

(E) Manta AK, Papadopoulou D, Polyzos AP, Fragopoulou AF, Skouroliakou AS, Thanos D, Stravopodis DJ, Margaritis LH. Mobile-phone radiation-induced perturbation of gene-

expression profiling, redox equilibrium and sporadic-apoptosis control in the ovary of Drosophila melanogaster. Fly (Austin). 11(2):75-95, 2017. (VO, AE, GE, OX)

The daily use by people of wireless communication devices has increased exponentially in the last decade, begetting concerns regarding its potential health hazards. Drosophila melanogaster four days-old adult female flies were exposed for 30 min to radiation emitted by a commercial mobile phone at a SAR of 0.15 W/kg and a SAE of 270 J/kg. ROS levels and apoptotic follicles were assayed in parallel with a genome-wide microarrays analysis. ROS cellular contents were found to increase by 1.6-fold (x), immediately after the end of exposure, in follicles of pre-choriogenic stages (germarium - stage 10), while sporadically generated apoptotic follicles (germarium 2b and stages 7-9) presented with an averaged 2x upregulation in their sub-population mass, 4 h after fly's irradiation with mobile device. Microarray analysis revealed 168 genes being differentially expressed, 2 h post-exposure, in response to radiofrequency (RF) electromagnetic field-radiation exposure (≥1.25x, $P < 0.05$) and associated with multiple and critical biological processes, such as basic metabolism and cellular subroutines related to stress response and apoptotic death. Exposure of adult flies to mobile-phone radiation for 30 min has an immediate impact on ROS production in animal's ovary, which seems to cause a global, systemic and non-targeted transcriptional reprogramming of gene expression, 2 h post-exposure, being finally followed by induction of apoptosis 4 h after the end of exposure. Conclusively, this unique type of pulsed radiation, mainly being derived from daily used mobile phones, seems capable of mobilizing critical cytopathic mechanisms, and altering fundamental genetic programs and networks in D. melanogaster.

(E) Manti L, Braselmann H, Calabrese ML, Massa R, Pugliese M, Scampoli P, Sicignano G, Grossi G. Effects of modulated microwave radiation at cellular telephone frequency (1.95 GHz) on X-ray-induced chromosome aberrations in human lymphocytes in vitro. Radiat Res 169:575-583, 2008. (VT, AE, GT, IX)

The case for a DNA-damaging action produced by radiofrequency (RF) signals remains controversial despite extensive research. With the advent of the Universal Mobile Telecommunication System (UMTS) the number of RF-radiation-exposed individuals is likely to escalate. Since the epigenetic effects of RF radiation are poorly understood and since the potential modifications of repair efficiency after exposure to known cytotoxic agents such as ionizing radiation have been investigated infrequently thus far, we studied the influence of UMTS exposure on the yield of chromosome aberrations induced by X rays. Human peripheral blood lymphocytes were exposed in vitro to a UMTS signal (frequency carrier of 1.95 GHz) for 24 h at 0.5 and 2.0 W/kg specific absorption rate (SAR) using a previously characterized waveguide system. The frequency of chromosome aberrations was measured on metaphase spreads from cells given 4 Gy of X rays immediately before RF radiation or sham exposures by fluorescence in situ hybridization. Unirradiated controls were RF-radiation- or sham-exposed. No significant variations due to the UMTS exposure were found in the fraction of aberrant cells. However, the frequency of exchanges per cell was affected by the SAR, showing a small but statistically significant increase of 0.11 exchange per cell compared to 0 W/kg SAR. We

conclude that, although the 1.95 GHz signal (UMTS modulated) does not exacerbate the yield of aberrant cells caused by ionizing radiation, the overall burden of X-ray-induced chromosomal damage per cell in first-mitosis lymphocytes may be enhanced at 2.0 W/kg SAR. Hence the SAR may either influence the repair of X-ray-induced DNA breaks or alter the cell death pathways of the damage response.

(E) Marinelli F, La Sala D, Cicciotti G, Cattini L, Trimarchi C, Putti S, Zamparelli A, Giuliani L, Tomassetti G, Cinti C. Exposure to 900 MHz electromagnetic field induces an unbalance between pro-apoptotic and pro-survival signals in T-lymphoblastoid leukemia CCRF-CEM cells. J Cell Physiol. 198(2):324-332, 2004.(VT, AE, GT, GE)

It has been recently established that low-frequency electromagnetic field (EMFs) exposure induces biological changes and could be associated with increased incidence of cancer, while the issue remains unresolved as to whether high-frequency EMFs can have hazardous effect on health. Epidemiological studies on association between childhood cancers, particularly leukemia and brain cancer, and exposure to low- and high-frequency EMF suggested an etiological role of EMFs in inducing adverse health effects. To investigate whether exposure to high-frequency EMFs could affect in vitro cell survival, we cultured acute T-lymphoblastoid leukemia cells (CCRF-CEM) in the presence of unmodulated 900 MHz EMF, generated by a transverse electromagnetic (TEM) cell, at various exposure times. We evaluated the effects of high-frequency EMF on cell growth rate and apoptosis induction, by cell viability (MTT) test, FACS analysis and DNA ladder, and we investigated pro-apoptotic and pro-survival signaling pathways possibly involved as a function of exposure time by Western blot analysis. At short exposure times (2-12 h), unmodulated 900 MHz EMF induced DNA breaks and early activation of both p53-dependent and -independent apoptotic pathways while longer continuous exposure (24-48 h) determined silencing of pro-apoptotic signals and activation of genes involved in both intracellular (Bcl-2) and extracellular (Ras and Akt1) pro-survival signaling. Overall our results indicate that exposure to 900 MHz continuous wave, after inducing an early self-defense response triggered by DNA damage, could confer to the survivor CCRF-CEM cells a further advantage to survive and proliferate.

(E) Markova E, Hillert L, Malmgren L, Persson BR, Belyaev IY. Microwaves from GSM mobile telephones affect 53BP1 and gamma-H2AX foci in human lymphocytes from hypersensitive and healthy persons. Environ Health Perspect. 113(9):1172-1177, 2005. (VT, AE, GT, WS)

The data on biologic effects of nonthermal microwaves (MWs) from mobile telephones are diverse, and these effects are presently ignored by safety standards of the International Commission for Non-Ionizing Radiation Protection (ICNIRP). In the present study, we investigated effects of MWs of Global System for Mobile Communication (GSM) at different carrier frequencies on human lymphocytes from healthy persons and from persons reporting hypersensitivity to electromagnetic fields (EMFs). We measured the changes in chromatin conformation, which are indicative of stress response and genotoxic effects, by the method of anomalous viscosity time dependence, and we analyzed tumor suppressor p53-binding protein 1 (53BP1) and phosphorylated histone H2AX (gamma-H2AX), which have been shown to

colocalize in distinct foci with DNA double-strand breaks (DSBs), using immunofluorescence confocal laser microscopy. We found that MWs from GSM mobile telephones affect chromatin conformation and 53BP1/gamma-H2AX foci similar to heat shock. For the first time, we report here that effects of MWs from mobile telephones on human lymphocytes are dependent on carrier frequency. On average, the same response was observed in lymphocytes from hypersensitive and healthy subjects.

(E)Markovà E, Malmgren LO, Belyaev IY. Microwaves from mobile phones inhibit 53BP1 focus formation in human stem cells more strongly than in differentiated cells: possible mechanistic link to cancer risk. Environ Health Perspect. 118(3):394-399, 2010. (VT, AE, GT)

Background: It is widely accepted that DNA double-strand breaks (DSBs) and their misrepair in stem cells are critical events in the multistage origination of various leukemias and tumors, including gliomas. Objectives: We studied whether microwaves from mobile telephones of the Global System for Mobile Communication (GSM) and the Universal Global Telecommunications System (UMTS) induce DSBs or affect DSB repair in stem cells. Methods: We analyzed tumor suppressor TP53 binding protein 1 (53BP1) foci that are typically formed at the sites of DSB location (referred to as DNA repair foci) by laser confocal microscopy. Results: Microwaves from mobile phones inhibited formation of 53BP1 foci in human primary fibroblasts and mesenchymal stem cells. These data parallel our previous findings for human lymphocytes. Importantly, the same GSM carrier frequency (915 MHz) and UMTS frequency band (1947.4 MHz) were effective for all cell types. Exposure at 905 MHz did not inhibit 53BP1 foci in differentiated cells, either fibroblasts or lymphocytes, whereas some effects were seen in stem cells at 905 MHz. Contrary to fibroblasts, stem cells did not adapt to chronic exposure during 2 weeks. Conclusions: The strongest microwave effects were always observed in stem cells. This result may suggest both significant misbalance in DSB repair and severe stress response. Our findings that stem cells are most sensitive to microwave exposure and react to more frequencies than do differentiated cells may be important for cancer risk assessment and indicate that stem cells are the most relevant cellular model for validating safe mobile communication signals.

(E) Martin C, Percevault F, Ryder K, Sani E, Le Cun JC, Zhadobov M, Sauleau R, Le Dréan Y, Habauzit D. Effects of radiofrequency radiation on gene expression: a study of gene expressions of human keratinocytes from different origins. Bioelectromagnetics. 41(7):552-557, 2020. (VT, AE, GE)

(No abstract available.) Human keratinocytes from difference origins showed different patterns of gene expression after acute exposure to a 60-GHz RFR.

(E) Mashevich M, Folkman D, Kesar A, Barbul A, Korenstein R, Jerby E, Avivi L. Exposure of human peripheral blood lymphocytes to electromagnetic fields associated with cellular phones leads to chromosomal instability. Bioelectromagnetics 24:82-90, 2003. (VT, AE, GT)

Whether exposure to radiation emitted from cellular phones poses a health hazard is at the focus of current debate. We have examined whether in vitro exposure of human peripheral blood lymphocytes (PBL) to continuous 830 MHz electromagnetic fields causes losses and gains of chromosomes (aneuploidy), a major "somatic mutation" leading to genomic instability and thereby to cancer. PBL were irradiated at different average absorption rates (SAR) in the range of 1.6-8.8 W/kg for 72 hr in an exposure system based on a parallel plate resonator at temperatures ranging from 34.5-37.5 °C. The averaged SAR and its distribution in the exposed tissue culture flask were determined by combining measurements and numerical analysis based on a finite element simulation code. A linear increase in chromosome 17 aneuploidy was observed as a function of the SAR value, demonstrating that this radiation has a genotoxic effect. The SAR dependent aneuploidy was accompanied by an abnormal mode of replication of the chromosome 17 region engaged in segregation (repetitive DNA arrays associated with the centromere), suggesting that epigenetic alterations are involved in the SAR dependent genetic toxicity. Control experiments (i.e., without any RF radiation) carried out in the temperature range of 34.5-38.5 °C showed that elevated temperature is not associated with either the genetic or epigenetic alterations observed following RF radiation - the increased levels of aneuploidy and the modification in replication of the centromeric DNA arrays. These findings indicate that the genotoxic effect of the electromagnetic radiation is elicited via a non-thermal pathway. Moreover, the fact that aneuploidy is a phenomenon known to increase the risk for cancer, should be taken into consideration in future evaluation of exposure guidelines.

(E) Mazor R, Korenstein-Ilan A, Barbul A, Eshet Y, Shahadi A, Jerby E, Korenstein R. Increased levels of numerical chromosome aberrations after in vitro exposure of human peripheral blood lymphocytes to radiofrequency electromagnetic fields for 72 hours. Radiat Res. 169(1):28-37, 2008. (VT, AE, GT)

We investigated the effects of 72 h in vitro exposure of 10 human lymphocyte samples to radiofrequency electromagnetic fields (800 MHz, continuous wave) on genomic instability. The lymphyocytes were exposed in a specially designed waveguide resonator at specific absorption rates (SARs) of 2.9 and 4.1 W/kg in a temperature range of 36-37 degrees C. The induced aneuploidy of chromosomes 1, 10, 11 and 17 was determined by interphase FISH using semi-automated image analysis. We observed increased levels of aneuploidy depending on the chromosome studied as well as on the level of exposure. In chromosomes 1 and 10, there was increased aneuploidy at the higher SAR, while for chromosomes 11 and 17, the increases were observed only for the lower SAR. Multisomy (chromosomal gains) appeared to be the primary contributor to the increased aneuploidy. The effect of temperature on the level of aneuploidy was examined over the range of 33.5-40 degrees C for 72 h with no statistically significant difference in the level of aneuploidy compared to 37 degrees C. These findings suggest the possible existence of an athermal effect of RF radiation that causes increased levels of aneuploidy. These results contribute to the assessment of potential health risks after continuous chronic exposure to RF radiation at SARs close to the current levels set by ICNIRP guidelines.

(NE) McNamee JP, Bellier PV, Gajda GB, Miller SM, Lemay EP, Lavallee BF, Marro L, Thansandote A. DNA damage and micronucleus induction in human leukocytes after acute in

vitro exposure to a 1.9 GHz continuous-wave radiofrequency field. Radiat Res 158(4):523-533, 2002a. (VT, AE, GT)

Human blood cultures were exposed to a 1.9 GHz continuous-wave (CW) radiofrequency (RF) field for 2 h using a series of six circularly polarized, cylindrical waveguides. Mean specific absorption rates (SARs) of 0.0, 0.1, 0.26, 0.92, 2.4 and 10 W/kg were achieved, and the temperature within the cultures during a 2-h exposure was maintained at 37.0 +/- 0.5 degrees C. Concurrent negative (incubator) and positive (1.5 Gy (137)Cs gamma radiation) control cultures were run for each experiment. DNA damage was quantified immediately after RF-field exposure using the alkaline comet assay, and four parameters (tail ratio, tail moment, comet length and tail length) were used to assess DNA damage for each comet. No evidence of increased primary DNA damage was detected by any parameter for RF-field-exposed cultures at any SAR tested. The formation of micronuclei in the RF-field-exposed blood cell cultures was assessed using the cytokinesis-block micronucleus assay. There was no significant difference in the binucleated cell frequency, incidence of micronucleated binucleated cells, or total incidence of micronuclei between any of the RF-field-exposed cultures and the sham-exposed controls at any SAR tested. These results do not support the hypothesis that acute, nonthermalizing 1.9 GHz CW RF-field exposure causes DNA damage in cultured human leukocytes.

(NE) McNamee JP, Bellier PV, Gajda GB, Lavallee BF, Lemay EP, Marro L, Thansandote A. DNA Damage in human leukocytes after acute in vitro exposure to a 1.9 GHz pulse-modulated radiofrequency field. Radiat Res 158(4):534-537, 2002b. (VT, AE, GT)

Blood cultures from human volunteers were exposed to an acute 1.9 GHz pulse-modulated radiofrequency (RF) field for 2 h using a series of six circularly polarized, cylindrical waveguides. Mean specific absorption rates (SARs) ranged from 0 to 10 W/kg, and the temperature within the cultures during the exposure was maintained at 37.0 +/- 0.5 degrees C. DNA damage was quantified in leukocytes by the alkaline comet assay and the cytokinesis-block micronucleus assay. When compared to the sham-treated controls, no evidence of increased primary DNA damage was detected by any parameter for any of the RF-field-exposed cultures when evaluated using the alkaline comet assay. Furthermore, no significant differences in the frequency of binucleated cells, incidence of micronucleated binucleated cells, or total incidence of micronuclei were detected between any of the RF-field-exposed cultures and the sham-treated control at any SAR tested. These results do not support the hypothesis that acute, nonthermalizing 1.9 GHz pulse-modulated RF-field exposure causes DNA damage in cultured human leukocytes.

(NE) McNamee, J. P., Bellier, P. V., Gajda, G. B., Lavallee, B. F., Marro, L., Lemay, E. and Thansandote, A. No evidence for genotoxic effects from 24 h exposure of human leukocytes to 1.9 GHz radiofrequency fields. Radiat Res 159:693-697, 2003. (VT, AE, GT)

The current study extends our previous investigations of 2-h radiofrequency (RF)-field exposures on genotoxicity in human blood cell cultures by examining the effect of 24-h continuous-wave (CW) and pulsed-wave (PW) 1.9 GHz RF-field exposures on both primary DNA damage and micronucleus induction in human leukocyte cultures. Mean specific absorption

rates (SARs) ranged from 0 to 10 W/kg, and the temperature within the cultures was maintained at 37.0 +/- 1.0 degrees C for the duration of the 24-h exposure period. No significant differences in primary DNA damage were observed between the sham-treated controls and any of the CW or PW 1.9 GHz RF-field-exposed cultures when processed immediately after the exposure period by the alkaline comet assay. Similarly, no significant differences were observed in the incidence of micronuclei, incidence of micronucleated binucleated cells, frequency of binucleated cells, or proliferation index between the sham-treated controls and any of the CW or PW 1.9 GHz RF-field-exposed cultures. In conclusion, the current study found no evidence of 1.9 GHz RF-field-induced genotoxicity in human blood cell cultures after a 24-h exposure period.

(NE) McNamee JP, Bellier PV, Konkle AT, Thomas R, Wasoontarajaroen S, Lemay E, Gajda GB. Analysis of gene expression in mouse brain regions after exposure to 1.9 GHz radiofrequency fields. Int J Radiat Biol. 92(6):338-350, 2016. (VO, LE, GE)

PURPOSE: To assess 1.9 GHz radiofrequency (RF) field exposure on gene expression within a variety of discrete mouse brain regions using whole genome microarray analysis MATERIALS AND METHODS: Adult male C57BL/6 mice were exposed to 1.9 GHz pulse-modulated or continuous-wave RF fields for 4 h/day for 5 consecutive days at whole body average (WBA) specific absorption rates of 0 (sham), ~0.2 W/kg and ~1.4 W/kg. Total RNA was isolated from the auditory cortex, amygdala, caudate, cerebellum, hippocampus, hypothalamus, and medial prefrontal cortex and differential gene expression was assessed using Illumina MouseWG-6 (v2) BeadChip arrays. Validation of potentially responding genes was conducted by RT-PCR. RESULTS: When analysis of gene expression was conducted within individual brain regions when controlling the false discovery rate (FDR), no differentially expressed genes were identified relative to the sham control. However, it must be noted that most fold changes among groups were observed to be less than 1.5-fold and this study had limited ability to detect such small changes. While some genes were differentially expressed without correction for multiple-comparisons testing, no consistent pattern of response was observed among different RF-exposure levels or among different RF-modulations. CONCLUSIONS: The current study provides the most comprehensive analysis of potential gene expression changes in the rodent brain in response to RF field exposure conducted to date. Within the exposure conditions and limitations of this study, no convincing evidence of consistent changes in gene expression was found in response to 1.9 GHz RF field exposure.

(E) Meena R, Kumari K, Kumar J, Rajamani P, Verma HN, Kesari KK. Therapeutic approaches of melatonin in microwave radiations-induced oxidative stress-mediated toxicity on male fertility pattern of Wistar rats. Electromag Biol Med. 33: 81-91, 2014. (VO. LE, GT, OX, RP)

Microwave (MW) radiation produced by wireless telecommunications and a number of electrical devices used in household or in healthcare institutions may adversely affects the reproductive pattern. Present study aimed to investigate the protective effects of melatonin (is well known antioxidant that protects DNA, lipids and proteins from free radical damage) against oxidative

stress-mediated testicular impairment due to long-term exposure of MWs. For this, 70-day-old male Wistar rats were divided into four groups (n = 6/group): Sham exposed, Melatonin (Mel) treated (2 mg/kg), 2.45 GHz MWs exposed and MWs + Mel treated. Exposure took place in Plexiglas cages for 2 h a day for 45 days where, power density (0.21 mW/cm(2)) and specific absorption rate (SAR 0.14 W/Kg) were estimated. After the completion of exposure period, rats were sacrificed and various stress related parameters, that is LDH-X (lactate dehydrogenase isoenzyme) activity, xanthine oxidase (XO), ROS (reactive oxygen species), protein carbonyl content, DNA damage and MDA (malondialdehyde) were performed. Result shows that melatonin prevent oxidative damage biochemically by significant increase ($p < 0.001$) in the levels of testicular LDH-X, decreased ($p < 0.001$) levels of MDA and ROS in testis ($p < 0.01$). Meanwhile, it reversed the effects of MWs on XO, protein carbonyl content, sperm count, testosterone level and DNA fragmentation in testicular cells. These results concluded that the melatonin has strong antioxidative potential against MW induced oxidative stress mediated DNA damage in testicular cells.

(E) Megha K, Deshmukh PS, Ravi AK, Tripathi AK, Abegaonkar MP, Banerjee BD. Effect of Low-Intensity Microwave Radiation on Monoamine Neurotransmitters and Their Key Regulating Enzymes in Rat Brain. Cell Biochem Biophys. 73(1):93-100, 2015a. (VO, LE, GE, LI)

The increasing use of wireless communication devices has raised major concerns towards deleterious effects of microwave radiation on human health. The aim of the study was to demonstrate the effect of low-intensity microwave radiation on levels of monoamine neurotransmitters and gene expression of their key regulating enzymes in brain of Fischer rats. Animals were exposed to 900 MHz and 1800 MHz microwave radiation for 30 days (2 h/day, 5 days/week) with respective specific absorption rates as 5.953 × 10(-4) and 5.835 × 10(-4) W/kg. The levels of monoamine neurotransmitters viz. dopamine (DA), norepinephrine (NE), epinephrine (E) and serotonin (5-HT) were detected using LC-MS/MS in hippocampus of all experimental animals. In addition, mRNA expression of key regulating enzymes for these neurotransmitters viz. tyrosine hydroxylase (TH) (for DA, NE and E) and tryptophan hydroxylase (TPH1 and TPH2) (for serotonin) was also estimated. Results showed significant reduction in levels of DA, NE, E and 5-HT in hippocampus of microwave-exposed animals in comparison with sham-exposed (control) animals. In addition, significant downregulation in mRNA expression of TH, TPH1 and TPH2 was also observed in microwave-exposed animals ($p < 0.05$). In conclusion, the results indicate that low-intensity microwave radiation may cause learning and memory disturbances by altering levels of brain monoamine neurotransmitters at mRNA and protein levels.

(E) Megha K, Deshmukh PS, Banerjee BD, Tripathi AK, Ahmed R, Abegaonkar MP. Low intensity microwave radiation induced oxidative stress, inflammatory response and DNA damage in rat brain. NeuroToxicol. 51: 158-165, 2015b. (VO, LE, GT, LI, OX)

Over the past decade people have been constantly exposed to microwave radiation mainly from wireless communication devices used in day to day life. Therefore, the concerns over potential adverse effects of microwave radiation on human health are increasing. Until now no study has been proposed to investigate the underlying causes of genotoxic effects induced by low intensity

microwave exposure. Thus, the present study was undertaken to determine the influence of low intensity microwave radiation on oxidative stress, inflammatory response and DNA damage in rat brain. The study was carried out on 24 male Fischer 344 rats, randomly divided into four groups (n=6 in each group): group I consisted of sham exposed (control) rats, group II-IV consisted of rats exposed to microwave radiation at frequencies 900, 1800 and 2450 MHz, specific absorption rates (SARs) 0.59, 0.58 and 0.66 mW/kg, respectively in gigahertz transverse electromagnetic (GTEM) cell for 60 days (2h/day, 5 days/week). Rats were sacrificed and decapitated to isolate hippocampus at the end of the exposure duration. Low intensity microwave exposure resulted in a frequency dependent significant increase in oxidative stress markers viz. malondialdehyde (MDA), protein carbonyl (PCO) and catalase (CAT) in microwave exposed groups in comparison to sham exposed group ($p<0.05$). Whereas, levels of reduced glutathione (GSH) and superoxide dismutase (SOD) were found significantly decreased in microwave exposed groups ($p<0.05$). A significant increase in levels of pro-inflammatory cytokines (IL-2, IL-6, TNF-α, and IFN-γ) was observed in microwave exposed animal ($p<0.05$). Furthermore, significant DNA damage was also observed in microwave exposed groups as compared to their corresponding values in sham exposed group ($p<0.05$). In conclusion, the present study suggests that low intensity microwave radiation induces oxidative stress, inflammatory response and DNA damage in brain by exerting a frequency dependent effect. The study also indicates that increased oxidative stress and inflammatory response might be the factors involved in DNA damage following low intensity microwave exposure.

Meltz M. Radiofrequency exposure and mammalian cell toxicity, genotoxicity, and transformation. Bioelectromagnetics Suppl 6:S196-S213, 2003. (review)

The published in vitro literature relevant to the issue of the possible induction of toxicity, genotoxicity, and transformation of mammalian cells due to radiofrequency field (RF) exposure is examined. In some instances, information about related in vivo studies is presented. The review is from the perspective of technical merit and also biological consistency, especially with regard to those publications reporting a positive effect. The weight of evidence available indicates that, for a variety of frequencies and modulations with both short and long exposure times, at exposure levels that do not (or in some instances do) heat the biological sample such that there is a measurable increase in temperature, RF exposure does not induce (a). DNA strand breaks, (b). chromosome aberrations, (c). sister chromatid exchanges (SCEs), (d). DNA repair synthesis, (e). phenotypic mutation, or (f). transformation (cancer-like changes). While there is limited experimental evidence that RF exposure induces micronuclei formation, there is abundant evidence that it does not. There is some evidence that RF exposure does not induce DNA excision repair, suggesting the absence of base damage. There is also evidence that RF exposure does not inhibit excision repair after the induction of thymine dimers by UV exposure, as well as evidence that indicates that RF is not a co-carcinogen or a tumor promoter. The article is in part a tutorial, so that the reader can consider similarities and discrepancies between reports of RF-induced effects relative to one another.

(NE) Meltz ML, Eagan P, Erwin DN, Proflavin and microwave radiation: absence of a mutagenic interaction. Bioelectromagnetics 11(2):149-157, 1990. (VT, AE, GT, IX)

The potential ability of radiofrequency electromagnetic radiation (RFR) in the microwave range

to induce mutagenesis, chromosomal aberrations, and sister chromatid exchanges in mammalian cells is being explored in our laboratories. In addition, we have also been examining the ability of simultaneous exposure to RFR and chemical mutagens to alter the genotoxic damage induced by chemical mutagens acting alone. We have performed experiments to determine whether there is an interaction between 2.45-GHz, pulsed-wave, RFR and proflavin, a DNA-intercalating drug. The endpoint studied was forward mutation at the thymidine kinase locus in L5178Y mouse leukemic cells. Any effect on the size distribution of the resulting colonies of mutated cells was also examined. The exposures were performed at net forward powers of 500 or 600 W, resulting in a specific absorption rate (SAR) of approximately 40 W/kg. The culture-medium temperature reached a 3 degrees C maximal increase during the 4-h exposure; appropriate 37 degrees C and convection-heating temperature controls (TC) were performed. In no case was there any indication of a statistically significant increase in the induced mutant frequency due to the simultaneous exposure to RFR and proflavin, as compared with the proflavin exposures alone. There was also no indication of any change in the colony-size distribution of the resulting mutant colonies, neither, and there was no evidence in these experiments of any mutagenic action by the RFR exposure alone.

(E)Migdal P, Bieńkowski P, Cebrat M, Berbeć E, Plotnik M, Murawska A, Sobkiewicz P, Łaszkiewicz A, Latarowski K. Exposure to a 900 MHz electromagnetic field induces a response of the honey bee organism on the level of enzyme activity and the expression of stress-related genes. PLoS One 18(5):e0285522, 2023. (VO, AE, GE)

There are many artificial sources of radiofrequency electromagnetic field (RF-EMF) in the environment, with a value between 100 MHz and 6 GHz. The most frequently used signal is with a frequency of around 900 MHz. The direction of these changes positively impacts the quality of life, enabling easy communication from almost anywhere in the world. All living organisms in the world feel the effects of the electromagnetic field on them. The observations regarding the influence of a RF-EMF on honey bees, describing the general impact of RF-EMF on the colony and/or behavior of individual bees, such as reduction in the number of individuals in colonies, extended homing flight duration, decrease in breeding efficiency, changes in flight direction (movement of bees toward the areas affected by RF-EMF), increase in the intensity and frequency of sounds characteristic for those announcing the impending danger. In this work, we describe the changes in the levels of some of the stress-related markers in honey bees exposed to varying intensities and duration of RF-EMF. One-day-old honeybee worker bees were used for the study. The bees were randomly assigned to 9 experimental groups which were exposed to the following 900 MHz EMF intensities: 12 V/m, 28 V/m, and 61 V/m for 15 min, 1 h and 3 h. The control group was not exposed to the RF-EMF. Each experimental group consisted of 10 cages in which were 100 bees. Then, hemolymph was collected from the bees, in which the activity was assessed AST, ALT, ALP, GGTP, and level of nonenzymatic antioxidants albumin, creatinine, uric acid, and urea. Bees were also collected for the analysis of rps5, ppo, hsp10, hsp70, hsp90, and vitellogenin gene expression. Our study shows that exposure to a 900 MHz electromagnetic field induces a response in the honey bees that can be detected in the level of enzyme activity and the expression of stress-related genes. The response is similar to the one previously

described as a result of exposition to UVB irradiation and most likely cannot be attributed to increased temperature.

(E) Mildažienė V, Aleknavičiūtė V, Žūkienė R, Paužaitė G, Naučienė Z, Filatova I, Lyushkevich V, Haimi P, Tamošiūnė I, Baniulis D. Treatment of common sunflower (Helianthus annus L.) seeds with radio-frequency electromagnetic field and cold plasma induces changes in seed phytohormone balance, seedling development and leaf protein expression. Sci Rep. 9(1):6437, 2019. (VT, AE, GE)

Treatment of plant seeds with electromagnetic fields or non-thermal plasmas aims to take advantage of plant functional plasticity towards stimulation of plant agricultural performance. In this study, the effects of pre-sowing seed treatment using 200 Pa vacuum (7 min), 5.28 MHz radio-frequency cold plasma (CP -2, 5, and 7 min) and electromagnetic field (EMF -5, 10, 15 min) on seed germination kinetics, content of phytohormones, morphometric parameters of seedlings and leaf proteome were assessed. CP 7 min and EMF 15 min treatments caused 19-24% faster germination in vitro; germination in the substrate was accelerated by vacuum (9%) and EMF 15 min (17%). The stressors did not change the seed germination percentage, with exception of EMF 5 min treatment that caused a decrease by 7.5%. Meanwhile both CP 7 min and EMF 15 min treatments stimulated germination, but the EMF treatment resulted in higher weight of leaves. Stressor-specific changes in phytohormone balance were detected in seeds: vacuum treatment decreased zeatin amount by 39%; CP treatments substantially increased gibberellin content, but other effects strongly varied with the treatment duration; the abscisic acid content was reduced by 55-60% after the EMF treatment. Analysis of the proteome showed that short exposure of seeds to the EMF or CP induced a similar long-term effect on gene expression in leaves, mostly stimulating expression of proteins involved in photosynthetic processes and their regulation.

(E) Millenbaugh NJ, Roth C, Sypniewska R, Chan V, Eggers JS, Kiel JL, Robert V Blystone RV, Mason PA. Gene expression changes in the skin of rats induced by prolonged 35 GHz millimeter-wave exposure. Radiat Res 169(3):288-300, 2008. (VO, AE, GE)

To better understand the cellular and molecular responses to overexposure to millimeter waves, alterations in the gene expression profile and histology of skin after exposure to 35 GHz radiofrequency radiation were investigated. Rats were subjected to sham exposure, to 42 degrees C environmental heat, or to 35 GHz millimeter waves at 75 mW/cm(2). Skin samples were collected at 6 and 24 h after exposure for Affymetrix GeneChip analysis. The skin was harvested from a separate group of rats at 3-6 h or 24-48 h after exposure for histopathology analysis. Microscopic findings observed in the dermis of rats exposed to 35 GHz millimeter waves included aggregation of neutrophils in vessels, degeneration of stromal cells, and breakdown of collagen. Changes were detected in 56 genes at 6 h and 58 genes at 24 h in the millimeter-wave-exposed rats. Genes associated with regulation of transcription, protein folding, oxidative stress, immune response, and tissue matrix turnover were affected at both times. At 24 h, more genes

related to extracellular matrix structure and chemokine activity were altered. Up-regulation of Hspa1a, Timp1, S100a9, Ccl2 and Angptl4 at 24 h by 35 GHz millimeter-wave exposure was confirmed by real-time RT-PCR. These results obtained from histopathology, microarrays and RT-PCR indicate that prolonged exposure to 35 GHz millimeter waves causes thermally related stress and injury in skin while triggering repair processes involving inflammation and tissue matrix recovery.

(NE) Miyakoshi, J., Yoshida, M., Tarusawa, Y., et al. Effects of high- frequency electromagnetic fields on DNA strand breaks using comet assay method. Electrical Eng. Japan. 141: 9–15, 2002. (VT, AS, GT)

We investigated whether exposure to high-frequency electromagnetic fields causes DNA damage in cells, using the alkaline comet assay. The exposure device made for this study used a TE01 circular waveguide operating at a frequency of 2.45 GHz. Cells of the human brain tumor-derived MO54 cell line were exposed to an electromagnetic field (input power: 7.8 W, average SAR in the middle well of an annular culture plate: CW 50 W/kg) for 2 hours and the tail moments of the cells in the inner, middle, and outer wells of the plate were compared with those of sham-exposed cells. There was no significant difference between the high-frequency electromagnetic field-exposed groups and the sham-exposed groups. Three studies performed under the same conditions gave similar results. Next, cells were exposed to a stronger electromagnetic field (input power: 13 W, average SAR in the middle well: CW 100 W/kg) for 2 hours and compared with sham-exposed cells. There was also no significant difference in the tail moments of cells in the inner, middle, and outer wells of the plate in the high-frequency electromagnetic field-exposed groups and the sham-exposed groups. These findings suggest that a high-frequency electromagnetic field does not cause direct DNA damage, and does not induce DNA strand breaks, even at a SAR of 100 W/kg.

(NE) Miyakoshi J, Tonomura H, Koyama S, Narita E, Shinohara N. Effects of Exposure to 5.8 GHz Electromagnetic Field on Micronucleus Formation, DNA Strand Breaks, and Heat Shock Protein Expressions in Cells Derived From Human Eye. IEEE Trans Nanobioscience 18(2):257-260, 2019. (VT, AE, GT)

In the near future, electrification will be introduced to heavy-duty vehicles and passenger cars. However, the wireless power transfer (WPT) requires high energy levels, and the suitability of various types of WPT systems must be assessed. This paper describes a method for solving technical and safety issues associated with this technology. We exposed human corneal epithelial (HCE-T) cells derived from the human eye to 5.8-GHz electromagnetic fields for 24 h. We observed no statistically significant increase in micronucleus (MN) frequency in cells exposed to a 5.8-GHz field at 1 mW/cm^2 (the general public level in ICNIRP) relative to sham-exposed or incubator controls. Similarly, the DNA strand breaks, and the expression of heat shock protein (Hsp) Hsp27, Hsp70, and Hsp 90α exhibited no statistically significant effects as a result of exposure. These results indicate that the exposure to 5.8-GHz electromagnetic fields at 1 mW/cm^2 for 24 h has little or no effect on micronucleus formation, DNA strand breaks, and Hsp expression in human eye cells.

(E) Moghadasi N, Alimohammadi I, Variani AS, Ashtarinezhad A. The Effect of Mobile Radiation on the Oxidative Stress Biomarkers in Pregnant Mice. J Family Reprod Health. 15(3):172-178, 2021. (VO, LE, GT, OX)

Objective: Due to the growing use of communication instruments such as cell phones and wireless devices, there is growing public concern about possible harmful effects, especially in sensitive groups such as pregnant women. This study aimed to investigate the oxidative stress induced by exposure to 900 MHz mobile phone radiation and the effect of vitamin C intake on reducing possible changes in pregnant mice. Materials and methods: Twenty-one pregnant mice were divided into three groups (control, mobile radiation-exposed, and mobile radiation plus with vitamin C intake co-exposed (200 mg /kg)). The mice in exposure groups were exposed to 900 MHz, 2 watts, and a power density of 0.045 µw /cm2 mobile radiation for eight hours/day for ten consecutive days. After five days of rest, MDA (Malondialdehyde), 8-OHdG (8-hydroxy-2' -deoxyguanosine), and TAC (Total Antioxidant Capacity) levels were measured in the blood of animals. The results were analyzed by SPSS.22.0 software. Results: The results showed that exposure to mobile radiation increased MDA (P=0.002), and 8-OHdG (P=0.001) significantly and decreased Total Antioxidant Capacity in the exposed groups (P=0.001). Taking vitamin C inhibited the significant increase in MDA and 8-OHdG levels in exposed groups. Conclusion: Although exposure to mobile radiation can cause oxidative stress in the blood of pregnant mice, vitamin C as an antioxidant can prevent it.

(E) Mokarram P, Sheikhi M, Mortazavi SMJ, Saeb S, Shokrpour N. Effect of Exposure to 900 MHz GSM Mobile Phone Radiofrequency Radiation on Estrogen Receptor Methylation Status in Colon Cells of Male Sprague Dawley Rats. J Biomed Phys Eng 7(1):79-86, 2017. (VO, LE, GE, DE, RP, EP)

Background: Over the past several years, the rapidly increasing use of mobile phones has raised global concerns about the biological effects of exposure to radiofrequency (RF) radiation. Numerous studies have shown that exposure to electromagnetic fields (EMFs) can be associated with effects on the nervous, endocrine, immune, cardiovascular, hematopoietic and ocular systems. In spite of genetic diversity, the onset and progression of cancer can be controlled by epigenetic mechanisms such as gene promoter methylation. There are extensive studies on the epigenetic changes of the tumor suppressor genes as well as the identification of methylation biomarkers in colorectal cancer. Some studies have revealed that genetic changes can be induced by exposure to RF radiation. However, whether or not RF radiation is capable of inducing epigenetic alteration has not been clarified yet. To date, no study has been conducted on the effect of radiation on epigenetic alterations in colorectal cancer (CRC). Several studies have also shown that methylation of estrogen receptor α (ERα), MYOD, MGMT, SFRP2 and P16 play an important role in CRC. It can be hypothesized that RF exposure can be a reason for the high incidence of CRC in Iran. This study aimed to investigate whether epigenetic pattern of ERα is susceptible to RF radiation and if RF radiation can induce radioadaptive response as epigenetic changes after receiving the challenge dose (γ-ray). **Material and method:** 40 male Sprague-Dawley rats were divided into 4 equal groups (Group I: exposure to RF radiation of a GSM cell

phone for 4 hours and sacrificed after 24 hours; Group II: RF exposure for 4 hours, exposure to Co-60 gamma radiation (3 Gy) after 24 hours and sacrificed after 72 hrs; Group III: only 3Gy gamma radiation; Group 4: control group). DNA from colon tissues was extracted to evaluate the methylation status by methylation specific PCR. **Results:** Our finding showed that exposure to GSM cell phone RF radiation was capable of altering the pattern of ERα gene methylation compared to that of non-exposed controls. Furthermore, no adaptive response phenomenon was induced in the pattern of ERα gene methylation after exposure to the challenging dose of Co-60 γ-rays. **Conclusion:** It can be concluded that exposure to RF radiation emitted by GSM mobile phones can lead to epigenetic detrimental changes in ERα promoter methylation pattern.

(NE) Nakatani-Enomoto S, Okutsu M, Suzuki S, Suganuma R, Groiss SJ, Kadowaki S, et al. Effects of 1950MHz W-CDMA-Like signal on human spermatozoa. Bioelectromagentics. 37: 373- 381, 2016. (VT, AE, GT)

There are growing concerns about how electromagnetic waves (EMW) emitted from mobile phones affect human spermatozoa. Several experiments have suggested harmful effects of EMW on human sperm quality, motility, velocity, or the deoxyribonucleic acid (DNA) of spermatozoa. In this study, we analyzed the effects on human spermatozoa (sperm motility and kinetic variables) induced by 1 h of exposure to 1950 MHz Wideband Code Division Multiple Access (W-CDMA)-like EMW with specific absorption rates of either 2.0 or 6.0 W/kg, using a computer-assisted sperm analyzer system. We also measured the percentage of 8-hydroxy-2'-deoxyguanosine (8-OHdG) positive spermatozoa with flow cytometry to evaluate damage to DNA. No significant differences were observed between the EMW exposure and the sham exposure in sperm motility, kinetic variables, or 8-OHdG levels. We conclude that W-CDMA-like exposure for 1 h under temperature-controlled conditions has no detectable effect on normal human spermatozoa. Differences in exposure conditions, humidity, temperature control, baseline sperm characteristics, and age of donors may explain inconsistency of our results with several previous studies.

(E) Narasimhan V, Huh WK, Altered restriction patterns of microwave irradiated lambdaphage DNA. Biochem Int 25(2):363-370, 1991.(VT, AE, GT)

Samples of lambdaphage DNA exposed to short pulses of microwave irradiation were subjected to restriction fragmentation by Eco RI and Bam HI. Eco RI digests of microwaved DNA samples yielded three additional fragments ranging in base pair lengths between 24,226 and 7,421 besides the six expected fragments. While Bam HI digests of the microwaved samples did not yield any additional fragments, mobilities of the Bam HI fragments from the microwaved DNA samples were slower and the bands were broader in comparison to those from native samples. We attribute these altered restriction patterns to the conformational anomolies in DNA resulting from single strand breaks and localized strand separations induced by microwave irradiation.

(E) Nikolova T, Czyz J, Rolletschek A, Blyszczuk P, Fuchs J, Jovtchev G, Schuderer J, Kuster N, Wobus AM. Electromagnetic fields affect transcript levels of apoptosis-related

genes in embryonic stem cell-derived neural progenitor cells. FASEB J 19(12):1686-1688, 2005. (VT, AE, GT, GE, WS)

Mouse embryonic stem (ES) cells were used as an experimental model to study the effects of electromagnetic fields (EMF). ES-derived nestin-positive neural progenitor cells were exposed to extremely low frequency EMF simulating power line magnetic fields at 50 Hz (ELF-EMF) and to radiofrequency EMF simulating the Global System for Mobile Communication (GSM) signals at 1.71 GHz (RF-EMF). Following EMF exposure, cells were analyzed for transcript levels of cell cycle regulatory, apoptosis-related, and neural-specific genes and proteins; changes in proliferation; apoptosis; and cytogenetic effects. Quantitative RT-PCR analysis revealed that ELF-EMF exposure to ES-derived neural cells significantly affected transcript levels of the apoptosis-related bcl-2, bax, and cell cycle regulatory "growth arrest DNA damage inducible" GADD45 genes, whereas mRNA levels of neural-specific genes were not affected. RF-EMF exposure of neural progenitor cells resulted in down-regulation of neural-specific Nurr1 and in up-regulation of bax and GADD45 mRNA levels. Short-term RF-EMF exposure for 6 h, but not for 48 h, resulted in a low and transient increase of DNA double-strand breaks. No effects of ELF- and RF-EMF on mitochondrial function, nuclear apoptosis, cell proliferation, and chromosomal alterations were observed. We may conclude that EMF exposure of ES-derived neural progenitor cells transiently affects the transcript level of genes related to apoptosis and cell cycle control. However, these responses are not associated with detectable changes of cell physiology, suggesting compensatory mechanisms at the translational and posttranslational level.

(E) Nittby H, Widegren B, Krogh M, Grafström G, Berlin H, Rehn G, Eberhardt JL, Malmgren L, Persson BRR, Salford L. Exposure to radiation from global system for mobile communications at 1,800 MHz significantly changes gene expression in rat hippocampus and cortex. Environmentalist 28(4), 458-465, 2008. (VO, AE, GE, LI)

We have earlier shown that radio frequency electromagnetic fields can cause significant leakage of albumin through the blood–brain barrier of exposed rats as compared to non-exposed rats, and also significant neuronal damage in rat brains several weeks after a 2 h exposure to a mobile phone, at 915 MHz with a global system for mobile communications (GSM) frequency modulation, at whole-body specific absorption rate values (SAR) of 200, 20, 2, and 0.2 mW/kg. We have now studied whether 6 h of exposure to the radiation from a GSM mobile test phone at 1,800 MHz (at a whole-body SAR-value of 13 mW/kg, corresponding to a brain SAR-value of 30 mW/kg) has an effect upon the gene expression pattern in rat brain cortex and hippocampus—areas where we have observed albumin leakage from capillaries into neurons and neuronal damage. Microarray analysis of 31,099 rat genes, including splicing variants, was performed in cortex and hippocampus of 8 Fischer 344 rats, 4 animals exposed to global system for mobile communications electromagnetic fields for 6 h in an anechoic chamber, one rat at a time, and 4 controls kept as long in the same anechoic chamber without exposure, also in this case one rat at a time. Gene ontology analysis (using the gene ontology categories biological processes, molecular functions, and cell components) of the differentially expressed genes of the exposed animals versus the control group revealed the following highly significant altered gene categories in both cortex and hippocampus: extracellular region, signal transducer activity, intrinsic to membrane, and integral to membrane. The fact that most of these categories are connected with

membrane functions may have a relation to our earlier observation of albumin transport through brain capillaries.

(E) Nylund R, Leszczynski D. Mobile phone radiation causes changes in gene and protein expression in human endothelial cell lines and the response seems to be genome- and proteome-dependent. Proteomics 6:4769-4780, 2006. (VT, AE, GE, CS)

We have examined in vitro cell response to mobile phone radiation (900 MHz GSM signal) using two variants of human endothelial cell line: EA.hy926 and EA.hy926v1. Gene expression changes were examined in three experiments using cDNA Expression Arrays and protein expression changes were examined in ten experiments using 2-DE and PDQuest software. Obtained results show that gene and protein expression were altered, in both examined cell lines, in response to one hour mobile phone radiation exposure at an average specific absorption rate of 2.8 W/kg. However, the same genes and proteins were differently affected by the exposure in each of the cell lines. This suggests that the cell response to mobile phone radiation might be genome- and proteome-dependent. Therefore, it is likely that different types of cells and from different species might respond differently to mobile phone radiation or might have different sensitivity to this weak stimulus. Our findings might also explain, at least in part, the origin of discrepancies in replication studies between different laboratories.

(E) Odacı E, Hancı H, Yuluğ E, Türedi S, Aliyazıcıoğlu Y, Kaya H, Çolakoğlu S. Effects of prenatal exposure to a 900 MHz electromagnetic field on 60-day-old rat testis and epididymal sperm quality. Biotech Histochem. 91(1):9-19, 2016. (VO, CE, GT, OX, DE, RP)

We investigated the effects of exposure in utero to a 900 megahertz (MHz) electromagnetic field (EMF) on 60-day-old rat testis and epididymis. Pregnant rats were divided into control (CG; no treatment) and EMF (EMFG) groups. The EMFG was exposed to 900 MHz EMF for 1 h each day during days 13 - 21 of pregnancy. Newborn rats were either newborn CG (NCG) or newborn EMF groups (NEMFG). On postnatal day 60, a testis and epididymis were removed from each animal. Epididymal semen quality, and lipid and DNA oxidation levels, apoptotic index and histopathological damage to the testis were compared. We found a higher apoptotic index, greater DNA oxidation levels and lower sperm motility and vitality in the NEMFG compared to controls. Immature germ cells in the seminiferous tubule lumen, and altered seminiferous tubule epithelium and seminiferous tubule structure also were observed in hematoxylin and eosin stained sections of NEMFG testis. Nuclear changes that indicated apoptosis were identified in TUNEL stained sections and large numbers of apoptotic cells were observed in most of the seminiferous tubule epithelium in the NEMFG. Sixty-day-old rat testes exposed to 900 MHz EMF exhibited altered sperm quality and biochemical characteristics.

(E) Ohtani S, Ushiyama A, Maeda M, Hattori K, Kunugita N, Wang J, Ishii K. Exposure time-dependent thermal effects of radiofrequency electromagnetic field exposure on the whole body of rats. J Toxicol Sci. 41(5):655-666, 2016. (VO, AE, GE) (thermal effect)

We investigated the thermal effects of radiofrequency electromagnetic fields (RF-EMFs) on the variation in core temperature and gene expression of some stress markers in rats. Sprague-

Dawley rats were exposed to 2.14 GHz wideband code division multiple access (W-CDMA) RF signals at a whole-body averaged specific absorption rate (WBA-SAR) of 4 W/kg, which causes behavioral disruption in laboratory animals, and 0.4 W/kg, which is the limit for the occupational exposure set by the International Commission on Non-Ionizing Radiation Protection guideline. It is important to understand the possible in vivo effects derived from RF-EMF exposures at these intensities. Because of inadequate data on real-time core temperature analyses using free-moving animal and the association between stress and thermal effects of RF-EMF exposure, we analyzed the core body temperature under nonanesthetic condition during RF-EMF exposure. The results revealed that the core temperature increased by approximately 1.5°C compared with the baseline and reached a plateau till the end of RF-EMF exposure. Furthermore, we analyzed the gene expression of heat-shock proteins (Hsp) and heat-shock transcription factors (Hsf) family after RF-EMF exposure. At WBA-SAR of 4 W/kg, some Hsp and Hsf gene expression levels were significantly upregulated in the cerebral cortex and cerebellum following exposure for 6 hr/day but were not upregulated after exposure for 3 hr/day. On the other hand, there was no significant change in the core temperature and gene expression at WBA-SAR of 0.4 W/kg. Thus, 2.14-GHz RF-EMF exposure at WBA-SAR of 4 W/kg induced increases in the core temperature and upregulation of some stress markers, particularly in the cerebellum.

(NE) Ono T, Saito Y, Komura J, Ikehata H, Tarusawa Y, Nojima T, Goukon K, Ohba Y, Wang J, Fujiwara O, Sato R. Absence of mutagenic effects of 2.45 GHz radiofrequency exposure in spleen, liver, brain, and testis of lacZ-transgenic mouse exposed in utero. Tohoku J Exp Med. 202(2):93-103, 2004. (VO, LE, GT)

A possible mutagenic effect of 2.45 GHz radiofrequency exposure was examined using lacZ-transgenic Muta mice. Pregnant animals were exposed intermittently at a whole-body averaged specific absorption rate of 0.71 W/kg (10 seconds on, 50 seconds off which is 4.3 W/kg during the 10 seconds exposure). Offspring that were exposed in utero for 16 hours a day, from the embryonic age of 0 to 15 days, were examined at 10 weeks of age. To minimize thermal effects, the exposure was given in repeated bursts of 10 seconds of exposure followed by 50 seconds of no exposure. Mutation frequencies at the lacZ gene in spleen, liver, brain, and testis were similar to those observed in non-exposed mice. Quality of mutation assessed by sequencing the nucleotides of mutant DNAs revealed no appreciable difference between exposed and non-exposed samples. The data suggest that the level of radiofrequency exposure studied is not mutagenic when administered in utero in short repeated bursts.

(E) Ozgur E, Guler G, Kismali G, Seyhan N. Mobile phone radiation alters proliferation of hepatocarcinoma cells. Cell Biochem Biophys. 70(2):983-991, 2014. (GT, WS)

This study investigated the effects of intermittent exposure (15 min on, 15 min off for 1, 2, 3, or 4 h, at a specific absorption rate of 2 W/kg) to enhanced data rates for global system for mobile communication evolution-modulated radiofrequency radiation (RFR) at 900- and 1,800-MHz frequencies on the viability of the Hepatocarcinoma cells (Hep G2). Hep G2 cell proliferation was measured by a colorimetric assay based on the cleavage of the tetrazolium salt WST-1 by mitochondrial dehydrogenases in viable cells. Cell injury was evaluated by analyzing the levels of lactate dehydrogenase (LDH) and glucose released from lysed cells into the culture medium.

Morphological observation of the nuclei was carried out by 4',6-diamidino-2-phenylindole (DAPI) staining using fluorescence microscopy. In addition, TUNEL assay was performed to confirm apoptotic cell death. It was observed that cell viability, correlated with the LDH and glucose levels, changed according to the frequency and duration of RFR exposure. Four-hour exposure produced more pronounced effects than the other exposure durations. 1,800-MHz RFR had a larger impact on cell viability and Hep G2 injury than the RFR at 900 MHz. Morphological observations also supported the biochemical results indicating that most of the cells showed irregular nuclei pattern determined by using the DAPI staining, as well as TUNEL assay which shows DNA damage especially in the cells after 4 h of exposure to 1,800-MHz RFR. Our results indicate that the applications of 900- and 1,800-MHz (2 W/kg) RFR cause to decrease in the proliferation of the Hep G2 cells after 4 h of exposure. Further studies will be conducted on other frequency bands of RFR and longer duration of exposure.

(E) Pacini S, Ruggiero M, Sardi I, Aterini S, Gulisano F, Gulisano M. Exposure to global system for mobile communication (GSM) cellular phone radiofrequency alters gene expression, proliferation, and morphology of human skin fibroblasts. Oncol Res 13: 19-24, 2002. (VT, AE, GE)

Human skin fibroblasts were exposed to global system for mobile communication (GSM) cellular phone radiofrequency for 1 h. GSM exposure induced alterations in cell morphology and increased the expression of mitogenic signal transduction genes (e.g., MAP kinase kinase 3, G2/mitotic-specific cyclin G1), cell growth inhibitors (e.g., transforming growth factor-beta), and genes controlling apoptosis (e.g., bax). A significant increase in DNA synthesis and intracellular mitogenic second messenger formation matched the high expression of MAP kinase family genes. These findings show that these electromagnetic fields have significant biological effects on human skin fibroblasts.

(E) Panagopoulos DJ, Chavdoula ED, Nezis IP, Margaritis LH. Cell death induced by GSM 900-MHz and DCS 1800-MHz mobile telephony radiation. Mutat Res 626:69-78, 2007. (VO, LE, GT, RP)

In the present study, the TUNEL (Terminal deoxynucleotidetransferasedUTP Nick End Labeling) assay - a well known technique widely used for detecting fragmented DNA in various types of cells - was used to detect cell death (DNA fragmentation) in a biological model, the early and mid stages of oogenesis of the insect Drosophila melanogaster. The flies were exposed in vivo to either GSM 900-MHz (Global System for Mobile telecommunications) or DCS 1800-MHz (Digital Cellular System) radiation from a common digital mobile phone, for few minutes per day during the first 6 days of their adult life. The exposure conditions were similar to those to which a mobile phone user is exposed, and were determined according to previous studies of ours [D.J Panagopoulos, A. Karabarbounis, L.H. Margaritis, Effect of GSM 900-MHz mobile phone radiation on the reproductive capacity of D. melanogaster, Electromagn. Biol Med 23 (2004) 29-43; D.J Panagopoulos, N. Messini, A. Karabarbounis, A.L. Philippetis, L.H. Margaritis, Radio frequency electromagnetic radiation within "safety levels" alters the physiological function of insects, in: P. Kostarakis, P. Stavroulakis (Eds.), Proceedings of the Millennium International Workshop on Biological Effects of Electromagnetic Fields, Heraklion, Crete, Greece, October 17-20, 2000, pp. 169-175, ISBN: 960-86733-0-5; D.J Panagopoulos,

L.H. Margaritis, Effects of electromagnetic fields on the reproductive capacity of D. melanogaster, in: P. Stavroulakis (Ed.), Biological Effects of Electromagnetic Fields, Springer, 2003, pp. 545-578], which had shown a large decrease in the oviposition of the same insect caused by GSM radiation. Our present results suggest that the decrease in oviposition previously reported, is due to degeneration of large numbers of egg chambers after DNA fragmentation of their constituent cells, induced by both types of mobile telephony radiation. Induced cell death is recorded for the first time, in all types of cells constituting an egg chamber (follicle cells, nurse cells and the oocyte) and in all stages of the early and mid-oogenesis, from germarium to stage 10, during which programmed cell death does not physiologically occur. Germarium and stages 7-8 were found to be the most sensitive developmental stages also in response to electromagnetic stress induced by the GSM and DCS fields and, moreover, germarium was found to be even more sensitive than stages 7-8.

(E) Panagopoulos DJ. Chromosome damage in human cells induced by UMTS mobile telephony radiation. Gen Physiol Biophys. 38(5):445-454, 2019. (VT, AE, GT)

Environmental exposure to modern microwave telecommunication electromagnetic fields (EMFs) has increased to unprecedented levels with consequent health complaints and concerns. Many studies have already reported genotoxic effects on a variety of organisms and cell/tissue types. Human peripheral blood lymphocytes from six healthy donors were stimulated for mitosis and exposed to microwave EMF of Universal Mobile Telecommunications System (UMTS) or third generation (3G) mobile telephony (MT) EMF/radiation emitted by a commercially available mobile phone handset. Lymphocytes exposed during the G2 phase of the cell division cycle and observed at metaphase, exhibited chromatid-type aberrations (gaps and breaks) at highly significant percentages - up to 275% - compared to the control (sham-exposed) samples. Each subject exhibited a different sensitivity to the microwave exposure. Moreover, the percentages of aberrations in the control samples among subjects were different due to genetic and environmental factors. The MT EMF exposure induced mainly achromatic lesions (gaps), and secondarily terminal deletions (breaks) in a smaller degree. In conclusion, the present study shows that microwave 3G MT EMF/radiation - within the current exposure limits - has significant genotoxic action on human cells, and human exposure to this EMF/radiation should be kept at levels as low as possible.

(E) Panagopoulos, D.J. Comparing chromosome damage induced by mobile telephony radiation and a high caffeine dose: Effect of combination and exposure duration. Gen. Physiol. Biophys. 39(6):531-544, 2020. (VT, AE, GT, IX)

I recently reported induction of chromatid-type aberrations in human peripheral blood lymphocytes after a single 15 min exposure to universal mobile telecommunications system (UMTS) mobile telephony (MT) electromagnetic field (EMF) from a mobile phone. Lymphocytes from six healthy subjects were stimulated for mitosis, and exposed during the G2/M phase at 1 cm distance from the handset during an active phone call in "talk" mode. The same type of cells from the same subjects treated with a high caffeine dose (~ 290 times above the permissible single dose for an adult human) exhibited the same type of aberrations in a little smaller but comparable degree. The combination of this caffeine dose and the 15 min MT EMF exposure increased dramatically the number of aberrations in all subjects. The combined effect

increased almost linearly with increasing duration of exposure to the MT EMF. Thus, MT EMF exposure ~ 136 times below the official limit (ICNIRP 2020) exerts a genotoxic action even greater than that of a caffeine dose ~ 290 times above the corresponding limit. Therefore, with a reasonable approximation, the limit for MT EMFs should be lowered by at least ~ 4×10^4 times (136 × 290) for short-term exposures, and ~ 4×10^6 times for long-term exposures.

(E) Pandey N, Giri S, Das S, Upadhaya P. Radiofrequency radiation (900 MHz)-induced DNA damage and cell cycle arrest in testicular germ cells in swiss albino mice. Toxicol Ind Health. 33(4):33-384, 2017. (VO, LE, GT, OX, RP)

Even though there are contradictory reports regarding the cellular and molecular changes induced by mobile phone emitted radiofrequency radiation (RFR), the possibility of any biological effect cannot be ruled out. In view of a widespread and extensive use of mobile phones, this study evaluates alterations in male germ cell transformation kinetics following RFR exposure and after recovery. Swiss albino mice were exposed to RFR (900 MHz) for 4 h and 8 h duration per day for 35 days. One group of animals was terminated after the exposure period, while others were kept for an additional 35 days post-exposure. RFR exposure caused depolarization of mitochondrial membranes resulting in destabilized cellular redox homeostasis. Statistically significant increases in the damage index in germ cells and sperm head defects were noted in RFR-exposed animals. Flow cytometric estimation of germ cell subtypes in mice testis revealed 2.5-fold increases in spermatogonial populations with significant decreases in spermatids. Almost fourfold reduction in spermatogonia to spermatid turnover (1C:2C) and three times reduction in primary spermatocyte to spermatid turnover (1C:4C) was found indicating arrest in the premeiotic stage of spermatogenesis, which resulted in loss of post-meiotic germ cells apparent from testis histology and low sperm count in RFR-exposed animals. Histological alterations such as sloughing of immature germ cells into the seminiferous tubule lumen, epithelium depletion and maturation arrest were also observed. However, all these changes showed recovery to varied degrees following the post-exposure period indicating that the adverse effects of RFR on mice germ cells are detrimental but reversible. To conclude, RFR exposure-induced oxidative stress causes DNA damage in germ cells, which alters cell cycle progression leading to low sperm count in mice.

(E) Pandey N, Giri S. Melatonin attenuates radiofrequency radiation (900 MHz)-induced oxidative stress, DNA damage and cell cycle arrest in germ cells of male Swiss albino mice.Toxicol Ind Health. 34(5):315-327, 2018. (VO, LE, GT, OX, RP)

Increasing male infertility of unknown aetiology can be associated with environmental factors. Extensive use of mobile phones has exposed the general population to unprecedented levels of radiofrequency radiations (RFRs) that may adversely affect male reproductive health. Therefore, the present study investigated the effect of RFR Global System for Mobile communication (GSM) type, 900 MHz and melatonin supplementation on germ cell development during spermatogenesis. Swiss albino mice were divided into four groups. One group received RFR exposure for 3 h twice/day for 35 days and the other group received the same exposure but with melatonin (N-acetyl-5-methoxytryptamine) (MEL; 5 mg/kg bw/day). Two other groups received

only MEL or remain unexposed. Sperm head abnormality, total sperm count, biochemical assay for lipid peroxides, reduced glutathione, superoxide dismutase activity and testis histology were evaluated. Additionally, flow cytometric evaluation of germ cell subtypes and comet assay were performed in testis. Extensive DNA damage in germ cells of RFR-exposed animals along with arrest in pre-meiotic stages of spermatogenesis eventually leading to low sperm count and sperm head abnormalities were observed. Furthermore, biochemical assays revealed excess free radical generation resulting in histological and morphological changes in testis and germ cells morphology, respectively. However, these effects were either diminished or absent in RFR-exposed animals supplemented with melatonin. Hence, it can be concluded that melatonin inhibits pre-meiotic spermatogenesis arrest in male germ cells through its anti-oxidative potential and ability to improve DNA reparative pathways, leading to normal sperm count and sperm morphology in RFR-exposed animals.

(NE) Paparini A, Rossi P, Gianfranceschi G, Brugaletta V, Falsaperla R, De Luca P, Romano Spica V. No evidence of major transcriptional changes in the brain of mice exposed to 1800 MHz GSM signal. Bioelectromagnetics. 29(4):312-323, 2008. (VO, AE, GE)

To analyze possible effects of microwaves on gene expression, mice were exposed to global system for mobile communication (GSM) 1800 MHz signal for 1 h at a whole body SAR of 1.1 W/kg. Gene expression was studied in the whole brain, where the average SAR was 0.2 W/kg, by expression microarrays containing over 22,600 probe sets. Comparison of data from sham and exposed animals showed no significant difference in gene expression modulation. However, when less stringent constraints were adopted to analyze microarray results, 75 genes were found to be modulated following exposure. Forty-two probes showed fold changes ranging from 1.5 to 2.8, whereas 33 were down-regulated from 0.67- to 0.29-fold changes, but these differences in gene expression were not confirmed by real-time PCR. Under these specific limited conditions, no consistent indication of gene expression modulation in whole mouse brain was found associated to GSM 1800 MHz exposure.

(E) Paulraj R, Behari J. Single strand DNA breaks in rat brain cells exposed to microwave radiation. Mutat Res 596:76-80, 2006. (VO, LE, GT)

This investigation concerns with the effect of low intensity microwave (2.45 and 16.5GHz, SAR 1.0 and 2.01W/kg, respectively) radiation on developing rat brain. Wistar rats (35 days old, male, six rats in each group) were selected for this study. These animals were exposed for 35 days at the above mentioned frequencies separately in two different exposure systems. After the exposure period, the rats were sacrificed and the whole brain tissue was dissected and used for study of single strand DNA breaks by micro gel electrophoresis (comet assay). Single strand DNA breaks were measured as tail length of comet. Fifty cells from each slide and two slides per animal were observed. One-way ANOVA method was adopted for statistical analysis. This study shows that the chronic exposure to these radiations cause statistically significant ($p<0.001$) increase in DNA single strand breaks in brain cells of rat.

(E) Pesnya DS, Romanovsky AV. Comparison of cytotoxic and genotoxic effects of plutonium-239 alpha particles and mobile phone GSM 900 radiation in the Allium cepa test. Mutat Res. 750(1-2):27-33, 2013. (VO, AE, GT)

The goal of this study was to compare the cytotoxic and genotoxic effects of plutonium-239 alpha particles and GSM 900 modulated mobile phone radiation in the Allium cepa test. Three groups of bulbs were exposed to mobile phone radiation during 0 (sham), 3 and 9hours. A positive control group was treated during 20 min with plutonium-239 alpha-radiation. Mitotic abnormalities, chromosome aberrations, micronuclei and mitotic index were analyzed. Exposure to alpha-radiation from plutonium-239 and exposure to modulated radiation from mobile phone during 3 and 9h significantly increased the mitotic index. GSM 900 mobile phone radiation as well as alpha-radiation from plutonium-239 induced both clastogenic and aneugenic effects. However, the aneugenic activity of mobile phone radiation was more pronounced. After 9 hours of exposure to mobile phone radiation, polyploid cells, three-groups metaphases, amitoses and some unspecified abnormalities were detected, which were not registered in the other experimental groups. Importantly, GSM 900 mobile phone radiation increased the mitotic index, the frequency of mitotic and chromosome abnormalities, and the micronucleus frequency in a time-dependent manner. Due to its sensitivity, the Allium cepa test can be recommended as a useful cytogenetic assay to assess cytotoxic and genotoxic effects of radiofrequency electromagnetic fields.

(E) Phillips, J.L., Ivaschuk, O., Ishida-Jones, T., Jones, R.A., Campbell-Beachler, M. and Haggren, W. DNA damage in Molt-4 T- lymphoblastoid cells exposed to cellular telephone radiofrequency fields in vitro. Bioelectrochem. Bioenerg. 45:103-110, 1998. (VT, AE, LI, GT)

Molt-4 T-lymphoblastoid cells have been exposed to pulsed signals at cellular telephone frequencies of 813.5625 MHz (iDEN signal) and 836.55 MHz (TDMA signal). These studies were performed at low SAR (average = 2.4 and 24 microwatt/g for iDEN and 2.6 and 26 microwatt/g for TDMA) in studies designed to look for athermal RF effects. The alkaline comet, or single cell gel electrophoresis, assay was employed to measure DNA single-strand breaks in cell cultures exposed to the radiofrequency (RF) signal as compared to concurrent sham-exposed cultures. Tail moment and comet extent were calculated as indicators of DNA damage. Statistical differences in the distribution of values for tail moment and comet extent between exposed and control cell cultures were evaluated with the SKolmogorov-Smirnoff distribution test. Data points for all experiments of each exposure condition were pooled and analyzed as single groups. It was found that: 1) exposure of cells to the iDEN signal at an SAR of 2.4 microwatt/g for 2 h or 21 h significantly decreased DNA damage; 2) exposure of cells to the TDMA signal at an SAR of 2.6 microwatt/g for 2 h and 21 h significantly decreased DNA damage; 3) exposure of cells to the iDEN signal at an SAR of 24 microwatt/g for 2 h and 21 h significantly increased DNA damage; 4) exposure of cells to the TDMA signal at an SAR of 26 microwatt/g for 2 h significantly decreased DNA damage. The data indicate a need to study the effects of exposure to RF signals on direct DNA damage and on the rate at which DNA damage is repaired.

(E) Piccinetti CC, De Leo A, Cosoli G, Scalise L, Randazzo B, Cerri G, Olivotto I. Measurement of the 100 MHz EMF radiation in vivo effects on zebrafish D. rerio embryonic development: A multidisciplinary study. Ecotoxicol Environ Saf. 154:268-279, 2018. (VO, CE, LI, GE)

The augmented exposure of both environment and human being to electromagnetic waves and the concomitant lack of an unequivocal knowledge about biological consequences of these radiations, raised public interest on electromagnetic pollution. In this context, the present study aims to evaluate the biological effects on zebrafish (ZF) embryos of 100 MHz radiofrequency electromagnetic field (RF-EMF) exposure through a multidisciplinary protocol. Because of the shared synteny between human and ZF genomes that validated its use in biomedical research, toxicology and developmental biology studies, ZF was here selected as experimental model and a measurement protocol and biological analyses have been set up to clearly discriminate between RF-EMF biological and thermal effects. The results showed that a 100 MHz EMF was able to affect ZF embryonic development, from 24 to 72 h post fertilization (hpf) in all the analyzed pathways. Particularly, at the 48 hpf stage, a reduced growth, an increased transcription of oxidative stress genes, the onset of apoptotic/autophagic processes and a modification in cholesterol metabolism were detected. ZF embryos faced stress induced by EMF radiation by triggering detoxification mechanisms and at 72 hpf they partially recovered from stress reaching the hatching time in a comparable way respect to the control group. Data here obtained showed unequivocally the in vivo effects of RF-EMF on an animal model, excluding thermal outcomes and thus represents the starting point for more comprehensive studies on dose response effects of electromagnetic fields radiations consequences.

(E) Pooam M, Jourdan N, Aguida B, Dahon C, Baouz S, Terry C, Raad H, Ahmad M. Exposure to 1.8 GHz radiofrequency field modulates ROS in human HEK293 cells as a function of signal amplitude. Commun Integr Biol 15(1):54-66, 2022.(VT, AE, OX, GE, WS)

The modern telecommunications industry is ubiquitous throughout the world, with a significant percentage of the population using cellular phones on a daily basis. The possible physiological consequences of wireless emissions in the GHz range are therefore of major interest, but remain poorly understood. Here, we show that exposure to a 1.8 GHz carrier frequency in the amplitude range of household telecommunications induces the formation of ROS (Reactive Oxygen Species) in human HEK293 cultured cells. The ROS concentrations detected by fluorescent imaging techniques increased significantly after 15 minutes of RF field exposure, and were localized to both nuclear and cytosolic cellular compartments. qPCR analysis showed altered gene expression of both anti-oxidative (SOD, GPX, GPX, and CAT) and oxidative (Nox-2) enzymes. In addition, multiple genes previously identified as responsive to static magnetic fields were found to also be regulated by RF, suggesting common features in response mechanisms. By contrast, many RF effects showed evidence of hormesis, whereby biological responsivity does not occur linearly as a function of signal amplitude. Instead, biphasic dose response curves occur with 'blind' spots at certain signal amplitudes where no measureable response occurs. We conclude that modulation of intracellular ROS can be a direct consequence of RF exposure dependent on signal frequency and amplitude. Since changes in intracellular ROS may have both harmful and beneficial effects, these could provide the basis for many reported physiological effects of RF exposure.

(E)Porcher A, Girard S, Bonnet P, Rouveure R, Guérin V, Paladian F, Vian A. Non thermal 2.45 GHz electromagnetic exposure causes rapid changes in Arabidopsis thaliana metabolism. J Plant Physiol 286:153999, 2023. (VO, AE, GE, OX, LI)

Numerous studies report different types of responses following exposure of plants to high frequency electromagnetic fields (HF-EMF). While this phenomenon is related to tissue heating in animals, the situation is much less straightforward in plants where metabolic changes seem to occur without tissue temperature increase. We have set up an exposure system allowing reliable measurements of tissue heating (using a reflectometric probe and thermal imaging) after a long exposure (30 min) to an electromagnetic field of 2.45 GHz transmitted through a horn antenna (about 100 V m-1 at the plant level). We did not observe any heating of the tissues, but we detected rapid increases (60 min) in the accumulation of transcripts of stress-related genes (TCH1 and ZAT12 transcription factor) or involved in ROS metabolism (RBOHF and APX1). At the same time, the amounts of hydrogen peroxide and dehydroascorbic acid increased while glutathione (reduced and oxidized forms), ascorbic acid, and lipid peroxidation remained stable. Therefore, our results unambiguously show that molecular and biochemical responses occur rapidly (within 60min) in plants after exposure to an electromagnetic field, in absence of tissue heating.

(NE) Port M, Abend M, Romer B, Van Beuningen D. Influence of high-frequency electromagnetic fields on different modes of cell death and gene expression. Int J Radiat Biol. 79(9):701-708, 2003.(VT, AE, GE)

PURPOSE: International thresholds for exposure to non-ionizing radiation leading to non-thermal effects were conservatively set by the International Commission on Non-Ionizing Radiation Protection (ICNIRP). The aim of this study was to examine whether biological effects such as different modes of cell death and gene expression modifications related to tumorgenesis are detectable above the threshold defined. MATERIALS AND METHODS: Human leukaemia cells (HL-60) grown in vitro were exposed to electromagnetic fields (EMF; t 1/2(r) about 1 ns; field strength about 25 times higher than the ICNIRP reference levels for occupational exposure) leading to non-thermal effects using a high-voltage-improved GTEM cell 5302 (EMCO) connected to a pulse generator NP20 (C = 1 nF, U(Load) = 20kV). HL-60 cells were harvested at 0, 24, 48 and 72 h after radiation exposure. Micronuclei, apoptosis and abnormal cells (e.g. necrosis) were determined using morphological criteria. In parallel, the expression of 1176 genes was measured using Atlas Human 1.2. Array. Based on high data reproducibility calculated from two independent experiments (> 99%), array analysis was performed. RESULTS: No significant change in apoptosis, micronucleation, abnormal cells and differential gene expression was found. CONCLUSIONS: Exposure of HL-60 cells to EMFs 25 times higher than the ICNIRP reference levels for occupational exposure failed to induce any changes in apoptosis, micronucleation, abnormal morphologies and gene expression. Further experiments using EMFs above the conservatively defined reference level set by the ICNIRP may be desirable.

(E) Pyrpasopoulou A, Kotoula V, Cheva A, Hytiroglou P, Nikolakaki E, Magras IN, Xenos TD, Tsiboukis TD, Karkavelas G. Bone morphogenetic protein expression in newborn rat kidneys after prenatal exposure to radiofrequency radiation. Bioelectromagnetics

25(3):216-227, 2004. (CE, GE, IU, VO)

Effects of nonthermal radiofrequency radiation (RFR) of the global system of mobile communication (GSM) cellular phones have been as yet mostly studied at the molecular level in the context of cellular stress and proliferation, as well as neurotransmitter production and localization. In this study, a simulation model was designed for the exposure of pregnant rats to pulsed GSM-like RFR (9.4 GHz), based on the different resonant frequencies of man and rat. The power density applied was 5 microW/cm^2, in order to avoid thermal electromagnetic effects as much as possible. Pregnant rats were exposed to RFR during days 1-3 postcoitum (p.c.) (embryogenesis, pre-implantation) and days 4-7 p.c. (early organogenesis, peri-implantation). Relative expression and localization of bone morphogenetic proteins (BMP) and their receptors (BMPR), members of a molecular family currently considered as major endocrine and autocrine morphogens and known to be involved in renal development, were investigated in newborn kidneys from RFR exposed and sham irradiated (control) rats. Semi-quantitative duplex RT-PCR for BMP-4, -7, BMPR-IA, -IB, and -II showed increased BMP-4 and BMPR-IA, and decreased BMPR-II relative expression in newborn kidneys. These changes were statistically significant for BMP-4, BMPR-IA, and -II after exposure on days 1-3 p.c. ($P < .001$ each), and for BMP-4 and BMPR-IA after exposure on days 4-7 p.c. ($P < .001$ and $P = .005$, respectively). Immunohistochemistry and in situ hybridization (ISH) showed aberrant expression and localization of these molecules at the histological level. Our findings suggest that GSM-like RFR interferes with gene expression during early gestation and results in aberrations of BMP expression in the newborn. These molecular changes do not appear to affect renal organogenesis and may reflect a delay in the development of this organ. The differences of relative BMP expression after different time periods of exposure indicate the importance of timing for GSM-like RFR effects on embryonic development.

(E) Qin F, Zhang J, Cao H, Guo W, Chen L, Shen O, Sun J, Yi C, Li J, Wang J, Tong J. Circadian alterations of reproductive functional markers in male rats exposed to 1800-MHz radiofrequency field. Chronobiol Int. 3191):123-133, 2014. (VO, LE, GE, LI)

In this study, we explored the circadian effects of daily radiofrequency field (RF) exposure on reproductive functional markers in adult male Sprague-Dawley rats. Animals in circadian rhythm (as indicated by melatonin measurements), were divided into several groups and exposed to 1800 MHz RF at 205 μw/cm^2 power density (specific absorption rate 0.0405 W/kg) for 2 h/day for 32 days at different zeitgeber time (ZT) points, namely, ZT0, ZT4, ZT8, ZT12, ZT16 and ZT20. Sham-exposed animals were used as controls in the study. From each rat, testicular and epididymis tissues were collected and assessed for testosterone levels, daily sperm production and sperm motility, testis marker enzymes γ-GT and ACP, cytochrome P450 side-chain cleavage (p450cc) mRNA expression, and steroidogenic acute regulatory protein (StAR) mRNA expression. Via these measurements, we confirmed the existence of circadian rhythms in sham-exposed animals. However, rats exposed to RF exhibited a disruption of circadian rhythms, decreased testosterone levels, lower daily sperm production and sperm motility, down-regulated activity of γ-GT and ACP, as well as altered mRNA expression of cytochrome P450 and StAR. All of these observations were more pronounced when rats were exposed to RF at ZT0. Thus,

our findings indicate potential adverse effects of RF exposure on male reproductive functional markers, in terms of both the daily overall levels as well as the circadian rhythmicity.

(E) Qin F, Cao H, Yuan H, Guo W, Pei H, Cao Y, Tong J. 1800 MHz radiofrequency fields inhibits testosterone production via CaMKI /RORα pathway. Reprod Toxicol. 81:229-236, 2018. (VO, LE, GE, LI)

Exposure to radiofrequency fields (RF) has been reported to induce adverse effects on testosterone production and its daily rhythm. However, the mechanisms underneath this effect remain unknown. In this study, male mice were exposed to 1800 MHz radiofrequency fields (RF, 40 μW/cm^2 power intensity and 0.0553 W/Kg SAR) 2 h per day for 32 days. The data suggested that RF exposure: (i) significantly reduced testosterone levels, (ii) altered the expression of genes involved in its synthesis (Star, P450scc, P450c17 and 3β-Hsd) in testicular tissue, (iii) significantly reduced regulatory protein CaMKI/RORα. Similar observations were also made in cultured primary Leydig cells exposed in vitro to RF. However, all of these observations were blocked by CaMK inhibitor, KN-93, and ionomycin reversed the down-regulation effects on intracellular [Ca2$^+$]i and CaMKI/RORα expression induced by RF exposure. Thus, the data provided the evidence that RF-induced inhibition of testosterone synthesis might be mediated through CaMKI/RORα signaling pathway. Capsule: CaMKI/RORα signaling pathway was involved in the inhibition of testosterone synthesis induced by RF exposure.

(E) Qin F, Shen T, Cao H, Qian J, Zou D, Ye M, Pei H. CeO_2NPs relieve radiofrequency radiation, improve testosterone synthesis, and clock gene expression in Leydig cells by enhancing antioxidation. Int J Nanomedicine. 14:4601-4611, 2019. (VT, AE, GE, OX)

Introduction: The ratio of Ce^{3+}/Ce^{4+} in their structure confers unique functions on cerium oxide nanoparticles (CeO_2NPs) containing rare earth elements in scavenging free radicals and protecting against oxidative damage. The potential of CeO_2NPs to protect testosterone synthesis in primary mouse Leydig cells during exposure to 1,800 MHz radiofrequency (RF) radiation was examined in vitro. **Methods:** Leydig cells were treated with different concentrations of CeO_2NPs to identify the optimum concentration for cell proliferation. The cells were pretreated with the optimum dose of CeO_2NPs for 24 hrs and then exposed to 1,800 MHz RF at a power density of 200.27 μW/cm^2 (specific absorption rate (SAR), 0.116 W/kg) for 1 hr, 2 hrs, or 4 hrs. The medium was used to measure the testosterone concentration. The cells were collected to determine the antioxidant indices (catalase [CAT], malondialdehyde [MDA], and total antioxidant capacity [T-AOC]), and the mRNA expression of the testosterone synthase genes (*Star, Cyp11a1*, and *Hsd-3β*) and clock genes (*Clock, Bmal1*, and *Rorα*). **Results:** Our preliminary result showed that 128 μg/mL CeO_2NPs was the optimum dose for cell proliferation. Cells exposed to RF alone showed reduced levels of testosterone, T-AOC, and CAT activities, increased MDA content, and the downregulated genes expression of *Star, Cyp11a1, Hsd-3β, Clock, Bmal1*, and *Rorα*. Pretreatment of the cells with 128 μg/mL CeO_2NPs for 24 hrs followed by RF exposure significantly increased testosterone synthesis, upregulated the expression of the testosterone synthase and clock genes, and increased the resistance to oxidative

damage in Leydig cells compared with those in cells exposed to RF alone. **Conclusion:** Exposure to 1,800 MHz RF had adverse effects on testosterone synthesis, antioxidant levels, and clock gene expression in primary Leydig cells. Pretreatment with CeO_2NPs prevented the adverse effects on testosterone synthesis induced by RF exposure by regulating their antioxidant capacity and clock gene expression in vitro. Further studies of the mechanism underlying the protective function of CeO_2NPs against RF in the male reproductive system are required.

(E) Qin F, Cao H, Feng C, Zhu T, Zhu B, Zhang J, Tong J, Pei H. Microarray profiling of LncRNA expression in the testis of pubertal mice following morning and evening exposure to 1800 MHz radiofrequency fields. Chronobiol Int 38(12):1745-1760, 2021. (VO, LE, GE, RP)

In this paper, the chronotoxicity of radiofrequency fields (RF) in the pubertal testis development and the involved molecular pathways were investigated by exposing four-week-old mice to RF (1800 MHz, SAR, 0.50 W/kg) in the morning and evening of each day for three weeks. Then, pathological changes and functional indices within the testis were determined. We also used a long non-coding RNA (lncRNA) microarray and GO/KEGG pathway analyses to determine lncRNA expression profiles and predict their potential functions. The cis and trans regulation of lncRNAs were investigated, and an interaction network was constructed using Cytoscape software. RF exposure led to a range of pathological changes in the testes of adolescent mice, as testicular weights and daily sperm productions decreased, and the testosterone secretion reduced. Furthermore, RF induced dysregulation in the expression of testicular lncRNAs. We identified 615 and 183 differentially expressed lncRNAs that were associated with morning and evening exposure to RF, respectively. From 15 differential expression lncRNAs both in morning RF group and evening RF group, we selected 6 lncRNAs to be validated by quantitative reverse transcription PCR (qRT-PCR). The differentially expressed lncRNAs induced by morning RF exposure were highly correlated with many different pathways, including Fanconi syndrome, metabolic processes, cell cycle, DNA damage, and DNA replication. Trans-regulation analyses further showed that differentially expressed lncRNAs were involved in multiple transcription factor-regulated pathways, such as TCFAP4, NFkB, HINFP, TFDP2, FoxN1, and PAX5. These transcription factors have all been shown to be involved in the modulation of testis development, cell cycle progression, and spermatogenesis. These findings suggest that the extent to which 1800 MHz RF induced toxicity in the testes and changed the expression of lncRNAs showed differences between morning exposure and evening exposure. These data indicate that differentially expressed lncRNAs play crucial roles in the RF exposure damage to the developing pubertal testis. Collectively, our findings provide a better understanding of the mechanisms underlying the toxic effects of RF exposure on testicular development.

(NE) Qutob SS, Chauhan V, Bellier PV, Yauk CL, Douglas GR, Berndt L, Williams A, Gajda GB, Lemay E, Thansandote A, McNamee JP. Microarray gene expression profiling of a human glioblastoma cell line exposed in vitro to a 1.9 GHz pulse-modulated radiofrequency field. Radiat Res 165:636-644, 2006. (VT, AE, GE)

The widespread use of mobile phones has led to public concerns about the health effects associated with exposure to radiofrequency (RF) fields. The paramount concern of most persons

relates to the potential of these fields to cause cancer. Unlike ionizing radiation, RF fields used for mobile telecommunications (800-1900 MHz) do not possess sufficient energy to directly damage DNA. Most rodent bioassay and in vitro genotoxicity/mutation studies have reported that RF fields at non-thermal levels have no direct mutagenic, genotoxic or carcinogenic effects. However, some evidence has suggested that RF fields may cause detectable postexposure changes in gene expression. Therefore, the purpose of this study was to assess the ability of exposure to a 1.9 GHz pulse-modulated RF field for 4 h at specific absorption rates (SARs) of 0.1, 1.0 and 10.0 W/kg to affect global gene expression in U87MG glioblastoma cells. We found no evidence that non-thermal RF fields can affect gene expression in cultured U87MG cells relative to the nonirradiated control groups, whereas exposure to heat shock at 43 degrees C for 1 h up-regulated a number of typical stress-responsive genes in the positive control group. Future studies will assess the effect of RF fields on other cell lines and on gene expression in the mouse brain after in vivo exposure.

(E) Racuciu M, Effects of radiofrequency radiation on root tip cells of Zea mays. Roumanian Biotechnological Letters 14 (3) 4365-4369, 2009. (VT, AE, GT)

In this study, the mutagenic effects of low power radiofrequency radiation on Zea mays root tip were studied. Cells in different division phases and chromosomal aberration assay were used to determine the mitotic index and chromosomal aberration frequency of Zea mays root tip cells induced by 900MHz radiofrequency radiation. Zea mays seeds, having a uniform genophond, have been exposed to RF field of low power density, for different time intervals, between 1.0 and 36.0 hours. Exposure to RF field was applied to seeds before germination process. Continuous wave on 900 MHz was used for irradiation. Incident field distribution in the irradiation area was characterized, so as an as possible as uniform field to be applied in the volume of the sample. The results showed that the mitotic index and chromosomal aberration frequency showed linear increasing for radiofrequency radiation treatment of increased exposure time.

(E) Rago R, Salacone P, Caponecchia L, Sebastianelli A, Marcucci I, Calogero AE, Condorelli R, Vicari E, Morgia G, Favilla V, Cimino S, Arcoria AF, La Vignera S. The semen quality of the mobile phone users. J Endocrinol Invest. 36(11):970-974, 2013. (HU, LE, GT, RP)

BACKGROUND: The increased use of mobile phones, the media's attention for general health, and the increase of idiopathic male infertility suggest to investigate the possible consequences of an excessive use of mobile phones on semen quality. AIM: To evaluate the conventional and some of the main biofunctional sperm parameters in healthy men according to the different use of the mobile phone. SUBJECTS AND METHODS: All the enrolled subjects in this study were divided into four groups according to their active cell phone use: group A= no use (no.=10 subjects); group B= <2 h/day (no.=16); group C= 2-4 h/day (no.=17); and group D= >4 h/day (no.=20). Among the subjects of the group D (>4 h/day), a further evaluation was made between the "trousers users"(no.=12) and "shirt users"(no.=8), and they underwent semen collection to evaluate conventional and biofunctional sperm parameters (density, total count, morphology, progressive motility, apoptosis, mithocondrial membrane potential, chromatin compaction, DNA fragmentation). RESULTS: None of the conventional sperm parameters examined were

significantly altered. However, the group D and the trousers users showed a higher percentage of sperm DNA fragmentation compared to other groups. CONCLUSION: These results suggest that the sperm DNA fragmentation could represent the only parameter significantly altered in the subjects who use the mobile phone for more than 4 h/day and in particular for those who use the device in the pocket of the trousers.

(E) Rammal M, Jebai F. Rammal H, Joumaa WH. Effects of long-term exposure to RF/MW radiations on the expression of mRNA of stress proteins in *Lycospersicon esculentum.* WSEAS Transect Biol Biomed. 11:10-14, 2014. (VT, AE, GE, LI)

Low levels of RF radiation exposure can modify the protein's activity by stimulating or inhibiting their expression in cells. The proteinase inhibitor (Pin II) and *Lycospersicon esculentum basic leucine Zipper1* (lebZIP1) are two wound-plants genes. The aim of this work is to study the rate of accumulation of pin II and lebZIP1 at the level of messenger RNA after 10 days of electromagnetic waves exposure. Using RT-PCR and RT-qPCR, the results show that Pin II and lebZIP1synthesis change at the level of cDNA. PinII and lebZIP1 surexpression influence the growth and the differentiation and evoke an increase of protein accumulation in the cell.

(NE) Regalbuto E, Anselmo A, De Sanctis S, Franchini V, Lista F, Benvenuto M, Bei R, Masuelli L, D'Inzeo G, Paffi A, Trodella E, Sgura A. Human fibroblasts in vitro exposed to 2.45 GHz continuous and pulsed wave signals: evaluation of biological effects with a multimethodological approach. Int J Mol Sci 21(19):E7069, 2020.(VT, AE, GT)

The increasing exposure to radiofrequency electromagnetic fields (RF-EMF), especially from wireless communication devices, raises questions about their possible adverse health effects. So far, several in vitro studies evaluating RF-EMF genotoxic and cytotoxic non-thermal effects have reported contradictory results that could be mainly due to inadequate experimental design and lack of well-characterized exposure systems and conditions. Moreover, a topic poorly investigated is related to signal modulation induced by electromagnetic fields. The aim of this study was to perform an analysis of the potential non-thermal biological effects induced by 2.45 GHz exposures through a characterized exposure system and a multimethodological approach. Human fibroblasts were exposed to continuous (CW) and pulsed (PW) signals for 2 h in a wire patch cell-based exposure system at the specific absorption rate (SAR) of 0.7 W/kg. The evaluation of the potential biological effects was carried out through a multimethodological approach, including classical biological markers (genotoxic, cell cycle, and ultrastructural) and the evaluation of gene expression profile through the powerful high-throughput next generation sequencing (NGS) RNA sequencing (RNA-seq) approach. Our results suggest that 2.45 GHz radiofrequency fields did not induce significant biological effects at a cellular or molecular level for the evaluated exposure parameters and conditions.

(E) Remondini D, Nylund R, Reivinen J, Poulletier de Gannes F, Veyret B, Lagroye I, Haro E, Trillo MA, Capri M, Franceschi C, Schlatterer K, Gminski R, Fitzner R, Tauber R, Schuderer J, Kuster N, Leszczynski D, Bersani F, Maercker C. Gene expression changes in human cells after exposure to mobile phone microwaves. Proteomics 6:4745-4754, 2006. (VT, AE, GE, CS)

Possible biological effects of mobile phone microwaves were investigated in vitro. In this study, which was part of the 5FP EU project REFLEX (Risk Evaluation of Potential Environmental Hazards From Low-Energy Electromagnetic Field Exposure Using Sensitive in vitro Methods), six human cell types, immortalized cell lines and primary cells, were exposed to 900 and 1800 MHz. RNA was isolated from exposed and sham-exposed cells and labeled for transcriptome analysis on whole-genome cDNA arrays. The results were evaluated statistically using bioinformatics techniques and examined for biological relevance with the help of different databases. NB69 neuroblastoma cells, T lymphocytes, and CHME5 microglial cells did not show significant changes in gene expression. In EA.hy926 endothelial cells, U937 lymphoblastoma cells, and HL-60 leukemia cells we found between 12 and 34 up- or down-regulated genes. Analysis of the affected gene families does not point towards a stress response. However, following microwave exposure, some but not all human cells might react with an increase in expression of genes encoding ribosomal proteins and therefore up-regulating the cellular metabolism.

(E) Romano-Spica V, Mucci N, Ursini CL, Ianni A, Bhat NK. Ets1 oncogene induction by ELF-modulated 50 MHz radiofrequency electromagnetic field. Bioelectromagnetics 21(1):8-18, 2000. (VT, AE, GE)

We have analyzed gene expression in hemopoietic and testicular cell types after their exposure to 50 MHz radiofrequency (RF) non-ionizing radiation modulated (80%) with a 16 Hz frequency. The exposure system generates a 0.2 microT magnetic field parallel to the ground and a 60 V/m electric field orthogonal to the earth's magnetic field. Exposure conditions were selected so as to interfere with the calcium ion flow. Under these electromagnetic field (EMF) conditions, we observed an overexpression of the ets1 mRNA in Jurkat T-lymphoblastoid and Leydig TM3 cell lines. This effect was observed only in the presence of the 16 Hz modulation, corresponding to the resonance frequency for calcium ion with a DC magnetic field of 45.7 microT. We have also identified a putative candidate gene repressed after EMF exposure. The experimental model described in this paper may contribute to the understanding of the biological mechanisms involved in EMF effects.

Romeo S, Zeni O, Sannino A, Lagorio S, Biffoni M, Scarfì MR. Genotoxicity of radiofrequency electromagnetic fields: Protocol for a systematic review of in vitro studies. Environ Int. 2021 Jan 21;148:106386. doi: 10.1016/j.envint.2021.106386. (Opinion)

Background: Exposure to radiofrequency electromagnetic fields (RF-EMF, 100 kHz - 300 GHz) emitted by wireless communication technologies is pervasive and ubiquitous. Concern has been raised about possible adverse effects to human health. In 2011 the International Agency for Research on Cancer has classified RF-EMF as possibly carcinogenic to humans, highlighting that the evidence is weak and far from conclusive. Updated systematic reviews of the scientific literature on this topic are lacking, especially for mechanistic studies. Objectives: To develop a protocol for a systematic review of experimental studies investigating genotoxic effects induced by RF-EMF in in vitro cellular models. Genotoxicity is one of the key-biological indicators of carcinogenicity, and the most common characteristics of established carcinogens. The predefined procedures for conducting the systematic review are outlined below. Methods: We will follow the guidelines developed by the National Toxicology Program-Office of Health Assessment and

Translation (NTP-OHAT), adapted to the evaluation of in vitro studies. Eligibility criteria: We will include experimental in vitro studies addressing the relationship between controlled exposures to RF-EMF and genotoxicity in mammalian cells only. Eligibility for inclusion will be further restricted to peer reviewed articles reporting findings from primary studies. Information sources: We will search the scientific literature databases NCBI PubMed, Web of Science, and EMF-Portal. No filter on publication date will be applied. Only studies published in English will be considered. The reference lists of the included papers and available reviews will be screened for unidentified relevant papers. References will be managed through Endnote X9 software. Data extraction and synthesis of results: Data from included papers will be extracted according to predefined forms. Heterogeneity within the available evidence will determine the type of evidence synthesis that is appropriate. Findings will be summarized in tables, graphical displays and in a narrative synthesis of the available evidences. A meta-analysis will be carried out if subgroups of studies homogeneous in terms of exposure characteristics, endpoint, and cell types will be identified. Risk of bias: The internal validity of included studies will be assessed using the NTP-OHAT Risk of Bias Rating Tool for animal studies, adapted to in vitro studies. This stage of the process will be managed through the Health Assessment Workspace Collaborative (HAWC). Evidence appraisal: To rate confidence in the body of evidence, we will use the OHAT GRADE-based approach for animal studies.

(NE) Ros-Llor I, Sanchez-Siles M, Camacho-Alonso F, Lopez-Jornet P. Effect of mobile phones on micronucleus frequency in human exfoliated oral mucosal cells. Oral Dis. 18:786-792, 2012. (HU, LE, GT)

Objective: In the last two decades, the use of mobile phones has increased enormously all over the world. The controversy regarding whether radiofrequency (RF) fields exert effects upon biological systems is a concern for the general population. An evaluation is made of DNA damage and cytokinetic defects, proliferative potential, and cell death because of RF radiation emitted by mobile phones in healthy young users. Study design: This cohort study was carried out in 50 Caucasian mobile phone users. We collected two cell samples from each subject (a total of 100 cell samples), corresponding to the right and left cheek mucosa, respectively. Case histories and personal information were assessed, including age, gender, body height and weight, history of cancer, smoking and alcohol consumption, exposure to chemical carcinogens or radiation, and dietary habits. Sampling comprised cell collection from both cheeks with a cytobrush, centrifugation, slide preparation, fixation, and staining, followed by fluorescent microscopic analysis. A total of 2000 exfoliated cells were screened for nuclear abnormalities, especially micronucleus. Results: No statistically significant changes were recorded in relation to age, gender, body mass index, or smoking status. A comparison of the results vs the control area according to the side of the face on which the mobile phone was placed, and in relation to the duration of exposure (years) to mobile phone radiation in the total 100 samples, yielded no significant differences. Conclusions: No genotoxic effects because of RF exposure were observed in relation to any of the study parameters.

(NE) Roti Roti JL , Malyapa RS, Bisht KS, Ahern EW, Moros EG, Pickard WF, Straube WL, Neoplastic Transformation in C3H 10T(1/2) cells after exposure to 835.62 MHz FDMA and 847.74 MHz CDMA radiations. Radiat Res 155(1):239-247, 2001.(VT, LE, GT)

The effect of radiofrequency (RF) radiation in the cellular phone communication range (835.62 MHz frequency division multiple access, FDMA; 847.74 MHz code division multiple access, CDMA) on neoplastic transformation frequency was measured using the in vitro C3H 10T(1/2) cell transformation assay system. To determine if 835.62 MHz FDMA or 847.74 MHz CDMA radiations have any genotoxic effects that induce neoplastic transformation, C3H 10T(1/2) cells were exposed at 37 degrees C to either of the above radiations [each at a specific absorption rate (SAR) of 0.6 W/kg] or sham-exposed at the same time for 7 days. After the culture medium was changed, the cultures were transferred to incubators and refed with fresh growth medium every 7 days. After 42 days, the cells were fixed and stained with Giemsa, and transformed foci were scored. To determine if exposure to 835.62 MHz FDMA or 847.74 MHz CDMA radiation has any epigenetic effects that can promote neoplastic transformation, cells were first exposed to 4.5 Gy of X rays to induce the transformation process and then exposed to the above radiations (SAR = 0.6 W/kg) in temperature-controlled irradiators with weekly refeeding for 42 days. After both the 7-day RF exposure and the 42-day RF exposure after X irradiation, no statistically significant differences in the transformation frequencies were observed between incubator controls, the sham-exposed (maintained in irradiators without power to the antenna), and the 835.62 MHz FDMA or 847.74 MHz CDMA-exposed groups.

(E) Roux D, Vian A, Girard S, Bonnet P, Paladian F, Davies E, Ledoigt G. Electromagnetic fields (900 MHz) evoke consistent molecular responses in tomato plants. Physiologia Plantarum 128: 283–288, 2006. (VO, AE, GE, LI)

Although the effects of high-frequency electromagnetic fields on biological systems have been studied frequently, unequivocal results have rarely been obtained, primarily because suitably controlled experiments could not be performed. In the present work, tomato plants were exposed to a homogeneous and isotropic field (900 MHz) using a mode stirred reverberation chamber, and the stress-related transcripts (calmodulin, protease inhibitor and chloroplast mRNA-binding protein) were assayed by real-time quantitative PCR. Exposure to an electromagnetic field induced a biphasic response, in which the levels of all three transcripts increased four- to six-fold 15 min after the end of electromagnetic stimulation, dropped to close to initial levels by 30 min, and then increased again at 60 min. We deliberately focused on the very early molecular responses to high-frequency electromagnetic fields in order to minimize secondary effects.

(E) Roux D, Vian A, Girard S, Bonnet P, Paladian F, Davies E, Ledoigt G. High frequency (900 MHz) low amplitude (5 V m-1) electromagnetic field: a genuine environmental stimulus that affects transcription, translation, calcium and energy charge in tomato. **Planta. 227(4):883-891, 2008. (VO, AE, GE)**

Using an especially-designed facility, the Mode Stirred Reverberation Chamber, we exposed tomato plants (Lycopersicon esculentum Mill. VFN8) to low level (900 MHz, 5 V m(-1)) electromagnetic fields for a short period (10 min) and measured changes in abundance of three specific mRNA soon after exposure. Within minutes of electromagnetic stimulation, stress-related mRNA (calmodulin, calcium-dependent protein kinase and proteinase inhibitor) accumulated in a rapid, large and 3-phase manner typical of an environmental stress response. Accumulation of these transcripts into the polysomal RNA also took place (indicating that the encoded proteins were translated) but was delayed (indicating that newly-synthesized mRNA was not immediately recruited into polysomes). Transcript accumulation was maximal at normal Ca(2+) levels and was depressed at higher Ca(2+), especially for those encoding calcium-binding proteins. Removal of Ca(2+) (by addition of chelating agents or Ca(2+) channel blocker) led to total suppression of mRNA accumulation. Finally, 30 min after the electromagnetic treatment, ATP concentration and adenylate energy charge were transiently decreased, while transcript accumulation was totally prevented by application of the uncoupling reagent, CCCP. These responses occur very soon after exposure, strongly suggesting that they are the direct consequence of application of radio-frequency fields and their similarities to wound responses strongly suggests that this radiation is perceived by plants as an injurious stimulus.

(NE) Roux D, Girard S, Paladian F, Bonnet P, Lalléchère S, Gendraud M, Davies E, Vian A. Human keratinocytes in culture exhibit no response when exposed to short duration, low amplitude, high frequency (900 MHz) electromagnetic fields in a reverberation chamber. Bioelectromagnetics 32(4):302-311, 2011. (VT, AE, LI, GE)

We exposed normal human epidermal keratinocytes to short duration, high frequency, and low amplitude electromagnetic fields, similar to that used by mobile phone technologies. We paid particular attention to the control of the characteristics of the electromagnetic environment generated within a mode stirred reverberation chamber (statistical homogeneity and isotropy of the field and SAR distribution). Two non-thermal exposure conditions were tested on the epidermal cells: 10-min exposure with a field amplitude of 8 V/m, and 30 min with 41 V/m. Corresponding specific absorption rates ranged from 2.6 to 73 mW/kg (continuous wave, 900 MHz carrier frequency). We collected RNA from cells subjected to these conditions and used it for a large-scale microarray screening of over 47000 human genes. Under these conditions, exposure of keratinocytes to the electromagnetic field had little effect; only 20 genes displayed significant modulation. The expression ratios were very small (close to 1.5-fold change), and none of them were shared by the two tested conditions. Furthermore, those assayed using polymerase chain reaction did not display significant expression modulation (overall mean of the exposed samples: 1.20 ± 0.18). In conclusion, the data presented here show that cultured keratinocytes are not significantly affected by EMF exposure.

(E) Sagripanti JL, Swicord ML. DNA structural changes caused by microwave radiation. Int J Radiat Biol Relat Stud Phys Chem Med. 50(1):47-50, 1986. (VT, AE, GT)

No Abstract available. "A 28 µl solution containing 10 µg purified DNA was exposed to 2.55GHz

microwave radiation at 20°C for 20min". "SAR_{min} and SAR_{max} ranges: 0, 2-8-5 and 21-85 mW/g". "These results suggest that exposure to microwave radiation can cause single as well as double-strand breaks in DNA in solution."

(E) Sagripanti JL, Swicord ML, Davis CC. Microwave effects on plasmid DNA. Radiat Res. 110(2):219-221, 1987. (VT, AE, GT)

The exposure of purified plasmid DNA to microwave radiation at nonthermal levels in the frequency range from 2.00 to 8.75 GHz produces single- and double-strand breaks that are detected by agarose gel electrophoresis. Microwave-induced damage to DNA depends on the presence of small amounts of copper. This effect is dependent upon both the microwave power and the duration of the exposure. Cuprous, but not cupric, ions were able to mimic the effects produced by microwaves on DNA.

(E) Sahin D. Ozgur E, Guler G, Tomruk A, Unlu I, Sepici-Dincel A, et al. The 2100 MHz radiofrequency radiation of a 3G-mobile phone and the DNA oxidative damage in brain. J Chem Neuroanatmy. 75: 94-98, 2016. (VO, LE, GT, OX)

We aimed to evaluate the effect of 2100MHz radiofrequency radiation emitted by a generator, simulating a 3G-mobile phone on the brain of rats during 10 and 40 days of exposure. The female rats were randomly divided into four groups. Group I; exposed to 3G modulated 2100MHz RFR signal for 6h/day, 5 consecutive days/wk for 2 weeks, group II; control 10 days, were kept in an inactive exposure set-up for 6h/day, 5 consecutive days/wk for 2 weeks, group III; exposed to 3G modulated 2100MHz RFR signal for 6h/day, 5 consecutive days/wk for 8 weeks and group IV; control 40 days, were kept in an inactive exposure set-up for 6h/day, 5 consecutive days/wk for 8 weeks. After the genomic DNA content of brain was extracted, oxidative DNA damage (8-hydroxy-2'deoxyguanosine, pg/mL) and malondialdehyde (MDA, nmoL/g tissue) levels were determined. Our main finding was the increased oxidative DNA damage to brain after 10 days of exposure with the decreased oxidative DNA damage following 40 days of exposure compared to their control groups. Besides decreased lipid peroxidation end product, MDA, was observed after 40 days of exposure. The measured decreased quantities of damage during the 40 days of exposure could be the means of adapted and increased DNA repair mechanisms.

(E) Said-Salman IH, Jebaii FA, Yusef HH, Moustafa ME. Global gene expression analysis of Escherichia coli K-12 DH5α after exposure to 2.4 GHz wireless fidelity radiation. Sci Rep. 9(1):14425, 2019. (VT, AE, GE)

This study investigated the non-thermal effects of Wi-Fi radiofrequency radiation of 2.4 GHz on global gene expression in Escherichia coli K-12 DH5α. High-throughput RNA-sequencing of 2.4 GHz exposed and non-exposed bacteria revealed that 101 genes were differentially expressed (DEGs) at $P \leq 0.05$. The up-regulated genes were 52 while the down-regulated ones were 49. QRT-PCR analysis of pgaD, fliC, cheY, malP, malZ, motB, alsC, alsK, appB and appX confirmed the RNA-seq results. About 7% of DEGs are involved in cellular component organization, 6% in response to stress stimulus, 6% in biological regulation, 6% in localization, 5% in locomotion and 3% in cell adhesion. Database for annotation, visualization and integrated

discovery (DAVID) functional clustering revealed that DEGs with high enrichment score included genes for localization of cell, locomotion, chemotaxis, response to external stimulus and cell adhesion. Kyoto encyclopedia of genes and genomes (KEGG) pathways analysis showed that the pathways for flagellar assembly, chemotaxis and two-component system were affected. Go enrichment analysis indicated that the up-regulated DEGs are involved in metabolic pathways, transposition, response to stimuli, motility, chemotaxis and cell adhesion. The down-regulated DEGs are associated with metabolic pathways and localization of ions and organic molecules. Therefore, the exposure of E. coli DH5α to Wi-Fi radiofrequency radiation for 5 hours influenced several bacterial cellular and metabolic processes.

(E) Saka VP, Chitra V, his Polon D. Protective role of hispolon and its derivatives against apoptosis in cortical neurons induced by electromagnetic radiation from 4G mobile phone. J Biochem Mol Toxicol 2023 Mar 30; e23351. doi: 10.1002/jbt.23351. Online ahead of print. (VT, AE, GE, OX)

Electromagnetic radiation (EMR) from wireless devices, particularly mobile phones, is a potentially growing public health concern. In this study, the neuronal effects of EMR on primary cortical neurons (PCNs) from neonatal rat cerebral cortex and the protective role of hispolon (HIS) and its derivatives were investigated as a measure of cranial exposure during mobile phone use. PCNs were isolated and cultured from day-old neonatal rats, then exposed for 2 h to EMR emitted by a mobile phone operating at a frequency of 2100 MHz with 1.6 W/Kg specific absorption rate (SAR) in call-answered mode treated with HIS and its derivatives. The induction of apoptosis through modulation of pro and anti-apoptotic genes via mitochondrial pathway and the protection by the test compounds was assessed. Pyrazole derivatives decreased apoptosis by modulating the levels of pro and anti-apoptotic genes by reducing the levels of reactive oxygen species (ROS) via mitochondrial damage, which was observed in the EMR exposed PCNs. The pyrazole compounds were found to have antioxidative and anti-apoptotic properties. Thus, the neuroprotective mechanisms of the pyrazole derivatives can be investigated further, which may make them appropriate as lead compounds in developing neuroprotective formulations.

(NE) Sakuma N, Komatsubara Y, Takeda H, Hirose H, Sekijima M, Nojima T, Miyakoshi J. DNA strand breaks are not induced in human cells exposed to 2.1425 GHz band CW and W-CDMA modulated radiofrequency fields allocated to mobile radio base stations. Bioelectromagnetics 27:51-57, 2006. (VT, AE, CT)

We conducted a large-scale in vitro study focused on the effects of low level radiofrequency (RF) fields from mobile radio base stations employing the International Mobile Telecommunication 2000 (IMT-2000) cellular system in order to test the hypothesis that modulated RF fields may act as a DNA damaging agent. First, we evaluated the responses of human cells to microwave exposure at a specific absorption rate (SAR) of 80 mW/kg, which corresponds to the limit of the average whole body SAR for general public exposure defined as a basic restriction in the International Commission on Non-Ionizing Radiation Protection (ICNIRP) guidelines. Second, we investigated whether continuous wave (CW) and Wideband Code Division Multiple Access (W-CDMA) modulated signal RF fields at 2.1425 GHz induced different levels of DNA damage. Human glioblastoma A172 cells and normal human IMR-90 fibroblasts from fetal lungs were exposed to mobile communication frequency radiation to

investigate whether such exposure produced DNA strand breaks in cell culture. A172 cells were exposed to W-CDMA radiation at SARs of 80, 250, and 800 mW/kg and CW radiation at 80 mW/kg for 2 and 24 h, while IMR-90 cells were exposed to both W-CDMA and CW radiations at a SAR of 80 mW/kg for the same time periods. Under the same RF field exposure conditions, no significant differences in the DNA strand breaks were observed between the test groups exposed to W-CDMA or CW radiation and the sham exposed negative controls, as evaluated immediately after the exposure periods by alkaline comet assays. Our results confirm that low level exposures do not act as a genotoxicant up to a SAR of 800 mW/kg.

(NE) Sakurai T, Kiyokawa T, Narita E, Suzuki Y, Taki M, Miyakoshi J. Analysis of gene expression in a human-derived glial cell line exposed to 2.45 GHz continuous radiofrequency electromagnetic fields. J Radiat Res. 52(2):185-192, 2011. (VT, AE, GE)

The increasing use of mobile phones has aroused public concern regarding the potential health risks of radiofrequency (RF) fields. We investigated the effects of exposure to RF fields (2.45 GHz, continuous wave) at specific absorption rate (SAR) of 1, 5, and 10 W/kg for 1, 4, and 24 h on gene expression in a normal human glial cell line, SVGp12, using DNA microarray. Microarray analysis revealed 23 assigned gene spots and 5 non-assigned gene spots as prospective altered gene spots. Twenty-two genes out of the 23 assigned gene spots were further analyzed by reverse transcription-polymerase chain reaction to validate the results of microarray, and no significant alterations in gene expression were observed. Under the experimental conditions used in this study, we found no evidence that exposure to RF fields affected gene expression in SVGp12 cells.

(E) Salameh M, Sukaina Zeitoun-Ghandour, Lina Sabra, Lina Ismail, Ahmad Daher, Ali Bazzi , Mahmoud Khalil, Wissam H Joumaa. Effects of continuous prenatal and postnatal global system for mobile communications electromagnetic waves (GSM-EMW) exposure on the oxidative stress biomarkers in female rat liver. Heliyon 8(12):e12367, 2022. (VO, LE, GE, OX, RP)

In light of the increased use of communication technologies, the harm caused by continuous exposure to emitted radiation on pregnancy and developing newborns is among the public concerns. Using Sprague-Dawley rats, our study investigates the effects of 24 h/day prenatal and postnatal 900 MHz radiofrequency electromagnetic radiation (RF-EMR) exposure of female rats on liver oxidative stress (OS) and other hepatic parameters at postnatal days (PND) 1, 9, and 21. Our results showed that RF-EMR exposure led to an increase in oxidative stress status as indicated by a significant elevation in MDA level at PND9 and PND21, a decrease in catalase (CAT) activity at all ages, a reduction (PND1 and PND9) in catalase amounts and mRNA expression, in addition to a decrease in GPx activity at PND21 in the exposed group. Current findings also showed a significant increase in cytoSOD at PND9 and 21 and a reduction in mitoSOD at PND21 in the exposed groups compared to the control groups. However, significant increases in glutathione peroxidase (GPx) level and mitoSOD activity were observed at all studied ages. Furthermore, cytoSOD activity showed a significant reduction in PND1, whereas in PND9 the value of this parameter increased compared to the non-exposed group. Moreover, while SOD1 mRNA expression increased at PND1, it decreased at PND9 and 21. However, GPx1 expression was shown to be always decreased in the exposed group. In addition, at PND1

and 9, exposed rats showed a similar response on Akt1, nuclear factor erythroïd 2-related factor 2 (Nrf-2), and intercellular adhesion molecule-1 (ICAM-1) expression. Therefore, an increased oxidative stress status produced from a continuous (24 h/day) GSM-modulated 900 MHz radiofrequency electromagnetic radiation (RF-EMR) exposure during the prenatal and postnatal periods may result in adverse health effects during future life stages.

(NE) Salmen SH, Alharbi SA, Faden AA, Wainwright M. Evaluation of effect of high frequency electromagnetic field on growth and antibiotic sensitivity of bacteria. Saudi J Biol Sci. 25(1):105-110, 2018. (VO, AE, GT)

This study was aimed to evaluate the impact of high frequency electromagnetic fields (HF-EMF at 900 and 1800 MHz) on DNA, growth rate and antibiotic susceptibility of *S. aureus*, *S. epidermidis*, and *P. aeruginosa*. In this study, bacteria were exposed to 900 and 1800 MHz for 2 h and then inoculated to new medium when their growth rate and antibiotic susceptibility were evaluated. Results for the study of bacterial DNA unsuccessful to appearance any difference exposed and non-exposed *S. aureus* and *S. epidermidis*. Exposure of *S. epidermidis* and *S. aureus* to electromagnetic fields mostly produced no statistically significant decrease in bacterial growth, except for *S. aureus* when exposure to 900 MHz at 12 h. Exposure of *P. aeruginosa* to electromagnetic fields at 900 MHz however, lead to a significant reduction in growth rate, while 1800 MHz had insignificant effect. With the exception of *S. aureus*, treated with amoxicillin (30 µg) and exposed to electromagnetic fields, radiation treatment had no significant effect on bacterial sensitivity to antibiotics.

(NE) Sannino A, Calabrese ML, d'Ambrosio G, Massa R, Petraglia G, Mita P, et al. Evaluation of cytotoxic and genotoxic effects in human peripheral blood leukocytes following exposure to 1950- MHz modulated signal. IEEE Trans Plasma Sci. 34: 1441-1448, 2006. (VT, AE, GT)

Human peripheral blood leukocytes from six volunteers were exposed to a Universal Mobile Telecommunication System (UMTS) signal (frequency carrier of 1950 MHz) for 24 h. The exposures were carried out in a waveguide system at specific absorption rates (SAR) of 0.5 and 2.0 W/kg, and for each blood donor, sham-exposed samples were also set up. The alkaline comet assay was used to quantify DNA damage, while cytotoxicity was determined by the Trypan blue exclusion method. The results obtained indicate the absence of genotoxic and cytotoxic effects at both SAR levels investigated, as assessed by comparing sham-exposed and exposed samples. Therefore, the findings indicate that, in the experimental conditions adopted, 24-h in vitro exposure to 1950-MHz radio-frequency radiation (UMTS signal) does not induce DNA damage in human leukocytes

(NE) Sannino A, Di Costanzo G, Brescia F, Sarti M, Zeni O, Juutilainen J, Scarfì MR. Human fibroblasts and 900 MHz radiofrequency radiation: evaluation of DNA damage after exposure and co-exposure to 3-Chloro-4-(dichloromethyl)-5-Hydroxy-2(5h)-furanone (MX). Radiat Res 171:743-751, 2009. (VT, AE, NT, IX)

The aim of this study was to investigate DNA damage in human dermal fibroblasts from a healthy subject and from a subject affected by Turner's syndrome that were exposed for 24 h to radiofrequency (RF) radiation at 900 MHz. The RF-radiation exposure was carried out alone or

in combination with 3-chloro-4-(dichloromethyl)-5-hydroxy-2(5H)-furanone (MX), a well-known environmental mutagen and carcinogen produced during the chlorination of drinking water. Turner's syndrome fibroblasts were also exposed for a shorter time (1 h). A signal similar to that emitted by Global System for Mobile Communications (GSM) mobile phones was used at a specific absorption rate of 1 W/kg under strictly controlled conditions of temperature and dosimetry. To evaluate DNA damage after RF-radiation exposure alone, the alkaline comet assay and the cytokinesis-block micronucleus assay were used. In the combined-exposure experiments, MX was given at a concentration of 25 microM for 1 h immediately after the RF-radiation exposure, and the effects were evaluated by the alkaline comet assay. The results revealed no genotoxic and cytotoxic effects from RF radiation alone in either cell line. As expected, MX treatment induced an increase in DNA migration in the comet assay, but no enhancement of the MX-induced DNA damage was observed in the cells exposed to RF radiation.

(E) Sannino A, Zeni O, Sarti M, Romeo S, Reddy SB, Belisario MA, et al. (2011) Induction of adaptive response in human blood lymphocytes exposed to 900 MHz radiofrequency fields: influence of cell cycle. Int J Radiat Biol. 87: 993–999, 2011. (VT, AE, GT, IX)

Purpose: To investigate the influence of cell cycle on the adaptive response (AR) induced by the exposure of human blood lymphocytes to radiofrequency fields (RF). **Materials and methods:** Human peripheral blood lymphocytes in G(0)-, G(1)- or S-phase of the cell cycle were exposed for 20 hours to an adaptive dose (AD) of 900 MHz RF at an average specific absorption rate of 1.25 W/kg and then treated with a challenge dose (CD) of 100 ng/ml mitomycin C (MMC). Un-exposed and sham-exposed controls as well as cells treated with MMC alone were included in the study. The incidence of micronuclei (MN) was evaluated to determine the induction of AR. **Results:** The results indicated that the cells which were exposed to AD of RF in G(0)- and G(1)-phase of the cell cycle did not exhibit AR while such a response was observed when the cells were exposed to AD of RF in S-phase of the cell cycle. **Conclusions:** These results confirmed the observations reported in our previous investigation where AR was observed in human blood lymphocytes exposed to AD of RF in S-phase of the cell cycle and further suggested that the timing of AD exposure of RF is important to elicit AR.

(E) Sannino A, Zeni O, Romeo S, Massa R, Gialanella G, Grossi G, et al. Adaptive response in human blood lymphocytes exposed to non-ionizing radiofrequency fields: resistance to ionizing radiation-induced damage. J Radiat Res. 55: 210-217, 2014. (VT, AE, GT, IX)

The aim of this preliminary investigation was to assess whether human peripheral blood lymphocytes which have been pre-exposed to non-ionizing radiofrequency fields exhibit an adaptive response (AR) by resisting the induction of genetic damage from subsequent exposure to ionizing radiation. Peripheral blood lymphocytes from four healthy donors were stimulated with phytohemagglutinin for 24 h and then exposed for 20 h to 1950 MHz radiofrequency fields (RF, adaptive dose, AD) at an average specific absorption rate of 0.3 W/kg. At 48 h, the cells were subjected to a challenge dose (CD) of 1.0 or 1.5 Gy X-irradiation (XR, challenge dose, CD). After a 72 h total culture period, cells were collected to examine the incidence of micronuclei (MN). There was a significant decrease in the number of MN in lymphocytes exposed to RF + XR (AD + CD) as compared with those subjected to XR alone (CD). These observations thus suggested a RF-induced AR and induction of resistance to subsequent damage

from XR. There was variability between the donors in RF-induced AR. The data reported in our earlier investigations also indicated a similar induction of AR in human blood lymphocytes that had been pre-exposed to RF (AD) and subsequently treated with a chemical mutagen, mitomycin C (CD). Since XR and mitomycin-C induce different kinds of lesions in cellular DNA, further studies are required to understand the mechanism(s) involved in the RF-induced adaptive response.

(E) Sannino A, Zeni O, Romeo S, Massa R, Scarfi MR. Adverse and beneficial effect in Chinese hamster lung fibroblast cells following radiofrequency exposure. Bioelectromagnetics. 38: 245-254, 2017. (VT, AE, GT)

In this study, the effect of radiofrequency (RF) exposure to 1950 MHz, Universal Mobile Telecommunication System signal, was investigated in Chinese hamster lung fibroblast cell line (V79). Genotoxic and cytotoxic effects of 20-h exposure at specific absorption rate (SAR) values from 0.15 W/kg to 1.25 W/kg were measured by means of cytokinesis-block micronucleus (MN) assay. Exposure was carried out blinded under strictly controlled conditions of dosimetry and temperature. The effect of RF exposure alone at four SAR values was tested, that is, 0.15, 0.3, 0.6, and 1.25 W/kg. A statistically significant increase in MN frequency was found in cultures exposed to 0.15 and 0.3 W/kg ($P < 0.05$) compared to sham-exposed ones, in the absence of cytotoxicity. SAR values of 0.6 and 1.25 W/kg did not exert any effect. Moreover, to evaluate the ability of RF to exert protective effects with respect to a chemical mutagen, cell cultures were also pre-exposed for 20 h at 0.3 or 1.25 W/kg, and then treated with 500 ng/ml of mitomycin-C (MMC). A significant reduction in the frequency of MN was detected in cultures pre-exposed to 1.25 W/kg compared to cultures treated with MMC alone ($P < 0.05$), indicating induction of adaptive response. Such a decrease was not induced by pre-exposure at 0.3 W/kg SAR. Taken together, our results indicated that V79 is a sensitive cell model to evidence either adverse or beneficial effects of RF exposure, depending on experimental conditions applied.

(E) Sarimov R, Malmgren L.O.G., Markova, E., Persson, B.R.R.. Belyaev, I.Y. Nonthermal GSM microwaves affect chromatin conformation in human lymphocytes similar to heat shock. IEEE Trans Plasma Sci 32:1600-1608, 2004. (VT, AE, GT, LI, WS)

Here we investigated whether microwaves (MWs) of Global System for Mobile Communication (GSM) induce changes in chromatin conformation in human lymphocytes. Effects of MWs were studied at different frequencies in the range of 895-915 MHz in experiments with lymphocytes from seven healthy persons. Exposure was performed in transverse electromagnetic transmission line cell (TEM-cell) using a GSM test-mobile phone. All standard modulations included 2 W output power in the pulses, specific absorbed rate (SAR) being 5.4 mW/kg. Changes in chromatin conformation, which are indicative of stress response and genotoxic effects, were measured by the method of anomalous viscosity time dependencies (AVTD). Heat shock and treatment with the genotoxic agent camptothecin, were used as positive controls. 30-min exposure to MWs at 900 and 905 MHz resulted in statistically significant condensation of chromatin in lymphocytes from 1 of 3 tested donors. This condensation was similar to effects of heat shock within the temperature window of 40/spl deg/C-44/spl deg/C.

Analysis of pooled data from all donors showed statistically significant effect of 30-min exposure to MWs. Stronger effects of MWs was found following 1-h exposure. In replicated experiments, cells from four out of five donors responded to 905 MHz. Responses to 915 MHz were observed in cells from 1 out of 5 donors, $p<0.002$. Dependent on donor, condensation, 3 donors, or decondensation, 1 donor, of chromatin was found in response to 1-h exposure. Analysis of pooled data from all donors showed statistically significant effect of 1-h exposure to MWs. In cells from one donor, this effect was frequency-dependent ($p<0.01$). Effects of MWs correlated statistically significantly with effects of heat shock and initial state of chromatin before exposure. MWs at 895 and 915 MHz affected chromatin conformation in transformed lymphocytes. The conclusion-GSM microwaves under specific conditions of exposure affected human lymphocytes similar to stress response. The data suggested that the MW effects differ at various GSM frequencies and vary between doners.

(E) Sarkar S, Ali S, Behari J, Effect of low power microwave on the mouse genome: a direct DNA analysis. Mutat Res 320(1-2):141-147, 1994. (VO, LE, GT)

The potential mutagenic effect of low power microwave at the DNA sequence level in the mouse genome was evaluated by direct DNA analysis. Animals were exposed to microwave at a power density of 1 mW/cm2 for 2 h/day at a frequency of 2.45 GHz over a period of 120, 150 and 200 days. HinfI digested DNA samples from testis and brain of control and exposed animals were hybridized with a synthetic oligo probe (OAT 36) comprising nine repeats of 5'-GACA-3'. As compared to control animals, band patterns in exposed animals were found to be distinctly altered in the range of 7-8 kb which was also substantiated by densitometric analysis. Though the mechanism of this rearrangement is not yet clear, the results obtained at the present dose are of significance. This dose, which has been set as the safe limit for general public exposure by the Non-Ionizing Radiation Committee of the International Radiation Protection Association, may imply a need for (re)evaluation of the mutagenic potential of microwaves at the prescribed safe limit for the personnel and people who are being exposed..

(E) Scarfi MR, Lioi MB, d'Ambrosio G, Massa R, Zeni O, Pietro RD, et al. Genotoxic effects of mitomycn-C and microwave radiation on bovine lymphocytes. Electro Magneto Biol. 15, 99-107, 1996. (VT, AE, GT)

The aim of this work was to ascertain whether microwave radiation (frequency 9 GHz; specific absorption rate 70 mW/gr, exposure duration 10 min) may produce genotoxic effects, assessed by cytokinesis-block micro-nucleus (MN) assay, in bovine *(Bos taurus* L.) peripheral blood lymphocyte cultures. As positive control for the MN induction, mitomycin-C (MMC) was used: it is known that this alkylating agent has a potent genotoxic (radiomimetic) action. To evaluate possible cooperative effects, some cultures from microwave-exposed samples were treated with MMC. The results obtained indicate that the optimal dose of MMC to induce MN frequency increase in this species is 0.044 µg/ml and that microwave radiation induces a statistically significant increase of MN frequency both with and without MMC.

(NE) Scarfi MR, Romano M, Di Pietro R, Zeni O, Doria A, Gallerano GP, et al. THz exposure of whole blood for the study of biological effects on human lymphocytes. J Biol Phy. 29: 171-177, 2003. (VT, AE, GT)

The aim of the present study is to investigate the genotoxic effect of THz radiation in human peripheral blood lymphocytes following 20 minutes exposure to 1 mW average power Free Electron Laser radiation in the frequency range 120-140 GHz. For this purpose 9 healthy donors were employed and cytokinesis block technique was applied to study micronucleus frequency and cell proliferation. The results obtained indicate that all the electromagnetic conditions adopted so far do not alter the investigated parameters, suggesting absence of direct chromosomal damage and alteration of cell cycle kinetics (two tailed paired Student's test:p> 0.05 in all cases.

(NE) Scarfi MR, Fresegna AM, Villani P, Pinto R, Marino C, Sarti M, Altavista P, Sannino A, Lovisolo GA. Exposure to radiofrequency radiation (900 MHz, GSM signal) does not affect micronucleus frequency and cell proliferation in human peripheral blood lymphocytes: an interlaboratory study. Radiat Res. 165(6):655-663, 2006. (VT, AE, GT)

The objective of this study was to investigate whether 24 h exposure to radiofrequency electromagnetic fields similar to those emitted by mobile phones induces genotoxic effects and/or effects on cell cycle kinetics in cultured human peripheral blood lymphocytes. The effect of 900 MHz exposure (GSM signal) was evaluated at four specific absorption rates (SARs, 0, 1, 5 and 10 W/kg peak values). The exposures were carried out in wire patch cells under strictly controlled conditions of both temperature and dosimetry, and the induction of genotoxic effects was evaluated in lymphocyte cultures from 10 healthy donors by applying the cytokinesis-block micronucleus assay. Positive controls were provided by using mitomycin C. Two research groups were involved in the study, one at ENEA, Rome, and the other at CNR-IREA, Naples. Each laboratory tested five donors, and the resulting slides were scored by both laboratories. Following this experimental scheme, it was also possible to compare the results obtained by cross-scoring of slides. The results obtained provided no evidence for the existence of genotoxic or cytotoxic effects in the range of SARs investigated. These findings were confirmed in the two groups of five donors examined in the two laboratories and when the same slides were scored by two operators.

(NE) Schuermann D, Ziemann C, Barekati Z, Capstick M, Oertel A, Focke F, Murbach M, Kuster N, Dasenbrock C, Schär P. Assessment of Genotoxicity in Human Cells Exposed to Modulated Electromagnetic Fields of Wireless Communication Devices. Genes (Basel). 11(4):347, 2020. (VT, AE, GT)

Modulated electromagnetic fields (wEMFs), as generated by modern communication technologies, have raised concerns about adverse health effects. The International Agency for Research on Cancer (IARC) classifies them as "possibly carcinogenic to humans" (Group 2B), yet, the underlying molecular mechanisms initiating and promoting tumorigenesis remain elusive. Here, we comprehensively assess the impact of technologically relevant wEMF

modulations on the genome integrity of cultured human cells, investigating cell type-specificities as well as time- and dose-dependencies. Classical and advanced methodologies of genetic toxicology and DNA repair were applied, and key experiments were performed in two separate laboratories. Overall, we found no conclusive evidence for an induction of DNA damage nor for alterations of the DNA repair capacity in cells exposed to several wEMF modulations (i.e., GSM, UMTS, WiFi, and RFID). Previously reported observations of increased DNA damage after exposure of cells to GSM-modulated signals could not be reproduced. Experimental variables, presumably underlying the discrepant observations, were investigated and are discussed. On the basis of our data, we conclude that the possible carcinogenicity of wEMF modulations cannot be explained by an effect on genome integrity through direct DNA damage. However, we cannot exclude non-genotoxic, indirect, or secondary effects of wEMF exposure that may promote tumorigenesis in other ways.

(E) Schwarz C, Kratochvil E, Pilger A, Kuster N, Adlkofer F, Rüdiger HW. Radiofrequency electromagnetic fields (UMTS, 1,950 MHz) induce genotoxic effects in vitro in human fibroblasts but not in lymphocytes. Int Arch Occup Environ Health 81:755-767, 2008. (VT, AE, GT, CS)

OBJECTIVE: Universal Mobile Telecommunication System (UMTS) was recently introduced as the third generation mobile communication standard in Europe. This was done without any information on biological effects and genotoxic properties of these particular high-frequency electromagnetic fields. This is discomforting, because genotoxic effects of the second generation standard Global System for Mobile Communication have been reported after exposure of human cells in vitro. METHODS: Human cultured fibroblasts of three different donors and three different short-term human lymphocyte cultures were exposed to 1,950 MHz UMTS below the specific absorption rate (SAR) safety limit of 2 W/kg. The alkaline comet assay and the micronucleus assay were used to ascertain dose and time-dependent genotoxic effects. Five hundred cells per slide were visually evaluated in the comet assay and comet tail factor (CTF) was calculated. In the micronucleus assay 1,000 binucleated cells were evaluated per assay. The origin of the micronuclei was determined by fluorescence labeled anticentromere antibodies. All evaluations were performed under blinded conditions. RESULTS: UMTS exposure increased the CTF and induced centromere-negative micronuclei (MN) in human cultured fibroblasts in a dose and time-dependent way. Incubation for 24 h at a SAR of 0.05 W/kg generated a statistically significant rise in both CTF and MN ($P = 0.02$). At a SAR of 0.1 W/kg the CTF was significantly increased after 8 h of incubation ($P = 0.02$), the number of MN after 12 h ($P = 0.02$). No UMTS effect was obtained with lymphocytes, either unstimulated or stimulated with Phytohemagglutinin. CONCLUSION: UMTS exposure may cause genetic alterations in some but not in all human cells in vitro.

(E) Sekeroğlu V, Akar A, Sekeroğlu ZA. Cytotoxic and genotoxic effects of high-frequency electromagnetic fields (GSM 1800 MHz) on immature and mature rats. Ecotoxicol Environ Saf. 80:140-144, 2012. (VO, LE, GT, DE)

We investigated the cytogenotoxic effects of high frequency electromagnetic fields (HF-EMF) for 45 day and the effect of a recovery period of 15 day after exposure to EMF on bone marrow

cells of immature and mature rats. The animals in treatment groups were exposed to 1800 MHz EMF at SAR of 0.37 W/kg and 0.49 W/kg for 2h/day for 45 day. Two recovery groups were kept for a recovery period of 15 day without EMF after exposure to HF-EMF. Two control groups for both immature and mature rats were also included. Significant differences were also observed in chromosome aberrations (CA), micronucleus (MN) frequency, mitotic index (MI) and ratio of polychromatic erythrocytes (PCEs) in all treatment groups. The cytogenotoxic damage was more remarkable in immature rats and, the recovery period did not improve this damage in immature rats. Because much higher and irreversible cytogenotoxic damage was observed in immature rats than in mature rats, further studies are needed to understand effects of EMF on DNA damage and DNA repair, and to determine safe limits for environment and human, especially for children.

(E) Sekeroglu ZA, Akar A, Sekeroglu V. Evaluation of the cytogenetic damage in immature and mature rats exposed to 900 MHz radiofrequency electromagnetic fields. Int J Radiat Biol. 89:985-892, 2013. (VO, RE, GT, DE)

Purpose: One of the most important issues regarding radiofrequency electromagnetic fields (RF-EMF) is their effect on genetic material. Therefore, we investigated the cytogenotoxic effects of 900 MHz radiofrequency electromagnetic fields (RF-EMF) and the effect of a recovery period after exposure to RF-EMF on bone marrow cells of immature and mature rats. **Materials and methods:** The immature and mature rats in treatment groups were exposed to RF-EMF for 2 h/day for 45 days. Average electrical field values for immature and mature rats were 28.1 ± 4.8 V/m and 20.0 ± 3.2 V/m, respectively. Whole-body specific absorption rate (SAR) values for immature and mature rats were in the range of 0.38-0.78 W/kg, and 0.31-0.52 W/kg during the 45 days, respectively. Two recovery groups were kept for 15 days after RF-EMF exposure. **Results:** Significant differences were observed in chromosome aberrations (CA), micronucleus (MN) frequency, mitotic index (MI) and ratio of polychromatic erythrocytes (PCE) in all treatment and recovery groups. The cytogenotoxic damage in immature rats was statistically higher than the mature rats. The recovery period did not reduce the damage to the same extent as the corresponding control groups.**Conclusions:** The exposure of RF-EMF leads to cytotoxic and genotoxic damage in immature and mature rats. More sensitive studies are required to elucidate the possible carcinogenic risk of EMF exposure in humans, especially children.

(NE) Sekijima M, Takeda H, Yasunaga K, Sakuma N, Hirose H, Nojima T, Miyakoshi J. 2-GHz band CW and W-CDMA modulated radiofrequency fields have no significant effect on cell proliferation and gene expression profile in human cells. J Radiat Res. 51(3):277-284, 2010. (VT, AE, GE)

We investigated the mechanisms by which radiofrequency (RF) fields exert their activity, and the changes in both cell proliferation and the gene expression profile in the human cell lines, A172 (glioblastoma), H4 (neuroglioma), and IMR-90 (fibroblasts from normal fetal lung) following exposure to 2.1425 GHz continuous wave (CW) and Wideband Code Division Multiple Access (W-CDMA) RF fields at three field levels. During the incubation phase, cells were exposed at the specific absorption rates (SARs) of 80, 250, or 800 mW/kg with both CW and W-CDMA RF fields for up to 96 h. Heat shock treatment was used as the positive control. No significant differences in cell growth or viability were observed between any test group exposed to W-

CDMA or CW radiation and the sham-exposed negative controls. Using the Affymetrix Human Genome Array, only a very small (< 1%) number of available genes (ca. 16,000 to 19,000) exhibited altered expression in each experiment. The results confirm that low-level exposure to 2.1425 GHz CW and W-CDMA RF fields for up to 96 h did not act as an acute cytotoxicant in either cell proliferation or the gene expression profile. These results suggest that RF exposure up to the limit of whole-body average SAR levels as specified in the ICNIRP guidelines is unlikely to elicit a general stress response in the tested cell lines under these conditions.

(E) Semin IuA, Shvartsburg LK, Dubovik BV. [Changes in the secondary structure of DNA under the influence of external low-intensity electromagnetic field] Radiats Biol Radioecol 35(1):36-41, 1995. (VT, AE, GT, WS)

The effect of weak RF on the stability of DNA secondary structure was studied in vitro. DNA was exposed in the presence of glycine and formaldehyde. Aminomethynol compounds, which form in this medium, react with DNA bases at single-strand sites, which prevents recovery from damage to the DNA secondary structure. The damage accumulates during the incubation, and its amount can be estimated from the dynamics of thermal DNA denaturalization after RF or sham exposure. Samples were exposed in an anechoic chamber at 18°C at 10 different microwave frequencies simultaneously (4- to 8 GHz, 25 ms pulses, 0.4 to 0.7 mW/cm2 peak power, 1- to 6-Hz repetition rate, no heating). Parallel control samples were sham exposed in a shielded area in the same chamber. The experiments established that irradiation at 3 or 4 Hz and 0.6 mW/cm2 peak power clearly increased the accumulated damage to the DNA secondary structure (P< .00001). However, changing the pulse repetition rate to 1, 5, 6 Hz, as well as changing the peak power to 0.4 or 0.7 mW/cm2, eliminated the effect entirely. Thus, the effect occurred only within narrow 'windows' of the peak intensities and modulation frequencies.

(NE) Senturk E, Tugrul S, Doğan R, Eren SB, Ozturan O, Koçyiğit A, Kesgin S. Effect of radiofrequency on DNA damage and oxidative status in patients with turbinate uypertrophy. Indian J Otolaryngol Head Neck Surg. 71(Suppl 3):1810-1815, 2019. (HU, LE, GT, OX)

The radiofrequency devices that are used generate radiofrequency in the frequency range of 1.5 and 2.5 MHz. This study aims to demonstrate whether systematic oxidative status and DNA are influenced in this frequency range. In study, 27 patients who received radiofrequency treatment on inferior turbinate as they were diagnosed with inferior turbinate hypertrophy. DNA damage was assessed by alkaline comet assay in peripheral lymphocyte cells. Plasma levels of total antioxidant status (TAS), total oxidative status (TOS) were determined by using an automated measurement method and oxidative stress index (OSI) was calculated (OSI was calculated as: OSI = (TOS/TAS) × 100). There were increased in the OSI and TOS values on days 1 and 15 as compared to the samples taken before the radiofrequency administration. Significant decreases were seen in TAS values on days 1 and 15. As for the DNA damage, no significant differences were found on day 15 compared to the preoperative values even though there was a statistically insignificant increase on day 1. Administration of radiofrequency radiation on inferior turbinates results in increased

oxidative stress in the acute period and a decrease in the anti-oxidative system. Although this effect causes a slight increase in the DNA damage in the early post-operative period, the damage is restored to the pre-operative levels on day 15. Therefore, we believe that a more conservative approach should be selected for radiofrequency treatment instead of using it routinely.

(E) Shah C, Nair A, Naik M2, Bakshi S. Cell phone radiation and genomic damage: In vitro exposure and assessment. Int J Innnov Res Sci Eng Technol. 4: 401-405, 2015. (VT, AE, GT)

The health concerns have been raised following the enormous increase in the use of wireless mobile telephones throughout the world. According to the International Agency for Research in Cancer (IARC), a part of World Health Organization (WHO) has designated cell phone radiation i.e. non-ionizing radiofrequency radiation as „Possible Human Carcinogen" [Class 2B] in May, 2011. It is believed that the effect is caused because of the electromagnetic frequency generated by the radio frequency which couples with the human tissues which results in induced electric and magnetic fields that cause field distribution in the body. Thus, human body acts as an antenna that receives electromagnetic waves externally. Therefore, effect of radiofrequency radiation needs to be studied by examining the target tissues that are directly exposed to electromagnetic waves i.e. brain tissue, circulating blood, and facial muscles. In this study, circulating blood was taken as target tissue and subjected to cell phone radiation in vitro and following short term cultures metaphase chromosomes were analyzed for frequency of breakage. The results indicated significant increase in chromosomal damage at higher power level and longer exposure times.

(E) Shahin S, Singh VP, Shukla RK, Dhawan A, Gangwar RK, Singh SP, Chaturvedi CM. 2.45 GHz microwave irradiation-induced oxidative stress affects implantation or pregnancy in mice, Mus musculus. Appl Biochem Biotechnol. 169(5):1727-1751, 2013. (VO, LE, GT, LI, OX)

The present experiment was designed to study the 2.45 GHz low-level microwave (MW) irradiation-induced stress response and its effect on implantation or pregnancy in female mice. Twelve-week-old mice were exposed to MW radiation (continuous wave for 2 h/day for 45 days, frequency 2.45 GHz, power density=0.033549 mW/cm(2), and specific absorption rate=0.023023 W/kg). At the end of a total of 45 days of exposure, mice were sacrificed, implantation sites were monitored, blood was processed to study stress parameters (hemoglobin, RBC and WBC count, and neutrophil/lymphocyte (N/L) ratio), the brain was processed for comet assay, and plasma was used for nitric oxide (NO), progesterone and estradiol estimation. Reactive oxygen species (ROS) and the activities of ROS-scavenging enzymes- superoxide dismutase, catalase, and glutathione peroxidase-were determined in the liver, kidney and ovary. We observed that implantation sites were affected significantly in MW-irradiated mice as compared to control. Further, in addition to a significant increase in ROS, hemoglobin ($p<0.001$), RBC and WBC counts ($p<0.001$), N/L ratio ($p<0.01$), DNA damage ($p<0.001$) in brain cells, and plasma estradiol concentration ($p<0.05$), a significant decrease was observed in NO level ($p<0.05$) and antioxidant enzyme activities of MW-exposed mice. Our

findings led us to conclude that a low level of MW irradiation-induced oxidative stress not only suppresses implantation, but it may also lead to deformity of the embryo in case pregnancy continues. We also suggest that MW radiation-induced oxidative stress by increasing ROS production in the body may lead to DNA strand breakage in the brain cells and implantation failure/resorption or abnormal pregnancy in mice.

(E) Shahin NN, El-Nabarawy NA, Gouda AS, Mégarbane B. The protective role of spermine against male reproductive aberrations induced by exposure to electromagnetic field - An experimental investigation in the rat. Toxicol Appl Pharmacol. 370:117-130, 2019. (VO, LE, GT, OX, RP)

The exponentially increasing use of electromagnetic field (EMF)-emitting devices imposes substantial health burden on modern societies with particular concerns of male infertility. Limited studies have addressed the modulation of this risk by protective agents. We investigated the hazardous effects of rat exposure to EMF (900 MHz, 2 h/day for 8 weeks) on male fertility and evaluated the possible protective effect of the polyamine, spermine, against EMF-induced alterations. Exposure to EMF significantly decreased sperm count, viability and motility, and increased sperm deformities. EMF-exposed rats exhibited significant reductions in serum inhibin B and testosterone along with elevated activin A, follicle-stimulating hormone, luteinizing hormone and estradiol concentrations. Testicular steroidogenic acute regulatory protein (StAR), c-kit mRNA expression and testicular activities of the key androgenic enzymes 3β- and 17β-hydroxysteroid dehydrogenases were significantly attenuated following exposure to EMF. Exposure led to testicular lipid peroxidation, decreased catalase and glutathione peroxidase activities and triggered nuclear factor-kappa B p65, inducible nitric oxide synthase, cyclooxygenase-2 and caspase-3 overexpression. EMF-exposed rats showed testicular DNA damage as indicated by elevated comet parameters. Spermine administration (2.5 mg/Kg/day intraperitoneally for 8 weeks) prevented EMF-induced alterations in the sperm and hormone profiles, StAR and c-kit expression and androgenic enzyme activities. Spermine hampered EMF-induced oxidative, inflammatory, apoptotic and DNA perturbations. Histological and histomorphometric analysis of the testes supported all biochemical findings. In conclusion, rat exposure to EMF disrupts sperm and hormone profiles with underlying impairment of steroidogenesis and spermatogenesis. Spermine confers protection against EMF-associated testicular and reproductive aberrations, at least in part, via antioxidant, anti-inflammatory and anti-apoptotic mechanisms.

(E) Sharma S, Shukla S. Effect of electromagnetic radiation on redox status, acetylcholine esterase activity and cellular damage contributing to the diminution of the brain working memory in rats. J Chem Neuroanat. 106:101784, 2020. (VO, LE, GT, OX)

Behavioral impairments are the most pragmatic outcome of long-term mobile uses but the underlying causes are still poorly understood. Therefore, the Aim of the present study to determine the possible mechanism of mobile induced behavioral alterations by observing redox status, cholinesterase activity, cellular, genotoxic damage and cognitive alterations in rat hippocampus. This study was carried out on 24 male Wistar rats, randomly divided into four groups (n = 6 in each group): group I consisted of sham-exposed (control) rats, group II-IV consisted of rats exposed to microwave radiation

(900 MHz) at different time duration 1 h, 2 h, and 4 h respectively for 90 days. After 90 days of exposure, rats were assessing learning ability by using T-Maze. A significantly increased level of malondialdehyde (MDA) with concomitantly depleted levels of superoxide dismutase (SOD), catalase (CAT) and redox enzymes (GSH, GPX, GR, GST, G-6PDH) indicated an exposure of mobile emitted EMR induced oxidative stress by the depleted redox status of brain cells. The depletion in the acetylcholinesterase (AChE) level reveals altered neurotransmission in brain cells. Resultant cellular degeneration was also observed in the radiation-exposed hippocampus. Conclusively, the present study revealed that microwave radiation induces oxidative stress, depleted redox status, and causes DNA damage with the subsequent reduction in working memory in a time-dependent manner. This study provides insight over the associative reciprocity between redox status, cellular degeneration and reduced cholinergic activity, which presumably leads to the behavioral alterations following mobile emitted electromagnetic radiation.

(E) Sharma A , Shrivastava S, Shukla S. Exposure of Radiofrequency Electromagnetic Radiation on Biochemical and Pathological Alterations. Neurol India 68(5):1092-1100, 2020. (VO, LE, GT)

Introduction: In the era of globalization, too much dependency on mobile phones is a cause of concern. **Objective:** The present study was designed to evaluate the risk assessment of microwave radiation (MWR) at 1800 MHz frequency and specific absorption rate 0.433 (W/kg) on male Wistar rats. **Methodology:** Animals were divided into two groups: the first group is the control group, and the second group was exposed to 1800 MHz radiation for 90 days at 4 h/5 days/week in a month. **Results:** Chronic exposure of MWR may alter GSH homeostasis due to alteration in various GSH cycle regulating enzymes such as GR, GPx, GST, and G6PDH which showed an imbalance in GSH content and causes an increase in the oxidative stress and release of inflammatory cytokines. A remarkable increase in the DNA damage was seen due to disorganization and pyknosis of neurons in exposed animal's brain when compared with the control group ($P \leq 0.05$). There was also a significant decline in AChE level. **Conclusion:** The study concludes that MWR may cause neurochemical and pathophysiological damage by initiating the inflammatory process in various brain regions, especially in hippocampus and cerebral cortex. These effects are further associated with a remarkable elevation in the genotoxicity of neurons with reference to the control group..

(E) Sharma S, Bahel S, Katnoria JK. Evaluation of oxidative stress and genotoxicity of 900 MHz electromagnetic radiations using Trigonella foenum-graecum test system. Protoplasma 260(1):209-224, 2023. (VO, LE, GT)

Unprecedented growth in the communication sector and expanded usage of the number of wireless devices in the past few decades have resulted in a tremendous increase in emissions of non-ionizing electromagnetic radiations (EMRs) in the environment. The widespread EMRs have induced many significant changes in biological systems leading to oxidative stress as well as DNA damage. Considering this, the present study was planned to study the effects of EMRs at 900 MHz frequency with the power density of 10.0 dBm (0.01 W) at variable exposure periods (0.5 h, 1 h, 2 h, 4 h, and 8 h per day for 7 days) on percentage germination, morphological

characteristics, protein content, lipid peroxidation in terms of malondialdehyde content (MDA), and antioxidant defense system of Trigonella foenum-graecum test system. The genotoxicity was also evaluated using similar conditions. It was observed that EMRs significantly decreased the germination percentage at an exposure time of 4 h and 8 h. Fresh weight and dry weight of root and shoot did not show significant variations, while the root and shoot length have shown significant variations for 4 h and 8 h exposure period. Further, EMRs enhanced MDA indicating lipid peroxidation. In response to exposure of EMRs, there was a significant up-regulation in the activities of enzymes such as ascorbate peroxidase (APX), superoxide dismutase (SOD), glutathione-S-transferase (GST), guaiacol peroxidase (POD), and glutathione reductase (GR) in the roots and shoots of Trigonella-foenum graecum. The genotoxicity study showed the induction of chromosomal aberrations in root tip cells of the Trigonella foenum-graecum test system. The present study revealed the induction of oxidative stress and genotoxicity of EMRs exposure in the test system.

(E) Shckorbatov YG, Pasiuga VN, Kolchigin NN, Grabina VA, Batrakov DO, Kalashnikov VV, et al. The influence of differently polarized microwave radiation on chromatin in human cells. Int J Radiat Biol. 85: 322-329, 2009. (VT, AE, GT, WS)

Purpose: To determine the possible biological effects of differently polarised microwave radiation on the chromatin state in human cells. **Materials and methods:** Isolated human buccal epithelium cells were irradiated by microwaves of frequency f = 35 GHz and surface power density E = 30 microW/cm(2). The state of chromatin in human cells was determined by methods of light and electron microscopy. The state of cell membranes was evaluated by the method of vital indigo carmine staining. **Results:** The microwave-induced condensation of chromatin in human cells is revealed. Degree of microwave-induced condensation depends on the state of polarisation of electromagnetic wave: In some cases left circularly polarised waves induce less effect than linearly polarised radiation. The linearly polarised electromagnetic waves induce cell membrane damage revealed by increase of cell staining. The data obtained are discussed in connection with mechanisms of biological effects of electromagnetic fields. **Conclusion:** The data obtained in this work demonstrate important biological effects of monochromatic microwave irradiation at 35 GHz. Low-level microwave irradiation induces chromatin condensation in human cells and damages of cell membranes.

(E) Shckorbatov YG, Pasiuga VN, Goncharuk EI, Petrenko TP, Grabina VA, Kolchigin NN, et al. Effects of differently polarized microwave radiation on the microscopic structure of the nuclei in human fibroblasts. J Zhejiang Univ Sci B. 11: 801-805, 2010. (VT, AE, GT)

To investigate the influence of microwave radiation on the human fibroblast nuclei, the effects of three variants of electromagnetic wave polarization, linear and left-handed and right-handed elliptically polarized, were examined. Experimental conditions were: frequency (f) 36.65 GHz, power density (P) at the surface of exposed object 1, 10, 30, and 100 μW/cm(2), exposure time 10 s. Human fibroblasts growing in a monolayer on a cover slide were exposed to microwave electromagnetic radiation. The layer of medium that covered cells during microwave exposure was about 1 mm thick. Cells were stained immediately after irradiation by 2% (w/v) orcein solution in 45% (w/v) acetic acid. Experiments were made at room temperature (25 °C), and

control cell samples were processed in the same conditions. We assessed heterochromatin granule quantity (HGQ) at 600× magnification. Microwave irradiation at the intensity of 1 μW/cm(2) produced no effect, and irradiation at the intensities of 10 and 100 μW/cm(2) induced an increase in HGQ. More intense irradiation induced more chromatin condensation. The right-handed elliptically polarized radiation revealed more biological activity than the left-handed polarized one.

(NE) Shi D, Zhu C, Lu R, Mao S, Qi Y. Intermediate frequency magnetic field generated by a wireless power transmission device does not cause genotoxicity in vitro. Bioelectromagnetics. 35(7):512-518, 2014. (VT, AE, GT)

The aim of this study was to evaluate effects of intermediate frequency magnetic fields (IFMF) generated by a wireless power transmission (WPT) based on magnetic resonance from the perspective of cellular genotoxicity on cultured human lens epithelial cells (HLECs). We evaluated the effects of exposure to 90 kHz magnetic fields at 93.36 μT on cellular genotoxicity in vitro for 2 and 4 h. The magnetic flux density is approximately 3.5 times higher than the reference level recommended by the International Commission on Non-Ionizing Radiation Protection (ICNIRP) guidelines. For assessment of genotoxicity, we studied cellular proliferation, apoptosis and DNA damage by Cell Counting Kit-8 (CCK-8) assay, flow cytometry analysis, alkaline comet assay and phosphorylated histone H2AX (γH2AX) foci formation test. We did not detect any effect of a 90 kHz IFMF generated by WPT based on magnetic resonance on cell proliferation, apoptosis, comet assay, and γH2AX foci formation test. Our results indicated that exposure to 90 kHz IFMF generated by WPT based on magnetic resonance at 93.36 μT for 2 and 4 h does not cause detectable cellular genotoxicity.

(NE) Silva V, Hilly O, Strenov Y, Tzabari C, Hauptman Y, Feinmesser R. Effect of cell phone-like electromagnetic radiation on primary human thyroid cells. Int J Radiat Biol. 92: 107-115, 2016. (VT, AE, GE)

Purpose: To evaluate the potential carcinogenic effects of radiofrequency energy (RFE) emitted by cell phones on human thyroid primary cells. **Materials and methods:** Primary thyroid cell culture was prepared from normal thyroid tissue obtained from patients who underwent surgery at our department. Subconfluent thyroid cells were irradiated under different conditions inside a cell incubator using a device that simulates cell phone-RFE. Proliferation of control and irradiated cells was assessed by the immunohistochemical staining of antigen Kiel clone-67 (Ki-67) and tumor suppressor p53 (p53) expression. DNA ploidy and the stress biomarkers heat shock protein 70 (HSP70) and reactive oxygen species (ROS) was evaluated by fluorescence-activated cell sorting (FACS).**Results:** Our cells highly expressed thyroglobulin (Tg) and sodium-iodide symporter (NIS) confirming the origin of the tissue. None of the irradiation conditions evaluated here had an effect neither on the proliferation marker Ki-67 nor on p53 expression. DNA ploidy was also not affected by RFE, as well as the expression of the biomarkers HSP70 and ROS. **Conclusion:** Our conditions of RFE exposure seem to have no potential carcinogenic effect on human thyroid cells. Moreover, common biomarkers usually

associated to environmental stress also remained unchanged. We failed to find an association between cell phone-RFE and thyroid cancer. Additional studies are recommended.

(E) Singh KV, Prakash C, Nirala JP, Nanda RK, Rajamani P. Acute radiofrequency electromagnetic radiation exposure impairs neurogenesis and causes neuronal DNA damage in the young rat brain. Neurotoxicology 94:46-58, 2023. (VO, AE, GT)

A mobile phone is now a commonly used device for digital media and communication among all age groups. Young adolescents use it for longer durations, which exposes them to radiofrequency electromagnetic radiation (RF-EMR). This exposure can lead to neuropsychiatric changes. The underlying cellular mechanism behind these changes requires detailed investigation. In the present study, we investigated the effect of RF-EMR emitted from mobile phones on young adolescent rat brains. Wistar rats (5 weeks, male) were exposed to RF-EMR signal (2115 MHz) at a head average specific absorption rate (SAR) of 1.51 W/kg continuously for 8 h. Higher level of lipid peroxidation, carbon-centered lipid radicals, and single-strand DNA damage was observed in the brain of rat exposed to RF-EMR. The number of BrdU-positive cells in the dentate gyrus (DG) decreased in RF-EMR-exposed rats, indicating reduced neurogenesis. RF-EMR exposure also induced degenerative changes and neuronal loss in DG neurons but had no effect on the CA3 and CA1 neurons of the hippocampus and cerebral cortex. The activity of Pro-caspase3 did not increase upon exposure in any of the brain regions, pointing out that degeneration observed in the DG region is not dependent on caspase activation. Results indicate that short-term acute exposure to RF-EMR induced the generation of carbon-centered lipid radicals and nuclear DNA damage, both of which likely played a role in the impaired neurogenesis and neuronal degeneration seen in the young brain's hippocampus region. The understanding of RF-EMR-induced alteration in the brain at the cellular level will help develop appropriate interventions for reducing its adverse impact.

Sisakht M, Darabian M, Mahmoodzadeh A, Bazi A, Shafiee SM, Mokarram P, Khoshdel Z. The role of radiation induced oxidative stress as a regulator of radio-adaptive responses. Int J Radiat Biol. 2020 Feb 7:1-16. (review)

Purpose: Various sources of radiation including radiofrequency, electromagnetic radiation (EMR), low- dose X-radiation, low-level microwave radiation and ionizing radiation (IR) are indispensable parts of modern life. In the current review, we discussed the adaptive responses of biological systems to radiation with a focus on the impacts of radiation-induced oxidative stress (RIOS) and its molecular downstream signaling pathways.**Materials and methods:** A comprehensive search was conducted in Web of Sciences, PubMed, Scopus, Google Scholar, Embase, and Cochrane Library. Keywords included Mesh terms of "radiation," "electromagnetic radiation," "adaptive immunity," "oxidative stress," and "immune checkpoints." Manuscripts published up until December 2019 were included.**Results:** RIOS induces various molecular adaptors connected with adaptive responses in radiation exposed cells. One of these adaptors includes p53 which promotes various cellular signaling pathways. RIOS also activates the intrinsic apoptotic pathway by depolarization of the mitochondrial membrane potential and activating the caspase apoptotic cascade. RIOS is also involved in radiation-induced proliferative responses through interaction with mitogen-activated

protein kinases (MAPks) including p38 MAPK, ERK, and c-Jun N-terminal kinase (JNK). Protein kinase B (Akt)/phosphoinositide 3-kinase (PI3K) signaling pathway has also been reported to be involved in RIOS-induced proliferative responses. Furthermore, RIOS promotes genetic instability by introducing DNA structural and epigenetic alterations, as well as attenuating DNA repair mechanisms. Inflammatory transcription factors including macrophage migration inhibitory factor (MIF), nuclear factor κB (NF-κB), and signal transducer and activator of transcription-3 (STAT-3) paly major role in RIOS-induced inflammation.**Conclusion:** In conclusion, RIOS considerably contributes to radiation induced adaptive responses. Other possible molecular adaptors modulating RIOS-induced responses are yet to be divulged in future studies.

(E)Smith-Roe SL, Wyde ME, Stout MD, Winters JW, Hobbs CA, Shepard KG, Green AS, Kissling GE, Shockley KR, Tice RR, Bucher JR, Witt KL. Evaluation of the genotoxicity of cell phone radiofrequency radiation in male and female rats and mice following subchronic exposure. Environ Mol Mutagen. 61(2):276-290, 2020. (VO, LE, GT)

The National Toxicology Program tested two common radiofrequency radiation (RFR) modulations emitted by cellular telephones in a 2-year rodent cancer bioassay that included interim assessments of additional animals for genotoxicity endpoints. Male and female Hsd:Sprague Dawley SD rats and B6C3F1/N mice were exposed from Gestation day 5 or Postnatal day 35, respectively, to code division multiple access (CDMA) or global system for mobile modulations over 18 hr/day, at 10-min intervals, in reverberation chambers at specific absorption rates of 1.5, 3, or 6 W/kg (rats, 900 MHz) or 2.5, 5, or 10 W/kg (mice, 1,900 MHz). After 19 (rats) or 14 (mice) weeks of exposure, animals were examined for evidence of RFR-associated genotoxicity using two different measures. Using the alkaline (pH > 13) comet assay, DNA damage was assessed in cells from three brain regions, liver cells, and peripheral blood leukocytes; using the micronucleus assay, chromosomal damage was assessed in immature and mature peripheral blood erythrocytes. Results of the comet assay showed significant increases in DNA damage in the frontal cortex of male mice (both modulations), leukocytes of female mice (CDMA only), and hippocampus of male rats (CDMA only). Increases in DNA damage judged to be equivocal were observed in several other tissues of rats and mice. No significant increases in micronucleated red blood cells were observed in rats or mice. In conclusion, these results suggest that exposure to RFR is associated with an increase in DNA damage.

(E) Sokolovic D, Djordjevic B, Kocic G, Stoimenov TJ, Stanojkovic Z, Sokolovic DM, et al. The effects of melatonin on oxidative stress parameters and DNA fragmentation in testicular tissue of rats exposed to microwave radiation. Adv Clin Exp Med. 24(3):429-436, 2015. (VO, LE, GT, OX, RP)

BACKGROUND: Microwaves from mobile phones are one of the environmental toxicants that are capable of compromising male fertility by inducing oxidative stress and apoptosis in the testes. Melatonin is a lipophilic tryptophan indole amine and a potent antioxidant. OBJECTIVES: The aim of the study was to evaluate the effect of melatonin treatment on oxidative stress parameters and DNA fragmentation in the testicular tissue of rats exposed to microwave radiation (4 h/day). MATERIAL AND METHODS: Adult Wistar rats were divided in 4 groups: I - treated with saline; II - treated with melatonin; III - exposed to microwaves; IV -

exposed to microwaves and treated with melatonin. The melatonin (2 mg/kg ip) was administered daily. The animals were sacrificed after 20, 40 and 60 days. RESULTS: Melatonin treatment prevented previously registered increases in malondialdehyde after only 20 days. Furthermore, it reversed the effects of microwave exposure on xanthine oxidase (after 40 days) and acid-DNase activity (after 20 days). However, neither protein carbonyl content nor catalase and alkaline Dnase activity were changed due to melatonin treatment. CONCLUSIONS: Melatonin exerts potent antioxidant effects in the testes of rats exposed to microwaves by decreasing the intensity of oxidative stress; it also reduces DNA fragmentation.

(E) Son Y, Park H-J, Jeong YJ, Choi H-D, Kim N, Lee H-J. Long-term radiofrequency electromagnetic fields exposure attenuates cognitive dysfunction in 5×FAD mice by regulating microglial function. Neural Regen Res 18(11):2497-2503, 2023. (VO, LE, GE)

We have previously found that long-term effects of exposure to radiofrequency electromagnetic fields in 5×FAD mice with severe late-stage Alzheimer's disease reduced both amyloid-β deposition and glial activation, including microglia. To examine whether this therapeutic effect is due to the regulation of activated microglia, we analyzed microglial gene expression profiles and the existence of microglia in the brain in this study. 5×FAD mice at the age of 1.5 months were assigned to sham- and radiofrequency electromagnetic fields-exposed groups and then animals were exposed to 1950 MHz radiofrequency electromagnetic fields at a specific absorption rate of 5 W/kg for 2 hours/day and 5 days/week for 6 months. We conducted behavioral tests including the object recognition and Y-maze tests and molecular and histopathological analysis of amyloid precursor protein/amyloid-beta metabolism in brain tissue. We confirmed that radiofrequency electromagnetic field exposure for 6 months ameliorated cognitive impairment and amyloid-β deposition. The expression levels of Iba1 (pan-microglial marker) and colony-stimulating factor 1 receptor (CSF1R; regulates microglial proliferation) in the hippocampus in 5×FAD mice treated with radiofrequency electromagnetic fields were significantly reduced compared with those of the sham-exposed group. Subsequently, we analyzed the expression levels of genes related to microgliosis and microglial function in the radiofrequency electromagnetic fields-exposed group compared to those of a CSF1R inhibitor (PLX3397)-treated group. Both radiofrequency electromagnetic fields and PLX3397 suppressed the levels of genes related to microgliosis (Csf1r, CD68, and Ccl6) and pro-inflammatory cytokine interleukin-1β. Notably, the expression levels of genes related to microglial function, including Trem2, Fcgr1a, Ctss, and Spi1, were decreased after long-term radiofrequency electromagnetic field exposure, which was also observed in response to microglial suppression by PLX3397. These results showed that radiofrequency electromagnetic fields ameliorated amyloid-β pathology and cognitive impairment by suppressing amyloid-β deposition-induced microgliosis and their key regulator, CSF1R.

(E) Soubere Mahamoud Y, Aite M, Martin C, Zhadobov M, Sauleau R, Le Dréan Y, Habauzit D. Additive Effects of Millimeter Waves and 2-Deoxyglucose Co-Exposure on the Human Keratinocyte Transcriptome.PLoS One.;11(8):e0160810, 2016. (VT, AE, GE, IX)

Millimeter Waves (MMW) will be used in the next-generation of high-speed wireless technologies, especially in future Ultra-Broadband small cells in 5G cellular networks. Therefore, their biocompatibilities must be evaluated prior to their massive deployment. Using a microarray-based approach, we analyzed modifications to the whole genome of a human keratinocyte model that was exposed at 60.4 GHz-MMW at an incident power density (IPD) of 20 mW/cm2 for 3 hours in athermic conditions. No keratinocyte transcriptome modifications were observed. We tested the effects of MMWs on cell metabolism by co-treating MMW-exposed cells with a glycolysis inhibitor, 2-deoxyglucose (2dG, 20 mM for 3 hours), and whole genome expression was evaluated along with the ATP content. We found that the 2dG treatment decreased the cellular ATP content and induced a high modification in the transcriptome (632 coding genes). The affected genes were associated with transcriptional repression, cellular communication and endoplasmic reticulum homeostasis. The MMW/2dG co-treatment did not alter the keratinocyte ATP content, but it did slightly alter the transcriptome, which reflected the capacity of MMW to interfere with the bioenergetic stress response. The RT-PCR-based validation confirmed 6 MMW-sensitive genes (SOCS3, SPRY2, TRIB1, FAM46A, CSRNP1 and PPP1R15A) during the 2dG treatment. These 6 genes encoded transcription factors or inhibitors of cytokine pathways, which raised questions regarding the potential impact of long-term or chronic MMW exposure on metabolically stressed cells.

(E) Souza LD, Cerqueira ED, Meireles JR. Assessment of nuclear abnormalities in exfoliated cells from the oral epithelium of mobile phone users. Electromagn Biol Med. 33(2):98-102, 2014. (HU, LE, GE)

Transmission and reception of mobile telephony signals take place through electromagnetic wave radiation, or electromagnetic radiofrequency fields, between the mobile terminal and the radio base station. Based on reports in the literature on adverse effects from exposure to this type of radiation, the objective of this study was to evaluate the genotoxic and cytotoxic potential of such exposure, by means of the micronucleus test on exfoliated cells from the oral epithelium. The sample included 45 individuals distributed in 3 groups according to the amount of time in hours per week (t) spent using mobile phones: group I, $t > 5$ h; group II, $t > 1$ h and ≤ 5 h; and group III, $t \leq 1$ h. Cells from the oral mucosa were analyzed to assess the numbers of micronuclei, broken egg structures and degenerative nuclear abnormalities indicative of apoptosis (condensed chromatin, karyorrhexis and pyknosis) or necrosis (karyolysis in addition to these changes). The occurrences of micronuclei and degenerative nuclear abnormalities did not differ between the groups, but the number of broken egg (structures that may be associated with gene amplification) was significantly greater in the individuals in group I ($p < 0.05$).

(E) Spandole-Dinu S, Catrina A-M, Voinea OC, Andone A, Radu S , Haidoiu C, Călborean O , Popescu DM , Suhăianu V, Baltag O , Tută L, Rosu G. Pilot Study of the Long-Term Effects of Radiofrequency Electromagnetic Radiation Exposure on the Mouse Brain. Int. J. Environ. Res. Public Health 20(4):3025, 2023. (VO, LE, GT, EP)

The increasing radiofrequency (RF) electromagnetic radiation pollution resulting from the development and use of technologies utilizing RF has sparked debate about the possible biological effects of said radiation. Of particular concern is the potential impact on the brain, due to the close proximity of communication devices to the head. The main aim of this study was to examine the effects of long-term exposure to RF on the brains of mice in a real-life scenario simulation compared to a laboratory setting. The animals were exposed continuously for 16 weeks to RF using a household Wi-Fi router and a laboratory device with a frequency of 2.45 GHz, and were compared to a sham exposed group. Before and after exposure, the mice underwent behavioral tests (open-field test and Y-maze); at the end of the exposure period, the brain was harvested for histopathological analysis and assessment of DNA methylation levels. Long-term exposure of mice to 2.45 GHz RF radiation increased their locomotor activity, yet did not cause significant structural or morphological changes in their brains. Global DNA methylation was lower in exposed mice compared to sham mice. Further research is needed to understand the mechanisms behind these effects and to understand the potential effects of RF radiation on brain function.

(NE) Speit G, Schütz P, Hoffmann H. Genotoxic effects of exposure to radiofrequency electromagnetic fields (RF-EMF) in cultured mammalian cells are not independently reproducible. Mutat Res. 626(1-2):42-47, 2007. (VT, AE, GT)

Conflicting results have been published regarding the induction of genotoxic effects by exposure to radiofrequency electromagnetic fields (RF-EMF). Using the comet assay, the micronucleus test and the chromosome aberration test with human fibroblasts (ES1 cells), the EU-funded "REFLEX" project (Risk Evaluation of Potential Environmental Hazards From Low Energy Electromagnetic Field Exposure Using Sensitive in vitro Methods) reported clearly positive effects for various exposure conditions. Because of the ongoing discussion on the biological significance of the effects observed, it was the aim of the present study to independently repeat the results using the same cells, the same equipment and the same exposure conditions. We therefore exposed ES1 cells to RF-EMF (1800 MHz; SAR 2 W/kg, continuous wave with intermittent exposure) for different time periods and then performed the alkaline (pH>13) comet assay and the micronucleus test (MNT). For both tests, clearly negative results were obtained in independently repeated experiments. We also performed these experiments with V79 cells, a sensitive Chinese hamster cell line that is frequently used in genotoxicity testing, and also did not measure any genotoxic effect in the comet assay and the MNT. Appropriate measures of quality control were considered to exclude variations in the test performance, failure of the RF-EMF exposure or an evaluation bias. The reasons for the difference between the results reported by the REFLEX project and our experiments remain unclear.

(NE) Speit G, Richard Gminski, Tauber R. Genotoxic effects of exposure to radiofrequency electromagnetic fields (RF-EMF) in HL-60 cells are not reproducible. Mutat Res 755(2):163-166, 2013. (VT, AE, GT)

Conflicting results have been published regarding the induction of genotoxic effects by exposure to radiofrequency electromagnetic fields (RF-EMF). Various results indicating a genotoxic

potential of RF-EMF were reported by the collaborative EU-funded REFLEX (Risk Evaluation of Potential Environmental Hazards From Low Energy Electromagnetic Field Exposure Using Sensitive in vitro Methods) project. There has been a long-lasting scientific debate about the reliability of the reported results and an attempt to reproduce parts of the results obtained with human fibroblasts failed. Another part of the REFLEX study was performed in Berlin with the human lymphoblastoid cell line HL-60; genotoxic effects of RF-EMF were measured by means of the comet assay and the micronucleus test. The plausibility and reliability of these results were also questioned. In order to contribute to a clarification of the biological significance of the reported findings, a repeat study was performed, involving scientists of the original study. Comet-assay experiments and micronucleus tests were performed under the same experimental conditions that had led to genotoxic effects in the REFLEX study. Here we report that the attempts to reproduce the induction of genotoxic effects by RF-EMF in HL-60 cells failed. No genotoxic effects of RF-EMF were measured in the repeat experiments. We could not find an explanation for the conflicting results. However, the negative repeat experiments suggest that the biological significance of genotoxic effects of RF-EMF reported by the REFLEX study should be re-assessed.

(E) Srujana Aravinda VS, Kandregula CR, Muppa R, Krishna MM, Nikitha BS, Yenni M. A cross-sectional and histological analysis to understand the cytological effects of cell phone radiation on buccal mucosa of children. J Indian Soc Pedod Prev Dent 40(1):74-80, 2022. (HU, LE, GT)

Context: The ongoing pandemic has affected all the spheres of life and one of the severely affected avenues is the education of a child. The online education has seen an upward curve since the start of COVID-19 pandemic. Schools globally have adopted online class tutorials as the main method to impart education and directly increasing the screen time for a child. **Aim:** The aim of the present study was to evaluate the cytological effects of prolonged mobile phone usage on the buccal mucosa of children. **Settings and design:** Stratified sampling was used for the selection of subjects for the study. After a questionnaire regarding the usage of a mobile phone was distributed among the parents of children. Among them, 90 children were selected on the basis of pattern and frequency of mobile phone usage in the child. **Materials and methodology:** The children were divided into three groups based on the per day hours of viewing of mobile phone, i.e., Group 1: Usage of 1-2 h a day, Group 2: Usage of 3-6 h a day, and Group 3: Usage of >6 h a day. The time frame taken into consideration was 1 year after the pandemic started. This was specifically to understand the impact of the online education. Swab was obtained by using the conventional ice-cream stick method from the buccal mucosa. **Statistical analysis:** The samples were subjected to histological and microscopical analysis to observe for cytological changes. One-way ANOVA was used to determine the statistical significance if any. **Results:** The results obtained clearly showed that Group 3 (>6 h usage per day) showed the highest number of cellular and chromosomal aberrations which was significant. **Conclusion:** The results indicated that impact due to the prolonged screen time on the buccal mucosa is significant. A direct proportionality was seen between the apoptotic changes and chromosomal aberrations and the number of daily hour usage.

(NE) Stronati L, Testa A, Moquet J, Edwards A, Cordelli E, Villani P, Marino C, Fresegna AM, Appolloni M, Lloyd D. 935 MHz cellular phone radiation. An in vitro study of genotoxicity in human lymphocytes. Int J Radiat Biol 82:339-346, 2006. (VT, AE, GT, IX)

Purpose: The possibility of genotoxicity of radiofrequency radiation (RFR) applied alone or in combination with x-rays was investigated in vitro using several assays on human lymphocytes. The chosen specific absorption rate (SAR) values are near the upper limit of actual energy absorption in localized tissue when persons use some cellular telephones. The purpose of the combined exposures was to examine whether RFR might act epigenetically by reducing the fidelity of repair of DNA damage caused by a well-characterized and established mutagen.Methods: Blood specimens from 14 donors were exposed continuously for 24 h to a Global System for Mobile Communications (GSM) basic 935 MHz signal. The signal was applied at two SAR; 1 and 2 W/Kg, alone or combined with a 1-min exposure to 1.0 Gy of 250 kVp x-rays given immediately before or after the RFR. The assays employed were the alkaline comet technique to detect DNA strand breakage, metaphase analyses to detect unstable chromosomal aberrations and sister chromatid exchanges, micronuclei in cytokinesis-blocked binucleate lymphocytes and the nuclear division index to detect alterations in the speed of in vitro cell cycling.Results: By comparison with appropriate sham-exposed and control samples, no effect of RFR alone could be found for any of the assay endpoints. In addition RFR did not modify any measured effects of the x-radiation. Conclusions: This study has used several standard in vitro tests for chromosomal and DNA damage in Go human lymphocytes exposed in vitro to a combination of x-rays and RFR. It has comprehensively examined whether a 24-h continuous exposure to a 935 MHz GSM basic signal delivering SAR of 1 or 2 W/Kg is genotoxic per se or whether, it can influence the genotoxicity of the well-established clastogenic agent; x-radiation. Within the experimental parameters of the study in all instances no effect from the RFR signal was observed.

(NE) Su L, Wei X, Xu Z, Chen G. RF-EMF exposure at 1800 MHz did not elicit DNA damage or abnormal cellular behaviors in different neurogenic cells. Bioelectromagnetics 38(3)175-185, 2017.(VT, AE, GT)

Despite many years of studies, the debate on genotoxic effects of radiofrequency electromagnetic fields (RF-EMF) continues. To systematically evaluate genotoxicity of RF-EMF, this study examined effects of RF-EMF on DNA damage and cellular behavior in different neurogenic cells. Neurogenic A172, U251, and SH-SY5Y cells were intermittently (5 min on/10 min off) exposed to 1800 MHz RF-EMF at an average specific absorption rate (SAR) of 4.0 W/kg for 1, 6, or 24 h. DNA damage was evaluated by quantification of γH2AX foci, an early marker of DNA double-strand breaks. Cell cycle progression, cell proliferation, and cell viability were examined by flow cytometry, hemocytometer, and cell counting kit-8 assay, respectively. Results showed that exposure to RF-EMF at an SAR of 4.0 W/kg neither significantly induced γH2AX foci formation in A172, U251, or SH-SY5Y cells, nor resulted in abnormal cell cycle progression, cell proliferation, or cell viability. Furthermore, prolonged incubation of these cells for up to 48 h after exposure did not significantly affect cellular behavior. Our data suggest that

1800 MHz RF-EMF exposure at 4.0 W/kg is unlikely to elicit DNA damage or abnormal cellular behaviors in neurogenic cells.

(NE) Su L, Yimaer A, Xu Z, Chen G. Effects of 1800 MHz RF-EMF exposure on DNA damage and cellular functions in primary cultured neurogenic cells. Int J Radiat Biol. 94(3):295-305, 2018. (VT, LE, GT)

PURPOSE: To systematically evaluate the effects of 1800 MHz radiofrequency electromagnetic fields (RF-EMF) exposure on DNA damage and cellular functions in primary cultured neurogenic cells. MATERIALS AND METHODS: The primary cultured astrocytes, microglia and cortical neurons were exposed to RF-EMF at a SAR of 4.0 W/kg. The DNA damage was evaluated by γH2AX foci formation assay. The secretions of pro-inflammatory cytokines (TNF-α, IL-6 and IL-1β) in astrocytes and microglia, microglial phagocytic activity and neuronal development were examined by enzyme-linked immunosorbent assay, phagocytosis assay and immunofluorescent staining on microtubule-associated protein tau, microtubule-associated protein 2, postsynaptic density 95 and gephyrin, respectively. RESULTS: RF-EMF exposure did not significantly induce γH2AX foci formation in three primary cultured neurogenic cells. Furthermore, RF-EMF exposure did not significantly affect the secretion of cytokines in astrocytes and microglia, and the morphological indicators of dendrites or synapses of cortical neurons. However, the exposure significantly reduced the phagocytic activity of microglia and inhibited the axon branch length and branch number of cortical neurons. CONCLUSIONS: Our data demonstrated that exposure to RF-EMF did not elicit DNA damage but inhibited the phagocytic ability of microglia and the axon branch length and branch number of cortical neurons.

(E) Sueiro Benavides RA, Leiro-Vidal JM, Rodriguez JA, Ares-Pena FJ, López-Martín E. The HL-60 human promyelocytic cell line constitutes an effective in vitro model for evaluating toxicity, oxidative stress and necrosis/apoptosis after exposure to black carbon particles and 2.45 GHz radio frequency. Sci Total Environ 867:161475, 2023. (VT, AE, GE)

The cellular and molecular mechanisms by which atmospheric pollution from particulate matter and/or electromagnetic fields (EMFs) may prove harmful to human health have not been extensively researched. We analyzed whether the combined action of EMFs and black carbon (BC) particles induced cell damage and a pro-apoptotic response in the HL-60 promyelocytic cell line when exposed to 2.45 GHz radio frequency (RF) radiation in a gigahertz transverse electromagnetic (GTEM) chamber at sub-thermal specific absorption rate (SAR) levels. RF and BC induced moderately significant levels of cell damage in the first 8 or 24 h for all exposure times/doses and much greater damage after 48 h irradiation and the higher dose of BC. We observed a clear antiproliferative effect that increased with RF exposure time and BC dose. Oxidative stress or ROS production increased with time (24 or 48 h of radiation), BC dose and the combination of both. Significant differences between the proportion of damaged and healthy cells were observed in all groups. Both radiation and BC participated separately and jointly in triggering necrosis and apoptosis in a programmed way. Oxidative-antioxidant action activated mitochondrial anti-apoptotic BCL2a gene expression after 24 h irradiation and exposure to BC.

After irradiation of the cells for 48 h, expression of FASR cell death receptors was activated, precipitating the onset of pro-apoptotic phenomena and expression and intracellular activity of caspase-3 in the mitochondrial pathways, all of which can lead to cell death. Our results indicate that the interaction between BC and RF modifies the immune response in the human promyelocytic cell line and that these cells had two fates mediated by different pathways: necrosis and mitochondria-caspase dependent apoptosis. The findings may be important in regard to antimicrobial, inflammatory and autoimmune responses in humans.

(E) Sun C, Wei X, Fei Y, Su L, Zhao X, Chen G, Xu Z. Mobile phone signal exposure triggers a hormesis-like effect in Atm+/+ and Atm-/- mouse embryonic fibroblasts. Sci Rep. 6:37423, 2016. (VT, AE, GT) (compensatory effect)

Radiofrequency electromagnetic fields (RF-EMFs) have been classified by the International Agency for Research on Cancer as possible carcinogens to humans; however, this conclusion is based on limited epidemiological findings and lacks solid support from experimental studies. In particular, there are no consistent data regarding the genotoxicity of RF-EMFs. Ataxia telangiectasia mutated (ATM) is recognised as a chief guardian of genomic stability. To address the debate on whether RF-EMFs are genotoxic, we compared the effects of 1,800 MHz RF-EMF exposure on genomic DNA in mouse embryonic fibroblasts (MEFs) with proficient ($Atm^{+/+}$) or deficient ($Atm^{-/-}$) ATM. In $Atm^{+/+}$ MEFs, RF-EMF exposure for 1 h at an average special absorption rate of 4.0 W/kg induced significant DNA single-strand breaks (SSBs) and activated the SSB repair mechanism. This effect reduced the DNA damage to less than that of the background level after 36 hours of exposure. In the $Atm^{-/-}$ MEFs, the same RF-EMF exposure for 12 h induced both SSBs and double-strand breaks and activated the two repair processes, which also reduced the DNA damage to less than the control level after prolonged exposure. The observed phenomenon is similar to the hormesis of a toxic substance at a low dose. To the best of our knowledge, this study is the first to report a hormesis-like effect of an RF-EMF.

(E) Sun LX, Yao K, He JL, Lu DQ, Wang KJ, Li HW.[Effect of acute exposure to microwave from mobile phone on DNA damage and repair of cultured human lens epithelial cells in vitro.] Zhonghua Lao Dong Wei Sheng Zhi Ye Bing ZaZhi. 24:465-467, 2006a. [Article in Chinese] (VT, AE, GT)

OBJECTIVE: To investigate the DNA damage of human lens epithelial cells (LECs) caused by acute exposure to low-power 217 Hz modulated 1.8 GHz microwave radiation and DNA repair. METHODS: Cultured LECs were exposed to 217 Hz modulated 1.8 GHz microwave radiation at SAR (specific absorption rate) of 0, 1, 2, 3 and 4 W/kg for 2 hours in an sXc-1800 incubator and irradiate system. The DNA single strand breaks were detected with comet assay in sham-irradiated cells and irradiated cells incubated for varying periods: 0, 30, 60, 120 and 240 min after irradiation. Images of comets were digitized and analyzed using an Imagine-pro plus software, and the indexes used in this study were tail length (TL) and tail moment (TM). RESULTS: The difference in DNA-breaks between the exposure and sham exposure groups induced by 1 and 2 W/kg irradiation was not significant at every detect time ($P > 0.05$). As for the dosage of 3 and 4 W/kg there was difference in both groups immediately after irradiation (P

< 0.01). At the time of 30 min after irradiation the difference went on at both group (P < 0.01). However, the difference disappeared after one hour's incubation in 3 W/kg group (P > 0.05), and existed in 4 W/kg group. CONCLUSION: No or repairable DNA damage was observed after 2 hour irradiation of 1.8 GHz microwave on LECs when SAR </= 3 W/kg. The DNA damages caused by 4 W/kg irradiation were irreversible.

(E) Sun LX, Yao K, Jiang H, He JL, Lu DQ, Wang KJ, Li HW [DNA damage and repair induced by acute exposure of microwave from mobile phone on cultured human lens epithelial cells] Zhonghua Yan Ke Za Zhi. 42(12):1084-1088, 2006b. [Article in Chinese] (VT, AE, GT)

OBJECTIVE: To investigate the effects of acute exposure of low-power 217 Hz modulated 1. 8 GHz microwave radiation on the DNA damage of human lens epithelial cells (hLECs) and repair. METHODS: Cultured hLECs were exposed to 217 Hz modulated 1. 8 GHz microwave radiation at SAR (specific absorption rate) of 1. 0, 2. 0, 3. O0 and 4. 0 W/kg for 2 hours in an sXc-1800 incubator and irradiate system, the DNA single strand breaks were detected with comet assay (single-cell gel electrophoresis) in sham-irradiated cells and irradiated cells incubated for varying periods: 0, 30 and 60 minutes after irradiation. Images of comets were digitized and analyzed using an Imagine-pro plus software, and the indexes used in this study were tail length (TL) and tail moment (TM). BrdU was added into the medium with additional one hour incubation after radiation, the cell proliferation rate was determined using a BrdU-kit. RESULTS: The difference of DNA-breaks between the exposure and sham exposure groups induced by 1.0 and 2.0 W/kg irradiation were not significant in each time points (P > 0.05) ; there were significant difference in both groups at the exposure dose of 3. 0 and 4. 0 W/kg immediately and at the time of 30 minutes after irradiation (P <0. 01) ; if the radiation exposure time was beyond one hour no differences were be able to detected in 3.0 W/kg group (P > 0. 05) compared with control, but the evidence of significant DNA damage still existed in 4. 0 W/kg group at the same time point. Cell proliferation rate had no significant difference when the application of SAR was < or = 3. 0 W/kg (P >0. 05) , however the cell proliferation was decreased significantly at the dose of 4. 0 W/kg irradiation (P < 0. 01). CONCLUSIONS: No effective DNA damage was induced using comet assay after 2 hours irradiation of 1. 8 GHz microwave on hLECs at the dose SAR < or = 3.0 W/kg. 4.0 W/kg irradiation caused significantly DNA damage and inhibition of hLECs proliferation.

(E) Sun Y, Zong L, Gao Z, Zhu S, Tong J, Cao Y. Mitochondrial DNA damage and oxidative damage in HL-60 cellsexposed to 900 MHz radiofrequency fields. Mutat Res. 797: 7-14, 2017. (VT, LE, GT, OX, GE. LI)

HL-60 cells, derived from human promyelocytic leukemia, were exposed to continuous wave 900MHz radiofrequency fields (RF) at 120μW/cm^2 power intensity for 4h/day for 5 consecutive days to examine whether such exposure is capable damaging the mitochondrial DNA (mtDNA) mediated through the production of reactive oxygen species (ROS). In addition, the effect of RF exposure was examined on 8-hydroxy-2'-dexoyguanosine (8-OHdG) which is a biomarker for oxidative damage and on the mitochondrial synthesis of adenosine triphosphate (ATP) which is the energy required for cellular functions. The results indicated a significant increase in ROS and

significant decreases in mitochondrial transcription factor A, mtDNA polymerase gamma, mtDNA transcripts and mtDNA copy number in RF-exposed cells compared with those in sham-exposed control cells. In addition, there was a significant increase in 8-OHdG and a significant decrease in ATP in RF-exposed cells. The response in positive control cells exposed to gamma radiation (GR, which is also known to induce ROS) was similar to those in RF-exposed cells. Thus, the overall data indicated that RF exposure was capable of inducing mtDNA damage mediated through ROS pathway which also induced oxidative damage. Prior-treatment of RF- and GR-exposed the cells with melatonin, a well-known free radical scavenger, reversed the effects observed in RF-exposed cells.

(E) Sykes PJ, McCallum BD, Bangay MJ, Hooker AM, Morley AA. Effect of exposure to 900 MHz radiofrequency radiation on intrachromosomal recombination in pKZ1 mice. Radiat Res 156(5):495-502, 2001. (VO, LE, GT)

Radiofrequency (RF) radiation emitted from mobile phones is not considered to be directly genotoxic, but it may have downstream effects on cellular DNA. We studied the effect of 4 W/kg pulsed 900 MHz RF radiation on somatic intrachromosomal recombination in the spleen in the pKZ1 recombination mutagenesis model. Somatic intrachromosomal recombination inversion events were detected in spleen tissue of pKZ1 mice by histochemical staining for E. coli beta-galactosidase protein in cells in which the lacZ transgene has undergone an inversion event. pKZ1 mice were exposed daily for 30 min to plane-wave fields of 900 MHz with a pulse repetition frequency of 217 Hz and a pulse width of 0.6 ms for 1, 5 or 25 days. Three days after the last exposure, spleen sections were screened for DNA inversion events. There was no significant difference between the control and treated groups in the 1- and 5-day exposure groups, but there was a significant reduction in inversions below the spontaneous frequency in the 25-day exposure group. This observation suggests that exposure to RF radiation can lead to a perturbation in recombination frequency which may have implications for recombination repair of DNA. The biological significance of a reduction below the spontaneous frequency is not known. The number of mice in each treatment group in this study was small (n = 10 or n = 20). Therefore, repetition of this study with a larger number of animals is required to confirm these observations.

(NE) Takahashi S, Inaguma S, Cho Y-M, Imaida K, Wang J, Fujiwara O, Shirai T, Lack of mutation induction with exposure to 1.5 GHz electromagnetic near fields used for cellular phones in brains of big blue mice. Cancer Res 62:1956-1960, 2002. (VO, LE, GT)

The possible mutagenic potential of exposure to 1.5 GHz electromagneticnear field (EMF) was investigated using brain tissues of BigBlue mice (BBM). Male BBM were locally exposed to EMF in the head region at 2.0, 0.67, and 0 W/kg specific absorption ratefor 90 min/day, 5 days/week, for 4 weeks. No gliosis or degenerative lesions were histopathologically noted in brain tissues, and no obvious differences in Ki-67 labeling and apoptotic indices of glial cells were evident among the groups. There was no significant variation in the frequency of independent mutations of the lacItrans gene in the brains. G:C to A:T transitions at CpG sites constituted the most prevalent mutations in all groups and at all time points. Deletion mutations were slightly increased in both

the high and low EMF exposure groups as compared with the sham-exposed group, but the differences were not statistically significant. These findings suggest that exposure to 1.5 GHz EMF is not mutagenic to mouse brain cells and does not create any increased hazard with regard to brain tumor development.

(E)Tarsaei M, Peyrovan ZS, Mahdavi SM, Chahardehi AM, Vafaie R, Heidari M. Effects of 2.45 GHz Non-Ionizing Radiation on Anxiety-Like Behavior, Gene Expression, and Corticosterone Level in Male Rats. J Lasers Med Sci 13:e56, 2022. (VO, LE, GE, OX)

Introduction: The effects of short-term and long-term exposures to 2.45 GHz radiofrequency electromagnetic radiation (RF-EMR) on anxiety-like behavior, corticosterone level, and gene expression were investigated. The goal of this study was to explore the effect of electromagnetic fields of 2.45 GHz on clinical signs such as body weight and anxiety-like behavior, including the elevated plus maze test and open-field test, and also on messenger RNA (mRNA) expression of Bax (Bcl2-associated x) and Bcl-2 (B-cell lymphoma 2) genes on the cognitive memory functions in an animal model of rats. Methods: The animals were classified into eight groups, sham groups and exposed groups for short-term and long-term exposures to the same dose of RF-EMR for one hour daily. The Wi-Fi equipment in the sham control group was not turned on during the experiment. Both genes were further confirmed by reverse transcriptase-polymerase chain reaction (RT-PCR). The semi-quantitative PCR method of electromagnetic fields in the 2.45 GHz range impacted the expression of Bax and Bcl-2 genes in the rat's memory. Results: The present study exhibited that short-term radiation could decrease the percentage of entry into the open arm and the percentage of time spent, while there were no substantial impacts on the long-term radiation effect. Our data support the hypothesis that short-term exposure worked as a systemic stressor, raising plasma corticosterone and changing glucocorticoid receptor expression in the hippocampus. Conclusion: Additional research on this specific frequency and amount of radiation is required to discover strategies for protecting the nervous system from the detrimental effects of RF-EMR radiation.

(E) Tice RR, Hook GG, Donner M, McRee DI, Guy AW. Genotoxicity of radiofrequency signals. I. Investigation of DNA damage and micronuclei induction in cultured human blood cells. Bioelectromagnetics 23:113-126, 2002. (VT, AE, GT)

As part of a comprehensive investigation of the potential genotoxicity of radiofrequency (RF) signals emitted by cellular telephones, in vitro studies evaluated the induction of DNA and chromosomal damage in human blood leukocytes and lymphocytes, respectively. The signals were voice modulated 837 MHz produced by an analog signal generator or by a time division multiple access (TDMA) cellular telephone, 837 MHz generated by a code division multiple access (CDMA) cellular telephone (not voice modulated), and voice modulated 1909.8 MHz generated by a global system of mobile communication (GSM)-type personal communication systems (PCS) cellular telephone. DNA damage (strand breaks/alkali labile sites) was assessed in leukocytes using the alkaline (pH>13) single cell gel electrophoresis (SCG) assay. Chromosomal damage was evaluated in lymphocytes mitogenically stimulated to divide postexposure using

the cytochalasin B-binucleate cell micronucleus assay. Cells were exposed at 37±1°C, for 3 or 24 h at average specific absorption rates (SARs) of 1.0-10.0 W/kg. Exposure for either 3 or 24 h did not induce a significant increase in DNA damage in leukocytes, nor did exposure for 3 h induce a significant increase in micronucleated cells among lymphocytes. However, exposure to each of the four RF signal technologies for 24 h at an average SAR of 5.0 or 10.0 W/kg resulted in a significant and reproducible increase in the frequency of micronucleated lymphocytes. The magnitude of the response (approximately four fold) was independent of the technology, the presence or absence of voice modulation, and the frequency (837 vs. 1909.8 MHz). This research demonstrates that, under extended exposure conditions, RF signals at an average SAR of at least 5.0 W/kg are capable of inducing chromosomal damage in human lymphocytes.

(E) Tiwari R, Lakshmi NK, Surender V, Rajesh AD, Bhargava SC, Ahuja YR. Combinative exposure effect of radio frequency signals from CDMA mobile phones and aphidicolin on DNA integrity. Electromagn Biol Med 27:418-425, 2008. (VT, AE, GT, IX)

The aim of present study is to assess DNA integrity on the effect of exposure to a radio frequency (RF) signal from Code Division Multiple Access (CDMA) mobile phones. Whole blood samples from six healthy male individuals were exposed for RF signals from a CDMA mobile phone for 1 h. Alkaline comet assay was performed to assess the DNA damage. The combinative exposure effect of the RF signals and APC at two concentrations on DNA integrity was studied. DNA repair efficiency of the samples was also studied after 2 h of exposure. The RF signals and APC (0.2 microg/ml) alone or in synergism did not have any significant DNA damage as compared to sham exposed. However, univariate analysis showed that DNA damage was significantly different among combinative exposure of RF signals and APC at 0.2 microg/ml ($p < 0.05$) and at 2 microg/ml ($p < 0.02$). APC at 2 microg/ml concentration also showed significant damage levels ($p < 0.05$) when compared to sham exposed. DNA repair efficiency also varied in a significant way in combinative exposure sets ($p < 0.05$). From these results, it appears that the repair inhibitor APC enhances DNA breaks at 2 microg/ml concentration and that the damage is possibly repairable. Thus, it can be inferred that the in vitro exposure to RF signals induces reversible DNA damage in synergism with APC.

(E) Tkalec M, Malarić K, Pavlica M, Pevalek-Kozlina B, Vidaković-Cifrek Z. Effects of radiofrequency electromagnetic fields on seed germination and root meristematic cells of Allium Cepa L. Mutat Res 672(2):76-81, 2009 . (VO, AE, GT)

The effects of exposure to radiofrequency electromagnetic fields (RF-EMFs) on seed germination, primary root growth as well as mitotic activity and mitotic aberrations in root meristematic cells were examined in Allium cepa L. cv. Srebrnjak Majski. Seeds were exposed for 2h to EMFs of 400 and 900MHz at field strengths of 10, 23, 41 and 120Vm(-1). The effect of longer exposure time (4h) and field modulation was investigated at 23Vm(-1) as well. Germination rate and root length did not change significantly after exposure to radiofrequency fields under any of the treatment conditions. At 900MHz, exposures to EMFs of higher field strengths (41 and 120Vm(-1)) or to modulated fields showed a significant increase of the mitotic index compared with corresponding controls, while the percentage of mitotic abnormalities increased after all exposure treatments. On the other hand, at 400MHz the mitotic index increased only after exposure to modulated EMF. At this frequency, compared with the control

higher numbers of mitotic abnormalities were found after exposure to modulated EMF as well as after exposure to EMFs of higher strengths (41 and 120Vm(-1)). The types of aberration induced by the EMFs of both frequencies were quite similar, mainly consisting of lagging chromosomes, vagrants, disturbed anaphases and chromosome stickiness. Our results show that non-thermal exposure to the radiofrequency fields investigated here can induce mitotic aberrations in root meristematic cells of A. cepa. The observed effects were markedly dependent on the field frequencies applied as well as on field strength and modulation. Our findings also indicate that mitotic effects of RF-EMF could be due to impairment of the mitotic spindle.

(E) Tkalec M, Stambuk A, Srut M, Malarić K, Klobučar GI. Oxidative and genotoxic effects of 900 MHz electromagnetic fields in the earthworm Eisenia fetida. Ecotoxicol Environ Saf. 90:7-12, 2013. (VO, AE, GT, OX, WS)

Accumulating evidence suggests that exposure to radiofrequency electromagnetic field (RF-EMF) can have various biological effects. In this study the oxidative and genotoxic effects were investigated in earthworms Eisenia fetida exposed in vivo to RF-EMF at the mobile phone frequency (900MHz). Earthworms were exposed to the homogeneous RF-EMF at field levels of 10, 23, 41 and 120Vm(-1) for a period of 2h using a Gigahertz Transversal Electromagnetic (GTEM) cell. At the field level of 23Vm(-1) the effect of longer exposure (4h) and field modulation (80% AM 1kHz sinusoidal) was investigated as well. All exposure treatments induced significant genotoxic effect in earthworms coelomocytes detected by the Comet assay, demonstrating DNA damaging capacity of 900MHz electromagnetic radiation. Field modulation additionally increased the genotoxic effect. Moreover, our results indicated the induction of antioxidant stress response in terms of enhanced catalase and glutathione reductase activity as a result of the RF-EMF exposure, and demonstrated the generation of lipid and protein oxidative damage. Antioxidant responses and the potential of RF-EMF to induce damage to lipids, proteins and DNA differed depending on the field level applied, modulation of the field and duration of E. fetida exposure to 900MHz electromagnetic radiation. Nature of detected DNA lesions and oxidative stress as the mechanism of action for the induction of DNA damage are discussed.

(E) Tohidi F, Sadr-Nabavi A, Haghir H, Fardid R, Rafatpanah H, Azimian H, Bahreyni-Toossi M. Long-term exposure to electromagnetic radiation from mobile phones can cause considerable changes in the balance of Bax/Bcl2 mRNA expression in the hippocampus of mice. Electromagn Biol Med 40(1):131-137, 2021. (VO, LE, GE)

The aim of the present study was the investigation of the effects of mobile phones at different daily exposure times on the hippocampal expression of two apoptotic genes. Forty-eight male BALB/c mice were randomly divided into six groups with 8 animals in each group. Four experimental groups were respectively exposed to electromagnetic waves for 0.5, 1, 2 and 4 hours twice a day for 30 consecutive days. One experimental group was radiated for 4 hours once a day, while the control group did not receive any radiation during the experiment. The expression of both *Bax* and *Bcl2* mRNAs was upregulated in the mice exposed for one and two hours. Whilst the highest expressions were observed in the two-hours radiation in the exposed group, the expression of both studied genes was downregulated in animals with longer exposure to radiation in a duration-dependent manner. The highest ratio of *Bax*/*Bcl2* expression was observed in the mice that received radiation for four hours twice a day. These results revealed that mobile phone radiation can cause considerable changes in the balance of *Bax/Bcl2* mRNA expression in laboratory mice hippocampus.

(NE) Tomruk A, Guler G, Dincel AS. The influence of 1800 MHz GSM-like signals on hepatic oxidative DNA and lipid damage in nonpregnant, pregnant, and newly born rabbits. Cell Biochem Biophys 56:39-47, 2010. (VO, LE, GT, OX, DE)

The aim of our study is to evaluate the possible biological effects of whole-body 1800 MHz GSM-like radiofrequency (RF) radiation exposure on liver oxidative DNA damage and lipid peroxidation levels in nonpregnant, pregnant New Zealand White rabbits, and in their newly borns. Eighteen nonpregnant and pregnant rabbits were used and randomly divided into four groups which were composed of nine rabbits: (i) Group I (nonpregnant control), (ii) Group II (nonpregnant-RF exposed), (iii) Group III (pregnant control), (iv) Group IV (pregnant-RF exposed). Newborns of the pregnant rabbits were also divided into two groups: (v) Group V (newborns of Group III) and (vi) Group VI (newborns of Group III). 1800 MHz GSM-like RF radiation whole-body exposure (15 min/day for a week) was applied to Group II and Group IV. No significant differences were found in liver 8 OHdG/10 dG levels of exposure groups (Group II and Group IV) compared to controls (Group I and Group III). However, in Group II and Group IV malondialdehyde (MDA) and ferrous oxidation in xylenol orange (FOX) levels were increased compared to Group I ($P < 0.05$, Mann-Whitney). No significant differences were found in liver tissue of 8 OHdG/10 dG and MDA levels between Group VI and Group V ($P > 0.05$, Mann-Whitney) while liver FOX levels were found significantly increased in Group VI with respect to Group V ($P < 0.05$, Mann-Whitney). Consequently, the whole-body 1800 MHz GSM-like RF radiation exposure may lead to oxidative destruction as being indicators of subsequent reactions that occur to form oxygen toxicity in tissues.

(E) Tran NT, Jokic L, Keller J, Geier JU, Kaldenhoff R. Impacts of Radio-Frequency Electromagnetic Field (RF-EMF) on Lettuce (*Lactuca sativa*)—Evidence for RF-EMF Interference with Plant Stress Responses. *Plants*. 12(5):1082, 2023. (VO, LE, GE)

The increased use of wireless technology causes a significant exposure increase for all living organisms to radio frequency electromagnetic fields (RF-EMF). This comprises bacteria, animals, and also plants. Unfortunately, our understanding of how RF-EMF influences plants and plant physiology remains inadequate. In this study, we examined the effects of RF-EMF radiation on lettuce plants (Lactuca sativa) in both indoor and outdoor environments using the frequency ranges of 1890–1900 MHz (DECT) at 2.4 GHz and 5 GHz (Wi-Fi). Under greenhouse conditions, RF-EMF exposure had only a minor impact on fast chlorophyll fluorescence kinetics and no effect on plant flowering time. In contrast, lettuce plants exposed to RF-EMF in the field showed a significant and systemic decrease in photosynthetic efficiency and accelerated flowering time compared to the control groups. Gene expression analysis revealed significant down-regulation of two stress-related genes in RF-EMF-exposed plants: violaxanthin de-epoxidase (VDE) and zeaxanthin epoxidase (ZEP). RF-EMF-exposed plants had lower Photosystem II's maximal photochemical quantum yield (F_V/F_M) and non-photochemical quenching (NPQ) than control plants under light stress conditions. In summary, our results imply that RF-EMF might interfere with plant stress responses and reduced plant stress tolerance.

(E) Trivino Pardo JC, Grimaldi S, Taranta M, Naldi I, Cinti C. Microwave electromagnetic field regulates gene expression in T-lymphoblastoid leukemia CCRF-CEM cell line exposed to 900 MHz. Electromagn Biol Med. 31(1):1-18, 2012. (VT, AE, GE)

Electric, magnetic, and electromagnetic fields are ubiquitous in our society, and concerns have been expressed regarding possible adverse effects of these exposures. Research on Extremely Low-Frequency (ELF) magnetic fields has been performed for more than two decades, and the methodology and quality of studies have improved over time. Studies have consistently shown increased risk for childhood leukemia associated with ELF magnetic fields. There are still inadequate data for other outcomes. More recently, focus has shifted toward Radio Frequencies (RF) exposures from mobile telephony. There are no persuasive data suggesting a health risk, but this research field is still immature with regard to the quantity and quality of available data. This technology is constantly changing and there is a need for continued research on this issue. To investigate whether exposure to high-frequency electromagnetic fields (EMF) could induce adverse health effects, we cultured acute T-lymphoblastoid leukemia cells (CCRF-CEM) in the presence of 900 MHz MW-EMF generated by a transverse electromagnetic (TEM) cell at short and long exposure times. We evaluated the effect of high-frequency EMF on gene expression and we identified functional pathways influenced by 900 MHz MW-EMF exposure.

(E) Trosic I. Multinucleated giant cell appearance after whole body microwave irradiation of rats. Int J Hyg Environ Health. 204(2-3):133-138, 2001. (VO, LE, GT)

Multinucleated giant cells are common for some chronic inflammatory processes in the lung. These cells are formed by fusion of macrophages, but how the process relates to the kinetics of alveolar macrophage generation is not clear. This study investigated the influence of 2450 MHz microwave irradiation on alveolar macrophage kinetics and formation of multinucleated giant cells after whole body irradiation of rats. The range of electromagnetic radiation was selected as 2450 MHz microwaves at a power density of 5-15 mW/cm2. A group of experimental animals was divided in four subgroups that received 2, 8, 13 and 22 irradiation treatments of two hours each. The animals were killed on experimental days 1, 8, 16, and 30. Free lung cell population was obtained by bronchoalveolar lavage. Cell response to the selected irradiation level was followed quantitatively, qualitatively and morphologically using standard laboratory methods. Total cell number retrieved by lavage slightly decreased in treated animals showing time- and dose-dependence. Cell viability did not significantly change in the irradiated animal group (G2) as compared with the control group (G1). Multinucleated cells significantly increased ($p < 0.01$) in treated animals. The elevation of the number of nuclei per cell was time- and dose-dependent. Macrophages with two nucleoli were more common in animals treated twice or eight times. Polynucleation, that is three and more nucleoli in a single cell, was frequently observed after 13 or 22 treatments. Binucleation and multinucleation of alveolar macrophages were sensitive time- and dose-dependent morphological indicators of pulmonary stress.

(E) Trosic I, Busljeta I. Frequency of micronucleated erythrocytes in rat bone marrow exposure to 2.45 GHz radiation. Physica Scripta T118: 168-170, 2005. (VO, LE, GT)

Wistar rats were exposed to 2.45 GHz continuous, radiofrequency microwave (RF/MW) field 2 hours daily, 7 days weekly, at power density 5–10mW/cm2. Four subgroups were created in order to be irradiated 4, 16, 30 and 60 hours. Sham exposed controls were included in the study. Animals were euthanized on the final irradiation day of each treated subgroup. Bone marrow smears were examined to determine the extent of genotoxicity after the particular

treatment time. Mann- Whitney testwas used for statistical evaluation of data. In comparison to the sham exposed subgroups, the findings of polychromatic erythrocytes revealed significant differences for the 8th and 15th experimental day. Bone marrow erythrocyte maturation and/or proliferation initiated by subthermogenic RF/MW irradiation showed temporary disturbance. Thereafter, the frequency of micronucleated bone marrow red cells was significantly increased after 15 irradiation treatments. Comparison of micronucleus frequency data obtained after 2, 8 and 30 irradiation treatments did not reveal statistically significant differences between sham and treated subgroups. Under the applied experimental conditions, RF/MWirradiation initiates transitory cytogenetic effect manifested with micronucleus formation in erythropoietic cells.

(E) Trosic I, Busljeta I. Erythropoietic dynamic equilibrium in rats maintained after microwave irradiation. Exp Toxicol Pathol. 57(3):247-251, 2006. (VO, LE, GT)

The aim of study was to define influence of radiofrequency microwave (RF/MW) radiation on erythropoiesis in rats. The kinetics of polychromatic erythrocytes (PCEs) and micronucleated (MN) PCEs in the bone marrow (BM) and peripheral blood (PB) of rats during the intermittent subchronic experiment was followed. Rats were exposed 2h/day, 7 days/week to RF/MW of 2.45 GHz and whole-body specific absorption rate (SAR) of 1.25+/-0.36W/kg. Control animals were included in the study. Each exposed and control group was killed on the final day of irradiation. Acridine-orange stained BM and blood smears were examined by fluorescence microscope. PCEs were obtained by inspection of 2000 BM and 1000 PB erythrocytes/slides. BMMNs and PBMNs frequency was obtained by observation of 1000 PCEs/slides. BMPCEs were increased on day 8 and 15, and PBPCEs were elevated on days 2 and 8 ($p<0.05$). The BMMN frequency was increased on experimental day 15, and MNPCEs in the PB was increased on day 8 ($p<0.05$). Findings of BM and PBPCEs or MNPCEs declined nearly to the control values until the end of the experiment. Such findings are considered to be indicators of radiation effects on BM erythropoiesis consequently reflected in the PB. Rehabilitated dynamic haemopoietc equilibrium in rats by the end of experiment indicates possibility of activation adaptation process in rats to the selected experimental conditions of subchronic RF/MW exposure.

(E) Trosic I, Busljeta I, Kasuba V, Rozgaj R. Micronucleus induction after whole-body microwave irradiation of rats. Mutat Res 521(1-2):73-79, 2002. (VO, LE, GT)

Adult male Wistar rats were exposed for 2h a day, 7 days a week for up to 30 days to continuous 2450 MHz radiofrequency microwave (rf/MW) radiation at a power density of 5-10 mW/cm(2). Sham-exposed rats were used as controls. After ether anesthesia, experimental animals were euthanized on the final irradiation day for each treated group. Peripheral blood smears were examined for the extent of genotoxicity, as indicated by the presence of micronuclei in polychromatic erythrocytes (PCEs). The results for the time-course of PCEs indicated significant differences ($P<0.05$) for the 2nd, the 8th and the 15th day between control and treated subgroups of animals. Increased influx of immature erythrocytes into the peripheral circulation at the beginning of the experiment revealed that the proliferation and maturation of nucleated erythropoietic cells were affected by exposure to the 2450MHz radiofrequency radiation. Such findings are indicators of radiation effects on bone-marrow

erythropoiesis and their subsequent effects in circulating red cells. The incidence of micronuclei/1000 PCEs in peripheral blood was significantly increased (P<0.05) in the subgroup exposed to rf/MW radiation after eight irradiation treatments of 2h each in comparison with the sham-exposed control group. It is likely that an adaptive mechanism, both in erythrocytopoiesis and genotoxicity appeared in the rat experimental model during the subchronic irradiation treatment.

(E) Trosic I, Busljeta I, Modlic B. Investigation of the genotoxic effect of microwave irradiation in rat bone marrow cells: in vivo exposure. Mutagenesis. 19(5):361-364, 2004. (VO, LE, GT)

An in vivo mammalian cytogenetic test (the erythrocyte micronucleus assay) was used to investigate the extent of genetic damage in bone marrow red cells of rats exposed to radiofrequency/microwave (RF/MW) radiation. Wistar rats (n = 40) were exposed to a 2.45 GHz continuous RF/MW field for 2 h daily, 7 days a week, at a power density of 5-10 mW/cm(2). The whole body average specific absorption rate (SARs) was calculated to be 1.25 +/- 0.36 (SE) W/kg. Four subgroups were irradiated for 4, 16, 30 and 60 h. Sham-exposed controls (n = 24) were included in the study. The animals of each treated subgroup were killed on the final day of irradiation. Bone marrow smears were examined to determine the extent of genotoxicity after particular treatment times. The results were statistically evaluated using non-parametric Mann-Whitney and Kruskal-Wallis tests. In comparison with the sham-exposed subgroups, the findings of polychromatic erythrocytes (PCE) revealed significant differences (P < 0.05) for experimental days 8 and 15. The frequency of micronucleated PCEs was also significantly increased on experimental day 15 (P < 0.05). Pair-wise comparison of data obtained after 2, 8 and 30 irradiation treatments did not reveal statistically significant differences between sham-exposed and treated subgroups. Under the applied experimental conditions the findings revealed a transient effect on proliferation and maturation of erythropoietc cells in the rat bone marrow and the sporadic appearance of micronucleated immature bone marrow red cells.

(E) Trosić I, Pavicić I, Milković-Kraus S, Mladinić M, Zeljezić D. Effect of electromagnetic radiofrequency radiation on the rats' brain, liver and kidney cells measured by comet assay. Coll Antropol 35:1259-1264, 2011. (VO, LE, GT)

The goal of study was to evaluate DNA damage in rat's renal, liver and brain cells after in vivo exposure to radiofrequency/microwave (Rf/Mw) radiation of cellular phone frequencies range. To determine DNA damage, a single cell gel electrophoresis/comet assay was used. Wistar rats (male, 12 week old, approximate body weight 350 g) (N = 9) were exposed to the carrier frequency of 915 MHz with Global System Mobile signal modulation (GSM), power density of 2.4 W/m2, whole body average specific absorption rate SAR of 0.6 W/kg. The animals were irradiated for one hour/day, seven days/week during two weeks period. The exposure set-up was Gigahertz Transversal Electromagnetic Mode Cell (GTEM--cell). Sham irradiated controls (N = 9) were apart of the study. The body temperature was measured before and after exposure. There were no differences in temperature in between control and treated animals. Comet assay parameters such as the tail length and tail intensity were evaluated. In comparison with tail length in controls (13.5 +/- 0.7 microm), the tail was slightly elongated in brain cells of irradiated

animals (14.0 +/- 0.3 microm). The tail length obtained for liver (14.5 +/- 0.3 microm) and kidney (13.9 +/- 0.5 microm) homogenates notably differs in comparison with matched sham controls (13.6 +/- 0.3 microm) and (12.9 +/- 0.9 microm). Differences in tail intensity between control and exposed animals were not significant. The results of this study suggest that, under the experimental conditions applied, repeated 915 MHz irradiation could be a cause of DNA breaks in renal and liver cells, but not affect the cell genome at the higher extent compared to the basal damage.

(E) Tsybulin O, Sidorik E, Brieieva O, Buchynska L, Kyrylenko S, Henshel D, Yakymenko I. GSM 900 MHz cellular phone radiation can either stimulate or depress early embryogenesis in Japanese quails depending on the duration of exposure. Int J Radiat Biol. 89(9):756-763, 2013. (VO, LE, LI, GT)

PURPOSE: Our study was designed to assess the effects of low intensity radiation of a GSM (Global System for Mobile communication) 900 MHz cellular phone on early embryogenesis in dependence on the duration of exposure. **MATERIALS AND METHODS:** Embryos of Japanese Quails were exposed in ovo to GSM 900 MHz cellular phone radiation during initial 38 h of brooding or alternatively during 158 h (120 h before brooding plus initial 38 h of brooding) discontinuously with 48 sec ON (average power density 0.25 μW/cm(2), specific absorption rate 3 μW/kg) followed by 12 sec OFF intervals. A number of differentiated somites were assessed microscopically. Possible DNA damage evoked by irradiation was assessed by an alkaline comet assay. **RESULTS:** Exposure to radiation from a GSM 900 MHz cellular phone led to a significantly altered number of differentiated somites. In embryos irradiated during 38 h the number of differentiated somites increased ($p < 0.001$), while in embryos irradiated during 158 h this number decreased ($p < 0.05$). The lower duration of exposure led to a significant ($p < 0.001$) decrease in a level of DNA strand breaks in cells of 38-h embryos, while the higher duration of exposure resulted in a significant ($p < 0.001$) increase in DNA damage as compared to the control. **CONCLUSION:** Effects of GSM 900 MHz cellular phone radiation on early embryogenesis can be either stimulating or deleterious depending on the duration of exposure.

(E) Usikalu MR, Obembe OO, Akinyemi ML, Zhu J. Short-duration exposure to 2.45 GHz microwave radiation induces DNA damage in Sprague Dawley rat's reproductive systems. African J Biotechnol. 12: 115-122, 2013.(VO, AE, GT, RP)

The genotoxic effects of 2.45 GHz microwave (MW) radiation on the testis and ovary of Sprague Dawley rats was investigated. The animals were exposed to varying levels of specific absorption rate (SAR) of 0 (control), 0.48, 0.95, 1.43, 1.91, 2.39, 2.90, 3.40, 3.80 and 4.30 Wkg^{-1}, for 10 min. The induction of DNA damages was assessed using DNA direct amplification of length polymorphisms (DALP) and validated with single cell gel electrophoresis (SCGE) comet assay for same cells at SAR 2.39 Wkg^{-1}. Potential damage at the organ level was assessed by histopathological study. The results show significant differences in the Olive moment and % DNA in the blood of the exposed animals when compared with the control ($p < 0.05$). Hyperchromasia was observed in the ovary of the animals exposed to MW radiation. Also, there was reduction in the number of germ cells and cell disorganization in the testis of exposed group with increasing SARs. These results suggest that MW radiation has the potential to affect both male and female fertility adversely.

(E)Üstündağ ÜV, Özen MS, Ünal İ, Ateş PS, Alturfan AA, Akalın M, Sancak E, Emekli-Alturfan E. Oxidative stress and apoptosis in electromagnetic waves exposed Zebrafish embryos and protective effects of conductive nonwoven fabric. Cell Mol Biol (Noisy-le-grand) 66(1):70-75, 2020. (VO, LE, GE, DE)

The amount of technological products including television, radio transmitters, and mobile phone that have entered our daily life has increased in recent years. But these devices may cause adverse effects on human health. Electromagnetic shielding fabrics may limit and inhibit electromagnetic waves. Aim of our study was to evaluate electromagnetic wave blocking performance of nonwoven textile surfaces on zebrafish embryos that were exposed to electromagnetic waves at specific frequencies. Oxidant-antioxidant system parameters were evaluated spectrophotometrically. The expressions of tp53 and casp3a were evaluated by RT-PCR. Results showed that electromagnetic shielding fabrics produced as conductive nonwoven textile surfaces improved oxidant-antioxidant status and tp53 expression that were impaired in electromagnetic waves exposed zebrafish embryos. Also, electromagnetic shielding fabrics decreased casp3a expression responsible for the execution phase of apoptosis that increased in electromagnetic waves exposed zebrafish embryos.

(E) Vafaei H, Kavari G, Izadi HR, Zahra Zare Dorahi ZZ, Dianatpour M, Daneshparvar A, Jamhiri I. Wi-Fi (2.4 GHz) affects anti-oxidant capacity, DNA repair genes expression and, apoptosis in pregnant mouse placenta. Iran J Basic Med Sci 23(6):833-840, 2020. (VO, LE, GE)

Objective(s): The placenta provides nutrients and oxygen to embryo and removes waste products from embryo's blood. As far as we know, the effects of exposure to Wi-Fi (2.4 GHz) signals on placenta have not been evaluated. Hence, we examined the effect of prenatal exposure to Wi-Fi signals on anti-oxidant capacity, expressions of CDKNA1, and GADD45a as well as apoptosis in placenta and pregnancy outcome. Materials and Methods: Pregnant mice were exposed to Wi-Fi signal (2.4 GHz) for 2 and 4 hr. Placenta tissues were examined to measure the MDA and SOD levels. To measure SOD, CDKNA1, GADD45a, Bax, and Bcl-2 expressions were compared by real-time PCR analysis. TUNEL assay was used to assess apoptosis in placenta tissues. The results were analyzed by one-way analysis of variance (ANOVA) using Prism version 6.0 software. Results: MDA and SOD levels had significantly increased in exposed Wi-Fi signal groups (P-value< 0.05). Also, quantitative PCR experiment showed that SOD mRNA expression significantly increased in Wi-Fi signal groups. The data showed that CDKN1A and GADD45a genes were increased in Wi-Fi groups (P-value<0.05). The quantitative PCR and the TUNEL assay showed that apoptosis increased in Wi-Fi groups (P-value<0.05). Conclusion: Our results provide evidence that Wi-Fi signals increase lipid peroxidation, SOD activity (oxidative stres), apoptosis and CDKN1A and GADD45a overexpression in mice placenta tissue. However, further experimental studies are warranted to investigate other genes and aspects of pregnancy to determine the role of Wi-Fi radiation on fertility and pregnancy.

(NE) Valbonesi P, Franzellitti S, Piano A, Contin A, Biondi C, Fabbri E. Evaluation of HSP70 expression and DNA damage in cells of a human trophoblast cell line exposed to 1.8

GHz amplitude-modulated radiofrequency fields. Radiat Res 169:270-279, 2008. (VT, AE, GT, GE)

The aim of this study was to determine whether high-frequency electromagnetic fields (EMFs) could induce cellular effects. The human trophoblast cell line HTR-8/SVneo was used as a model to evaluate the expression of proteins (HSP70 and HSC70) and genes (HSP70A, B, C and HSC70) of the HSP70 family and the primary DNA damage response after nonthermal exposure to pulse-modulated 1817 MHz sinusoidal waves (GSM-217 Hz; 1 h; SAR of 2 W/kg). HSP70 expression was significantly enhanced by heat, which was applied as the prototypical stimulus. The HSP70A, B and C transcripts were differentially expressed under basal conditions, and they were all significantly induced above basal levels by thermal stress. Conversely, HSC70 protein and gene expression was not influenced by heat. Exposing HTR-8/SVneo cells to high-frequency EMFs did not change either HSP70 or HSC70 protein or gene expression. A significant increase in DNA strand breaks was caused by exposure to HO, which was used as a positive stimulus; however, no effect was observed after exposure of cells to high-frequency EMFs. Overall, no evidence was found that a 1-h exposure to GSM-217 Hz induced a HSP70-mediated stress response or primary DNA damage in HTR-8/SVneo cells. Nevertheless, further investigations on trophoblast cell responses after exposure to GSM signals of different types and durations are needed.

(E) Valbonesi P, Franzellitti S, Bersani F, Contin A, Fabbri E. Effects of the exposure to intermittent 1.8 GHz radio frequency electromagnetic fields on HSP70 expression and MAPK signaling pathways in PC12 cells. Int J Radiat Biol. 90(5):382-391, 2014. (VT, AE, GE, WS)

Purpose: We previously reported effects on heat shock protein 70 (HSP70) mRNA expression, a cytoprotective protein induced under stressful condition, in human trophoblast cells exposed to amplitude-modulated Global System for Mobile Communication (GSM) signals. In the present work the same experimental conditions were applied to the rat PC12 cells, in order to assess the stress responses mediated by HSP70 and by the Mitogen Activated Protein Kinases (MAPK) in neuronal-like cells, an interesting model to study possible effects of mobile phone frequencies exposure. Materials and methods: HSP70 gene expression level was evaluated by reverse transcriptase polymerase chain reaction, HSP70 protein expression and MAPK phosphorylation were assessed by Western blotting. PC12 cells were exposed for 4, 16 or 24 h to 1.8 GHz continuous wave signal (CW, carrier frequency without modulation) or to two different GSM modulation schemes, GSM-217Hz and GSM-Talk (which generates temporal changes between two different GSM signals, active during talking or listening phases respectively, thus simulating a typical conversation). Specific adsorption rate (SAR) was 2 W/kg. Results: After PC12 cells exposure to the GSM-217Hz signal for 16 or 24 h, HSP70 transcription significantly increased, whereas no effect was observed in cells exposed to the CW or GSM-Talk signals. HSP70 protein expression and three different MAPK signaling pathways were not affected by the exposure to any of the three different 1.8 GHz signals. Conclusion: The positive effect on HSP70 mRNA expression, observed only in cells exposed to the GSM-217Hz signal, is a repeatable response previously reported in human trophoblast cells and now confirmed in PC12 cells. Further investigations towards a possible role of 1.8 GHz signal modulation are therefore advisable.

(NE) Valbonesi P, Franzellitti S, Bersani F, Contin A, Fabbri E. Activity and expression of acetylcholinesterase in PC12 cells exposed to intermittent 1.8 GHz 217-GSM mobile phone signal. Int J Radiat Biol. 92(1):1-10, 2016. (VT, AE, GE)

Purpose: Due to its role in learning, memory and in many neurodegenerative diseases, acetylcholinesterase (AChE) represents an interesting endpoint to assess possible targets of exposure to radiofrequency electromagnetic fields (RF-EMF) generated by mobile phones. We investigated possible alterations of enzymatic activity, gene and protein expression of AChE in neuronal-like cells exposed to a 1.8 GHz Global System for Mobile Communication (GSM) modulated signal (217-GSM). **Materials and methods:** Rat PC12 cells were exposed for 24 h to 1.8 GHz 217-GSM signal. Specific adsorption rate (SAR) was 2 W/kg. AChE enzyme activity was assessed spectrophotometrically by Ellman's method, mRNA expression level was evaluated by real time polymerase chain reaction, and protein expression was assessed by Western blotting. **Results:** AChE enzymatic activity increased of 1.4-fold in PC12 cells exposed to 217-GSM signal for 24 h, whilst AChE transcriptional or translational pathways were not affected. **Conclusion:** Our results provide the first evidence of effects on AChE activity after in vitro exposure of mammalian cells to the RF-EMF generated by GSM mobile phones, at the SAR value 2 W/kg. The obtained evidence promotes further investigations on AChE as a possible target of RF-EMF and confirm the ability of 1.8 GHz 217-GSM signal to induce biological effects in different mammalian cells.

(E)Vanishree, M., Manvikar V, Rudraraju A, Reddy KMP, Kumar NHP, and Quadri SJM. Significance of micronuclei in buccal smears of mobile phone users: A comparative study. J. Oral Maxillofacial Pathol. 22:448–452, 2018. (HU, LE, GT)

Aims and objectives: The present study was designed to evaluate the frequency of micronuclei (MN) in the buccal exfoliated cells of mobile phone users. In addition, comparison of MN frequency between high and low mobile phone users was also done. **Materials and methods:** A total of 30 male and 30 female participants between the age group of 20-28 years were selected from the Outpatient Department of Navodaya Dental College and Hospital, Raichur, Karnataka. The participants were divided into two groups: Group A - low mobile phone users and Group B - high mobile phone users. Cell sampling and preparation was done on the slide. All the slides were observed for a total of 1000 cells for the presence and number of MN in each cell. **Results:** There was a significant increase in the mean MN count in Group B in comparison to the Group A. There was highly significant difference in the mean MN count of participants using (code division multiple access) CDMA than (global system for mobiles) GSM mobile phones. The MN mean count was found to be significantly increased in nonheadphone users in comparison to headphone users. In Group B, the MN count on the side of mobile phone use was found to be statistically significantly elevated in comparison to the opposite side. **Conclusion:** Mobile phone radiation even in the permissible range when used for longer duration can cause significant genotoxicity. The genotoxicity accentuates when mobile phones are frequently used on the same side which may be due to more amount of radiation and increase in the temperature. Headphone usage reduces the genotoxicity of mobile phone radiation to some extent.

(E) Veerachari SB, Vasan SS. Mobile Phone Electromagnetic Waves and Its Effect on Human Ejaculated Semen: An in vitro Study. Int J Infertility Fetal Med 3(1):15-21, 2012. (VT, AE, GT)

Mobile phones usage has seen an exponential growth recently. With this increasing demand, the amount of electromagnetic radiation (EMR) exposed is also increasing. Hence, we studied the effect of these radiations on ejaculated human semen and speculate the contribution of these harmful radiations in male infertility. Samples exposed to EMR showed a significant decrease in sperm motility and viability, increase in reactive oxygen species (ROS) and DNA fragmentation index (DFI) compared to unexposed group. We concluded that mobile phones emit electromagnetic waves which lead to oxidative stress in human semen and also cause changes in DNA fragmentation. We extrapolate these findings to speculate that these radiations may negatively affect spermatozoa and impair male fertility.

(E)Verma M, Dutta SK. Microwave induced alteration in the neuron specific enolase gene expression. Cancer Biochem Biophys. 13(4):239-244, 1993. (VT, AE, GE)

Exposure of pNGE7, a recombinant clone containing the coding and regulatory sequences for the expression of neuron specific enolase gene, cells to electromagnetic radiations (915 MHz, 16 Hz AM, SAR 0.05 mW/kg) resulted in the elevation of neuron specific enolase (NSE), a diagnostic marker for neuron and lung cancer. Using ion-exchange chromatography we separated the neuron specific enolase activity from the non-neuronal enolase (NNE) activity and observed an alteration in the expression of neuron specific enolase and non-neuronal enolase. The clinical applications of the present studies have been discussed

(NE) Verschaeve L, Heikkinen P, Verheyen G, Van Gorp U, Boonen F, Vander Plaetse F, Maes A, Kumlin T, Maki-Paakkanen J, Puranen L, Juutilainen J. Investigation of co-genotoxic effects of radiofrequency electromagnetic fields in vivo. Radiat Res 165:598-607, 2006. (VO, LE, GT, IX)

We investigated the possible combined genotoxic effects of radiofrequency (RF) electromagnetic fields (900 MHz, amplitude modulated at 217 Hz, mobile phone signal) with the drinking water mutagen and carcinogen 3-chloro-4-(dichloromethyl)-5-hydroxy-2(5H)-furanone (MX). Female rats were exposed to RF fields for a period of 2 years for 2 h per day, 5 days per week at average whole-body specific absorption rates of 0.3 or 0.9 W/kg. MX was given in the drinking water at a concentration of 19 mug/ml. Blood samples were taken at 3, 6 and 24 months of exposure and brain and liver samples were taken at the end of the study (24 months). DNA damage was assessed in all samples using the alkaline comet assay, and micronuclei were determined in erythrocytes. We did not find significant genotoxic activity of MX in blood and liver cells. However, MX induced DNA damage in rat brain. Co-exposures to MX and RF radiation did not significantly increase the response of blood, liver and brain cells compared to MX exposure only. In conclusion, this 2-year animal study involving long-term exposures to RF radiation and MX did not provide any evidence for enhanced genotoxicity in rats exposed to RF radiation.

Verschaeve L. Genetic damage in subjects exposed to radiofrequency radiation. Mutat Res. 681(2-3):259-270 (2009). (Review)

Despite many research efforts and public debate there is still great concern about the possible adverse effects of radiofrequency (RF) radiation on human health. This is especially due to the

enormous increase of wireless mobile telephones and other telecommunication devices throughout the world. The possible genetic effects of mobile phone radiation and other sources of radiofrequencies constitute one of the major points of concern. In the past several review papers were published on laboratory investigations that were devoted to in vitro and in vivo animal (cyto)genetic studies. However, it may be assumed that some of the most important observations are those obtained from studies with individuals that were exposed to relatively high levels of radiofrequency radiation, either as a result of their occupational activity or as frequent users of radiofrequency emitting tools. In this paper the cytogenetic biomonitoring studies of RF-exposed humans are reviewed. A majority of these studies do show that RF-exposed individuals have increased frequencies of genetic damage (e.g., chromosomal aberrations) in their lymphocytes or exfoliated buccal cells. However, most of the studies, if not all, have a number of shortcomings that actually prevents any firm conclusion. Radiation dosimetry was lacking in all papers, but some of the investigations were flawed by much more severe imperfections. Large well-coordinated multidisciplinary investigations are needed in order to reach any robust conclusion.

(E) Vian A, Roux D, Girard S, Bonnet P, Paladian F, Davies E, Ledoigt G. Microwave irradiation affects gene expression in plants. Plant Signal Behav. 1(2):67-70, 2006.(VO, AE, GE)

The physiological impact of nonionizing radiation has long been considered negligible. However, here we use a carefully calibrated stimulation system that mimics the characteristics (isotropy and homogeneity) of electromagnetic fields present in the environment to measure changes in a molecular marker (mRNA encoding the stress-related bZIP transcription factor), and show that low amplitude, short duration, 900 MHz EMF evokes the accumulation of this mRNA. Accumulation is rapid (peaking 5-15 min after stimulation) and strong (3.5-fold), and is similar to that evoked by mechanical stimulations.

(E) Vijayalaxmi, Frei, MR, Dusch, SJ, Guel, V, Meltz, ML, Jauchem, JR, Frequency of micronuclei in the peripheral blood and bone marrow of cancer-prone mice chronically exposed to 2450 MHz radiofrequency radiation. *Radiat Res* 147(4):495-500, 1997a. (VO, LE, GT)

C3H/HeJ mice, which are prone to mammary tumors, were exposed for 20 h/day, 7 days/week, over 18 months to continuous-wave 2450 MHz radiofrequency (RF) radiation in circularly polarized wave guides at a whole-body average specific absorption rate of 1.0 W/kg. Sham-exposed mice were used as controls. The positive controls were the sentinel mice treated with mitomycin C during the last 24 h before necropsy. At the end of the 18 months, all mice were necropsied. Peripheral blood and bone marrow smears were examined for the extent of genotoxicity as indicated by the presence of micronuclei in polychromatic erythrocytes (PCEs). The results indicate that the incidence of micronuclei/1,000 PCEs was not significantly different between groups exposed to RF radiation (62 mice) and sham-exposed groups (58 mice), and the mean frequencies were 4.5 +/- 1.23 and 4.0 +/- 1.12 in peripheral blood and 6.1 +/- 1.78 and 5.7 +/- 1.60 in bone marrow, respectively. In contrast, the positive controls (7 mice) showed a significantly elevated incidence of micronuclei/1,000 PCEs in peripheral blood and bone marrow, and the mean frequencies were 50.9 +/- 6.18 and 55.2 +/- 4.65, respectively. When

the animals with mammary tumors were considered separately, there were no significant differences in the incidence of micronuclei/1,000 PCEs between the group exposed to RF radiation (12 mice) and the sham-exposed group (8 mice), and the mean frequencies were 4.6 +/- 1.03 and 4.1 +/- 0.89 in peripheral blood and 6.1 +/- 1.76 and 5.5 +/- 1.51 in bone marrow, respectively. Thus there was no evidence for genotoxicity in mice prone to mammary tumors that were exposed chronically to 2450 MHz RF radiation compared with sham-exposed controls.

A correction was published in a subsequent issue of the journal, stating that there was actually a significant increase in micronucleus formation in peripheral blood and bone marrow cells after chronic exposure to the radiofrequency radiation. "Vijayalaxmi, Frei ,MR, Dusch, SJ, Guel, V, Meltz, ML, Jauchem, JR, Correction of an error in calculation in the article "Frequency of micronuclei in the peripheral blood and bone marrow of cancer-prone mice chronically exposed to 2450 MHz radiofrequency radiation" (Radiat. Res. 147, 495-500, 1997). *Radiat Res* 149(3):308, 1998 "

(NE) Vijayalaxmi, Mohan N, Meltz ML, Wittler MA. Proliferation and cytogenetic studies in human blood lymphocytes exposed in vitro to 2450 MHz radiofrequency radiation. Int J Radiat Biol. 72: 751-757, 1997b. (VT, AE, GT)

Aliquots of human peripheral blood collected from two healthy human volunteers were exposed in vitro to continuous wave 2450 MHz radiofrequency radiation (RFR), either continuously for a period of 90 min or intermittently for a total exposure period of 90 min (30 min on and 30 min off, repeated three times). Blood aliquots which were sham-exposed or exposed in vitro to 150 cGy gamma radiation served as controls. The continuous wave 2450 MHz RFR was generated with a net forward power of 34.5 W and transmitted from a standard gain rectangular antenna horn in a vertically downward direction. The mean power density at the position of the cells was 5.0 mW/cm2. The mean specific absorption rate calculated by Finite Difference Time Domain analysis was 12.46 W/kg. Immediately after exposure, lymphocytes were cultured for 48 and 72 h to determine the incidence of chromosomal aberrations and micronuclei, respectively. Proliferation indices were also recorded. There were no significant differences between RFR-exposed and sham-exposed lymphocytes with respect to; (a) mitotic indices; (b) incidence of cells showing chromosome damage; (c) exchange aberrations; (d) acentric fragments; (e) binucleate lymphocytes, and (f) micronuclei, for either the continuous or intermittent RFR exposures. In contrast, the response of positive control cells exposed to 150 cGy gamma radiation was significantly different from RFR-exposed and sham-exposed lymphocytes. Thus, there is no evidence for an effect on mitogen-stimulated proliferation kinetics or for excess genotoxicity within 72 h in human blood lymphocytes exposed in vitro to 2450 MHz RFR.

(NE) Vijayalaxmi, Seaman RL, Belt ML, Doyle JM, Mathur SP, Prihoda TJ, Frequency of micronuclei in the blood and bone marrow cells of mice exposed to ultra-wideband electromagnetic radiation. Int J Radiat Biol 75(1):115-120, 1999. (VO, AE, LI, GT)

PURPOSE: To investigate the extent of genetic damage in the peripheral blood and bone marrow cells of mice exposed to ultra-wideband electromagnetic radiation (UWBR). MATERIALS AND METHODS: CF-1 male mice were exposed to UWBR for 15 min at an estimated whole-body

average specific absorption rate of 37 mWx kg(-1). Groups of untreated control and positive control mice injected with mitomycin C were also included in the study. After various treatments, half of the mice were killed at 18 h, and the other half at 24 h. Peripheral blood and bone marrow smears were examined to determine the extent of genotoxicity, as assessed by the presence of micronuclei (MN) in polychromatic erythrocytes (PCE). RESULTS: The percentages of PCE and the incidence of MN per 2000 PCE in both tissues in mice killed at 18 h were similar to the frequencies observed in mice terminated at 24 h. There were no significant differences in the percentage of PCE between control and the mice with or without UWBR exposure; the group mean values (+/- standard deviation) were in the range of 3.1+/-0.14 to 3.2+/-0.23 in peripheral blood, and 49.0+/-3.56 to 52.3+/-4.02 in bone marrow. The mean incidence of MN per 2000 PCE in control and in mice with or without UWBR exposure ranged from 7.7+/-2.00 to 9.7+/-2.54 in peripheral blood and 7.4+/-2.32 to 10.0+/-3.27 in bone marrow. Pairwise comparison of the data did not reveal statistically significant differences between the control and mice with or without UWBR exposure groups (excluding positive controls). CONCLUSION: Under the experimental conditions tested, there was no evidence for excess genotoxicity in peripheral blood or bone marrow cells of mice exposed to UWBR.

(NE) Vijayalaxmi, Leal BZ, Szilagyi M, Prihoda TJ, Meltz ML, Primary DNA damage in human blood lymphocytes exposed In vitro to 2450 MHz radiofrequency radiation. Radiat Res 153(4):479-486, 2000. (VT, AE, GT)

Human peripheral blood samples collected from three healthy human volunteers were exposed in vitro to pulsed-wave 2450 MHz radiofrequency (RF) radiation for 2 h. The RF radiation was generated with a net forward power of 21 W and transmitted from a standard gain rectangular antenna horn in a vertically downward direction. The average power density at the position of the cells in the flask was 5 mW/cm(2). The mean specific absorption rate, calculated by finite difference time domain analysis, was 2.135 (+/-0.005 SE) W/kg. Aliquots of whole blood that were sham-exposed or exposed in vitro to 50 cGy of ionizing radiation from a (137)Cs gamma-ray source were used as controls. The lymphocytes were examined to determine the extent of primary DNA damage (single-strand breaks and alkali-labile lesions) using the alkaline comet assay with three different slide-processing schedules. The assay was performed on the cells immediately after the exposures and at 4 h after incubation of the exposed blood at 37 +/- 1 degrees C to allow time for rejoining of any strand breaks present immediately after exposure, i.e. to assess the capacity of the lymphocytes to repair this type of DNA damage. At either time, the data indicated no significant differences between RF-radiation- and sham-exposed lymphocytes with respect to the comet tail length, fluorescence intensity of the migrated DNA in the tail, and tail moment. The conclusions were similar for each of the three different comet assay slide-processing schedules examined. In contrast, the response of lymphocytes exposed to ionizing radiation was significantly different from RF-radiation- and sham-exposed cells. Thus, under the experimental conditions tested, there is no evidence for induction of DNA single-strand breaks and alkali-labile lesions in human blood lymphocytes exposed in vitro to pulsed-wave

2450 MHz radiofrequency radiation, either immediately or at 4 h after exposure.

(NE) Vijayalaxmi , Leal BZ, Meltz ML, Pickard WF, Bisht KS, Roti Roti JL , Straube WL, Moros EG, Cytogenetic studies in human blood lymphocytes exposed in vitro to radiofrequency radiation at a cellular telephone frequency (835.62 MHz, FDMA). Radiat Res 155(1):113-121, 2001a. (VO, AE, GT)

Freshly collected peripheral blood samples from four healthy human volunteers were diluted with RPMI 1640 tissue culture medium and exposed in sterile T-75 tissue culture flasks in vitro for 24 h to 835.62 MHz radiofrequency (RF) radiation, a frequency employed for customer-to-base station transmission of cellular telephone communications. An analog signal was used, and the access technology was frequency division multiple access (FDMA, continuous wave). A nominal net forward power of 68 W was used, and the nominal power density at the center of the exposure flask was 860 W/m(2). The mean specific absorption rate in the exposure flask was 4.4 or 5.0 W/kg. Aliquots of diluted blood that were sham-exposed or exposed in vitro to an acute dose of 1.50 Gy of gamma radiation were used as negative or positive controls. Immediately after the exposures, the lymphocytes were stimulated with a mitogen, phytohemagglutinin, and cultured for 48 or 72 h to determine the extent of genetic damage, as assessed from the frequencies of chromosomal aberrations and micronuclei. The extent of alteration in the kinetics of cell proliferation was determined from the mitotic indices in 48-h cultures and from the incidence of binucleate cells in 72-h cultures. The data indicated no significant differences between RF-radiation- and sham-exposed lymphocytes with respect to mitotic indices, incidence of exchange aberrations, excess fragments, binucleate cells, and micronuclei. In contrast, the response of the lymphocytes exposed to gamma radiation was significantly different from both RF-radiation- and sham-exposed cells for all of these indices. Thus, under the experimental conditions tested, there is no evidence for the induction of chromosomal aberrations and micronuclei in human blood lymphocytes exposed in vitro for 24 h to 835.62 MHz RF radiation at SARs of 4.4 or 5.0 W/kg.

(NE) **Vijayalaxmi, Pickard WF, Bisht KS, Prihoda TJ, Meltz ML, LaRegina MC, Roti Roti JL, Straube WL, Moros EG. Micronuclei in the peripheral blood and bone marrow cells of rats exposed to 2450 MHz radiofrequency radiation. Int J Radiat Biol. 77(11):1109-1115, 2001b. (VT, AE, GT)**

PURPOSE: To determine the incidence of micronuclei in peripheral blood and bone marrow cells of rats exposed continuously for 24 h to 2450 MHz continuous wave radiofrequency radiation (RFR) at an average whole-body specific absorption rate (SAR) of 12 W/kg. MATERIALS AND METHODS: Eight adult male Sprague-Dawley rats were exposed to 2450 MHz RFR in circularly polarized waveguides. Eight sham-exposed rats were kept in similar waveguides without the transmission of RFR. Four rats were treated with mitomycin-C (MMC) and used as positive controls. All rats were necropsied 24 h after the end of RFR and sham exposures, and after the 24h treatment with MMC. Peripheral blood and bone marrow smears were examined to determine the frequency of micronuclei (MN) in polychromatic erythrocytes (PCE). RESULTS:

The results indicated that the incidence of MN/2000 PCE were not significantly different between RFR- and sham-exposed rats. The group mean frequencies of MN in the peripheral blood were 2.3+/-0.7 in RFR-exposed rats and 2.1+/-0.6 in sham-exposed rats. In bone marrow cells, the average MN incidence was 3.8+/-1.0 in RFR-exposed rats and 3.4+/-0.7 in sham-exposed rats. The corresponding values in positive control rats treated with MMC were 23.5+/-4.7 in the peripheral blood and 33.8+/-7.4 in bone marrow cells. CONCLUSION: There was no evidence for the induction of MN in peripheral blood and bone marrow cells of rats exposed for 24 h to 2450 MHz continuous wave RFR at a whole body average SAR of 12 W/kg.

(NE) Vijayalaxmi, Bisht KS, Pickard WF, Meltz ML, Roti Roti JL, Moros EG. Chromosome damage and micronucleus formation in human blood lymphocytes exposed in vitro to radiofrequency radiation at a cellular telephone frequency (847.74 MHz, CDMA). Radiat Res 156(4):430-432, 2001c.(VT, AE. GT)

Peripheral blood samples collected from four healthy nonsmoking human volunteers were diluted with tissue culture medium and exposed in vitro for 24 h to 847.74 MHz radiofrequency (RF) radiation (continuous wave), a frequency employed for cellular telephone communications. A code division multiple access (CDMA) technology was used with a nominal net forward power of 75 W and a nominal power density of 950 W/m(2) (95 mW/cm(2)). The mean specific absorption rate (SAR) was 4.9 or 5.5 W/kg. Blood aliquots that were sham-exposed or exposed in vitro to an acute dose of 1.5 Gy of gamma radiation were included in the study as controls. The temperatures of the medium during RF-radiation and sham exposures in the Radial Transmission Line facility were controlled at 37 +/- 0.3 degrees C. Immediately after the exposures, lymphocytes were cultured at 37 +/- 1 degrees C for 48 or 72 h. The extent of genetic damage was assessed from the incidence of chromosome aberrations and micronuclei. The kinetics of cell proliferation was determined from the mitotic indices in 48-h cultures and from the incidence of binucleate cells in 72-h cultures. The data indicated no significant differences between RF-radiation-exposed and sham-exposed lymphocytes with respect to mitotic indices, frequencies of exchange aberrations, excess fragments, binucleate cells, and micronuclei. The response of gamma-irradiated lymphocytes was significantly different from that of both RF-radiation-exposed and sham-exposed cells for all of these indices. Thus there was no evidence for induction of chromosome aberrations and micronuclei in human blood lymphocytes exposed in vitro for 24 h to 847.74 MHz RF radiation (CDMA) at SARs of 4.9 or 5.5 W/kg.

(NE) Vijayalaxmi, Sasser LB, Morris JE, Wilson BW, Anderson LE. Genotoxic potential of 1.6 GHz wireless communication signal: in vivo two-year bioassay. Radiat Res 159(4):558-564, 2003.(VO, LE, GT)

Timed-pregnant Fischer 344 rats (from nineteenth day of gestation) and their nursing offspring (until weaning) were exposed to a far-field 1.6 GHz Iridium wireless communication signal for 2 h/day, 7 days/week. Far-field whole-body exposures

were conducted with a field intensity of 0.43 mW/cm(2) and whole-body average specific absorption rate (SAR) of 0.036 to 0.077 W/kg (0.10 to 0.22 W/kg in the brain). This was followed by chronic, head-only exposures of male and female offspring to a near-field 1.6 GHz signal for 2 h/day, 5 days/week, over 2 years. Near-field exposures were conducted at an SAR of 0.16 or 1.6 W/kg in the brain. Concurrent sham-exposed and cage control rats were also included in the study. At the end of 2 years, all rats were necropsied. Bone marrow smears were examined for the extent of genotoxicity, assessed from the presence of micronuclei in polychromatic erythrocytes. The results indicated that the incidence of micronuclei/2000 polychromatic erythrocytes were not significantly different between 1.6 GHz-exposed, sham-exposed and cage control rats. The group mean frequencies were 5.6 +/- 1.8 (130 rats exposed to 1.6 GHz at 0.16 W/kg SAR), 5.4 +/- 1.5 (135 rats exposed to 1.6 GHz at 1.6 W/kg SAR), 5.6 +/- 1.7 (119 sham-exposed rats), and 5.8 +/- 1.8 (100 cage control rats). In contrast, positive control rats treated with mitomycin C exhibited significantly elevated incidence of micronuclei/2000 polychromatic erythrocytes in bone marrow cells; the mean frequency was 38.2 +/- 7.0 (five rats). Thus there was no evidence for excess genotoxicity in rats that were chronically exposed to 1.6 GHz compared to sham-exposed and cage controls.

(NE) Vijayalaxmi, Logani MK, Bhanushali A, Marvin C. Ziskin MC, Prihoda TJ. Micronuclei in peripheral blood and bone marrow cells of mice exposed to 42 GHz electromagnetic millimeter waves. Radiat Res. 161: 341–345, 2004. (VO, LE, GT)

The genotoxic potential of 42.2 +/- 0.2 GHz electromagnetic millimeter-wave radiation was investigated in adult male BALB/c mice. The radiation was applied to the nasal region of the mice for 30 min/day for 3 consecutive days. The incident power density used was 31.5 +/- 5.0 mW/cm2. The peak specific absorption rate was calculated as 622 +/- 100 W/kg. Groups of mice that were injected with cyclophosphamide (15 mg/kg body weight), a drug used in the treatment of human malignancies, were also included to determine if millimeter-wave radiation exposure had any influence on drug-induced genotoxicity. Concurrent sham-exposed and untreated mice were used as controls. The extent of genotoxicity was assessed from the incidence of micronuclei in polychromatic erythrocytes of peripheral blood and bone marrow cells collected 24 h after treatment. The results indicated that the incidence of micronuclei in 2000 polychromatic erythrocytes was not significantly different among untreated, millimeter wave-exposed, and sham-exposed mice. The group mean incidences were 6.0 +/- 1.6, 5.1 +/- 1.5 and 5.1 +/- 1.3 in peripheral blood and 9.1 +/- 1.1, 9.3 +/- 1.6 and 9.1 +/- 1.6 in bone marrow cells, respectively. Mice that were injected with cyclophosphamide exhibited significantly increased numbers of micronuclei, 14.6 +/- 2.7 in peripheral blood and 21.3 +/- 3.9 in bone marrow cells (P< 0.0001). The drug-induced micronuclei were not significantly different in millimeter wave-exposed and sham-exposed mice; the mean incidences were 14.3 +/- 2.8 and 15.4 +/- 3.0 in peripheral blood and 23.5 +/- 2.3 and 22.1 +/- 2.5 in bone marrow cells, respectively. Thus there was no evidence for the induction of genotoxicity in the peripheral blood and bone marrow cells of mice exposed to electromagnetic millimeter-wave radiation. Also, millimeter-wave radiation exposure did not influence cyclophosphamide-induced micronuclei in either type of cells.

(NE) Vijayalaxmi. Cytogenetic studies in human blood lymphocytes exposed in vitro to 2.45 GHz or 8.2 GHz radiofrequency radiation. Radiat Res 166, 532–538, 2006. (VT, AE, GT)

Peripheral blood samples collected from healthy human volunteers were exposed in vitro to 2.45 GHz or 8.2 GHz pulsed-wave radiofrequency (RF) radiation. The net forward power, average power density, mean specific absorption rate, and the temperature maintained during the 2-h exposure of the cells to 2.45 GHz or 8.2 GHz were, respectively, 21 W or 60 W, 5 mW/cm^2 or 10 mW/cm^2, 2.13 W/kg or 20.71 W/kg, and 36.9 ± 0.1°C or 37.5 ± 0.2°C. Aliquots of the same blood samples that were either sham-exposed or exposed in vitro to an acute dose of 1.5 Gy γ radiation were used as unexposed and positive controls, respectively. Cultured lymphocytes were examined to determine the extent of cytogenetic damage assessed from the incidence of chromosomal aberrations and micronuclei. Under the conditions used to perform the experiments, the levels of damage in RF-radiation-exposed and sham-exposed lymphocytes were not significantly different. Also, there were no significant differences in the response of unstimulated lymphocytes and lymphocytes stimulated with phytohemagglutinin when exposed to 8.2 GHz RF radiation. In contrast, the positive control cells that had been subjected to γ irradiation exhibited significantly more damage than RF-radiation- and sham-exposed lymphocytes.

Vijayalaxmi, Prihoda TJ. Comprehensive review of quality of publications and meta-analysis of genetic damage in mammalian cells exposed to non-ionizing radiofrequency fields. Radiat Res. 191(1):20-30, 2019. (Review)

There have been numerous published studies reporting on the extent of genetic damage observed in animal and human cells exposed in vitro and in vivo to non-ionizing radiofrequency fields (RF, electromagnetic waves that carry energy as they propagate in air and dense media). Overall, the data are inconsistent; while some studies have suggested significantly increased damage in cells exposed to RF energy compared to unexposed and/or sham-exposed control cells, others have not. Several variables in exposure conditions used in the experiments might have contributed to the controversy. In this comprehensive review, four specific quality control measures were used to determine the quality of 225 published studies in animal and human cells exposed in vitro and in vivo to RF energy, and the results from 2,160 tests with different sample sizes were analyzed. The four specific quality control measures were as follows: 1. "Blind" collection/analysis of the data to eliminate individual/observer "bias"; 2. Adequate description of "dosimetry" for independent replication/confirmation; 3. Inclusion of "positive controls" to confirm the outcomes; and 4. Inclusion of "sham-exposed controls" which are more appropriate to compare the data with those in RF exposure conditions. In addition, meta-analysis of the genetic damage in cells exposed to RF energy and control cells, thus far available in the RF literature database, was performed to obtain the "d" values, i.e., standardized mean difference between these two types of cells or the effect size. The relationship between d values and the above-mentioned quality control measures was ascertained. In addition, the correlation between the quality control measures and the conclusions reported in the publications (no significant difference between the cells exposed to RF energy and control cells; increased damage in former cells compared to the latter; increased, no significant difference and decreased damage in cells exposed to RF energy in the same experiment; or decreased damage in cells exposed to RF

energy) was examined. The overall conclusions were as follows: 1. When all four quality control measures were mentioned in the publication, the d values were smaller compared to those when one or more quality control measures were not mentioned in the investigation; 2. Based on the inclusion of quality control measures, the weighted outcome in cells exposed to RF energy (d values) indicated a very small effect, if any; 3. The number of published studies reporting no significant difference in genetic damage of cells exposed to RF energy, compared to that of control cells, increased with increased number of quality control measures employed in investigations; 4. The number of published studies reporting increased genetic damage in cells exposed to RF energy decreased with increased number of quality control measures; and 5. There was a "bias" towards the publications reporting increased genetic damage in cells exposed to RF energy even with very small sample size. Overall, the results from this study underscore the importance of including quality control measures in investigations so that the resulting data are useful, nationally and internationally, in evaluating "potential" health risks from exposure to RF energy

(E) Vilić, M., Tlak Gajger, I., Tucak, P., Štambuk, A., Šrut, M., Klobučar, G., Malarić, K., Žura Žaja, I., Pavelić, A., Manger, M., and Tkalec, M. Effects of short-term exposure to mobile phone radiofrequency (900 MHz) on the oxidative response and genotoxicity in honey bee larvae. J. Apic. Res. 56, 430–438, 2017. (VO, AE, GT, OX, WS)

Exposure of different animal species to radiofrequency electromagnetic fields (RF-EMF) could cause various biological effects such as oxidative stress, genotoxic effects and dysfunction of the immune system. However, there are a lack of results on oxidative stress response and genotoxicity in the honey bee (*Apis mellifera*) after exposure to RF-EMF. This study was performed to investigate the effects of exposure to RF-EMF on the activity of catalase, superoxide dismutase, glutathione *S*-transferase, lipid peroxidation level and DNA damage in honey bee larvae. Honey bee larvae were exposed to RF-EMF at 900 MHz and field levels of 10, 23, 41 and 120 V m^{-1} for 2 h. At a field level of 23 V m^{-1} the effect of 80% AM 1 kHz sinusoidal and 217 Hz modulation was investigated as well. Catalase activity and the lipid peroxidation level decreased significantly in the honey bee larvae exposed to the unmodulated field at 10 V m^{-1} compared to the control. Superoxide dismutase and glutathione *S*-transferase activity in the honey bee larvae exposed to unmodulated fields were not statistically different compared to the control. DNA damage increased significantly in honey bee larvae exposed to modulated (80% AM 1 kHz sinus) field at 23 V m^{-1} compared to the control and all other exposure groups. These results suggest that RF-EMF effects in honey bee larvae appeared only after exposure to a certain EMF conditions. The increase of the field level did not cause a linear dose-response in any of the measured parameters. Modulated RF-EMF produced more negative effects than the corresponding unmodulated field. Although honey bees in nature would not be exposed to such high field levels as used in our experiments, our results show the need for further intensive research in all stages of honey bee development.

(NE) Waldmann P, Bohnenberger S, Greinert R, Hermann-Then B, Heselich A, Klug SJ, Koenig J, Kuhr K, Kuster N, Merker M, Murbach M, Pollet D, Schadenboeck W, Scheidemann-Wesp U, Schwab B, Volkmer B, Weyer V, Blettner M. Influence of GSM signals on human peripheral lymphocytes: study of genotoxicity. Radiat Res. 179(2):243-253, 2013. (VT, AE, GT)

Exposure to radiofrequency (RF) electromagnetic fields (EMF) is continuously increasing worldwide. Yet, conflicting results of a possible genotoxic effect of RF EMF continue to be discussed. In the present study, a possible genotoxic effect of RF EMF (GSM, 1,800 MHz) in human lymphocytes was investigated by a collaboration of six independent institutes (institutes a, b, c, d, e, h). Peripheral blood of 20 healthy, nonsmoking volunteers of two age groups (10 volunteers 16-20 years old and 10 volunteers 50-65 years old) was taken, stimulated and intermittently exposed to three specific absorption rates (SARs) of RF EMF (0.2 W/kg, 2 W/kg, 10 W/kg) and sham for 28 h (institute a). The exposures were performed in a setup with strictly controlled conditions of temperature and dose, and randomly and automatically determined waveguide SARs, which were designed and periodically maintained by ITIS (institute h). Four genotoxicity tests with different end points were conducted (institute a): chromosome aberration test (five types of structural aberrations), micronucleus test, sister chromatid exchange test and the alkaline comet assay (Olive tail moment and % DNA). To demonstrate the validity of the study, positive controls were implemented. The genotoxicity end points were evaluated independently by three laboratories blind to SAR information (institute c = laboratory 1; institute d = laboratory 2; institute e = laboratory 3). Statistical analysis was carried out by institute b. Methods of primary statistical analysis and rules to adjust for multiple testing were specified in a statistical analysis plan based on a data review before unblinding. A linear trend test based on a linear mixed model was used for outcomes of comet assay and exact permutation test for linear trend for all other outcomes. It was ascertained that only outcomes with a significant SAR trend found by at least two of three analyzing laboratories indicated a substantiated suspicion of an exposure effect. On the basis of these specifications, none of the nine end points tested for SAR trend showed a significant and reproducible exposure effect. Highly significant differences between sham exposures and positive controls were detected by each analyzing laboratory, thus validating the study. In conclusion, the results show no evidence of a genotoxic effect induced by RF EMF (GSM, 1,800 MHz).

(E) Wang X, Liu C, Ma Q, Feng W, Yang L, Lu Y, Zhou Z, Yu Z, Li W, Zhang L. 8-oxoG DNA glycosylase-1 inhibition sensitizes Neuro-2a cells to oxidative DNA base damage induced by 900 MHz radiofrequency electromagnetic radiation. Cell Physiol Biochem. 37(3):1075-1088, 2015. (VT, AE, GT, OX)

BACKGROUND/AIMS: The purpose of this study was to explore the in vitro putative genotoxicity during exposure of Neuro-2a cells to radiofrequency electromagnetic fields (RF-EMFs) with or without silencing of 8-oxoG DNA glycosylase-1 (OGG1). METHODS: Neuro-2a cells treated with or without OGG1 siRNA were exposed to 900 MHz Global System for Mobile Communication (GSM) Talk signals continuously at a specific absorption rate (SAR) of 0, 0.5, 1 or 2 W/kg for 24 h. DNA strand breakage and DNA base damage were measured by the alkaline comet assay and a modified comet assay using formamidopyrimidine DNA glycosylase (FPG), respectively. Reactive oxygen species (ROS) levels and cell viability were monitored using the non-fluorescent probe 2, 7-dichlorofluorescein diacetate (DCFH-DA) and CCK-8 assay. RESULTS: Exposure to 900 MHz RF-EMFs with insufficient energy could induce oxidative DNA base damage in Neuro-2a cells. These increases were concomitant with similar increases in the generation of reactive oxygen species (ROS). Without OGG1 siRNA, 2 W/kg RF-EMFs induced oxidative DNA base damage in Neuro-2a cells. Interestingly, with OGG1

siRNA, RF-EMFs could cause DNA base damage in Neuro-2a cells as low as 1 W/kg. However, neither DNA strand breakage nor altered cell viability was observed. CONCLUSION: Even if further studies remain conducted we support the hypothesis that OGG1 is involved in the process of DNA base repair and may play a pivotal role in protecting DNA bases from RF-EMF induced oxidative damage.

(E) Wang Y, Jiang Z, Zhang L, Zhang Z, Liao Y, Cai P 3.5-GHz radiofrequency electromagnetic radiation promotes the development of Drosophila melanogaster. Environ Pollut 294:118646, 2022. (VO, LE, LI, GE)

With the rapidly increasing popularity of 5G mobile technology, the effect of radiofrequency radiation on human health has caused public concern. This study explores the effects of a simulated 3.5 GHz radiofrequency electromagnetic radiation (RF-EMF) environment on the development and microbiome of flies under intensities of 0.1 W/m^2, 1 W/m^2 and 10 W/m^2. We found that the pupation percentages in the first 3 days and eclosion rate in the first 2 days were increased under exposure to RF-EMF, and the mean development time was shortened. In a study on third-instar larvae, the expression levels of the heat shock protein genes hsp22, hsp26 and hsp70 and humoral immune system genes AttC, TotC and TotA were all significantly increased. In the oxidative stress system, DuoX gene expression was decreased, sod2 and cat gene expression levels were increased, and SOD and CAT enzyme activity also showed a significant increase. According to the 16S rDNA results, the diversity and species abundance of the microbial community decreased significantly, and according to the functional prediction analysis, the genera Acetobacter and Lactobacillus were significantly increased. In conclusion, 3.5 GHz RF-EMF may enhance thermal stress, oxidative stress and humoral immunity, cause changes in the microbial community, and regulate the insulin/TOR and ecdysteroid signalling pathways to promote fly development.

(NE) Whitehead TD, Brownstein BH, Parry JJ, Thompson D, Cha BA, Moros EG, Rogers BE, Roti Roti JL. Expression of the Proto-oncogene Fos after Exposure to Radiofrequency Radiation Relevant to Wireless Communications. Radiat Res. 164(4):420-430, 2005. (VT, AE, GE)

In this study the expression levels of the proto-oncogene Fos were measured after exposure to radiofrequency (RF) radiation at two relatively high specific absorption rates (SARs) of 5 and 10 W/kg for three types of modulated signals: 847.74 MHz code division multiple access (CDMA), 835.62 MHz frequency division multiple access (FDMA), and 836.55 MHz time division multiple access (TDMA). This work was undertaken to confirm a previous report by Goswami et al. (Radiat. Res. 151, 300-309, 1999) that CDMA and FDMA radiation caused small but statistically significant increases in Fos levels as cells entered plateau phase during exposure. No effects on Myc or Jun levels were observed in that study. Therefore, in the present study, analyses were restricted to Fos expression during the transition from exponential growth to plateau phase. Fos expression was measured using the real-time polymerase chain reaction (RT-PCR) technique. Serum-stimulated C3H 10T(1/2) cells were used as a positive control for Fos expression. Possible influences of final cell number or pH variability on Fos expression were evaluated. Expression of Fos mRNA in C3H 10T(1/2) cells was not significantly different from

that found after sham exposure at either SAR level for any signal modulation. Therefore, the results of Goswami et al. could not be confirmed.

(NE) Whitehead TD, Moros EG, Brownstein BH, Roti Roti JL. Gene expression does not change significantly in C3H 10T(1/2) cells after exposure to 847.74 CDMA or 835.62 FDMA radiofrequency radiation. Radiat Res. 165(6):626-635, 2006a. (VT, AE, GE)

.In vitro experiments with C3H 10T(1/2) mouse cells were performed to determine whether Frequency Division Multiple Access (FDMA) or Code Division Multiple Access (CDMA) modulated radiofrequency (RF) radiations induce changes in gene expression. After the cells were exposed to either modulation for 24 h at a specific absorption rate (SAR) of 5 W/ kg, RNA was extracted from both exposed and sham-exposed cells for gene expression analysis. As a positive control, cells were exposed to 0.68 Gy of X rays and gene expression was evaluated 4 h after exposure. Gene expression was evaluated using the Affymetrix U74Av2 GeneChip to detect changes in mRNA levels. Each exposure condition was repeated three times. The GeneChip data were analyzed using a two-tailed t test, and the expected number of false positives was estimated from t tests on 20 permutations of the six sham RF-field-exposed samples. For the X-ray-treated samples, there were more than 90 probe sets with expression changes greater than 1.3-fold beyond the number of expected false positives. Approximately one-third of these genes had previously been reported in the literature as being responsive to radiation. In contrast, for both CDMA and FDMA radiation, the number of probe sets with an expression change greater than 1.3-fold was less than or equal to the expected number of false positives. Thus the 24-h exposures to FDMA or CDMA RF radiation at 5 W/kg had no statistically significant effect on gene expression.

(NE) Whitehead TD, Moros EG, Brownstein BH, Roti Roti JL. The number of genes changing expression after chronic exposure to Code Division Multiple Access or Frequency DMA radiofrequency radiation does not exceed the false-positive rate. Proteomics. 6(17):4739-4744, 2006b. (VT, AE, GE)

Experiments with cultured C3H 10T 1/2 cells were performed to determine if exposure to cell phone radiofrequency (RF) radiations induce changes in gene expression. Following a 24 h exposure of 5 W/kg specific adsorption rate, RNA was extracted from the exposed and sham control cells for microarray analysis on Affymetrix U74Av2 Genechips. Cells exposed to 0.68 Gy of X-rays with a 4-h recovery were used as positive controls. The number of gene expression changes induced by RF radiation was not greater than the number of false positives expected based on a sham versus sham comparison. In contrast, the X-irradiated samples showed higher numbers of probe sets changing expression level than in the sham versus sham comparison.

(E)Wu H, Wang D, Shu Z, Zhou H, Zuo H, Wang S, Li Y, Xu X, Li N, Peng R. Cytokines produced by microwave-radiated Sertoli cells interfere with spermatogenesis in rat testis. Andrologia 44 Suppl 1:590-599, 2012. (VT, AE, GE, RP)

Microwave radiation resulted in degeneration, apoptosis or necrosis in germ cells at different stages. The molecular mechanisms by which microwaves induce spermatogenesis disorder have not been completely understood. Sertoli cells play crucial roles in mammalian spermatogenesis. Cytokines produced by Sertoli cells play pleiotropic roles in different conditions. At physiologically low concentration, TNFα, IL-1β and IL-6 behave as survival factors; while under pathological condition, these cytokines can induce apoptosis in testis. The effects of cytokines produced by microwave-radiated Sertoli cells on spermatogenesis are poorly understood. The purpose of this study was to determine the effect of cytokines produced by microwave-radiated Sertoli cells on the germ cells. We focused the effect of TNFα, IL-1β and IL-6 on the germ cells. The results showed that TNFα, IL-1β and IL-6 were increased in Sertoli cells after exposure to microwave radiation. These up-regulated cytokines can induce apoptosis and lipid peroxidation in the membrane of germ cells. In addition, germ cell apoptosis was associated with the up-regulation of Bax/Bcl-2 and caspase-3. These results suggest that cytokines produced by microwave-radiated Sertoli cells may disrupt spermatogenesis. Our data provided novel insight into the injury mechanism of germ cells induced by microwave radiation

(E) Wu W, Yao K, Wang KJ, Lu DQ, He JL, Xu LH, Sun WJ. [Blocking 1800 MHz mobile phone radiation-induced reactive oxygen species production and DNA damage in lens epithelial cells by noise magnetic fields.]Zhejiang Da XueXueBao Yi Xue Ban 37:34-38, 2008. [Article in Chinese] (VT, AE, GT, IX, OX)

OBJECTIVE: To investigate whether the exposure to the electromagnetic noise can block reactive oxygen species (ROS) production and DNA damage of lens epithelial cells induced by 1800 MHz mobile phone radiation. METHODS: The DCFH-DA method and comet assay were used respectively to detect the intracellular ROS and DNA damage of cultured human lens epithelial cells induced by 4 W/kg 1800 MHz mobile phone radiation or/and 2microT electromagnetic noise for 24 h intermittently. RESULT: 1800 MHz mobile phone radiation at 4 W/kg for 24 h increased intracellular ROS and DNA damage significantly ($P<0.05$). However, the ROS level and DNA damage of mobile phone radiation plus noise group were not significant enhanced ($P>0.05$) as compared to sham exposure group. Conclusion: Electromagnetic noise can block intracellular ROS production and DNA damage of human lens epithelial cells induced by 1800 MHz mobile phone radiation.

(E) Xu S, Zhong M, Zhang L, Zhou Z, Zhang W, Wang Y, Wang X, Li M, Chen Y, Chen C, He M, Zhang G, Yu Z. Exposure to 1800 MHz radiofrequency radiation induces oxidative damage to mitochondrial DNA in primary cultured neurons. Brain Res 1311:189-196. 2010. (VT, AE, GT, OX)

Increasing evidence indicates that oxidative stress may be involved in the adverse effects of radiofrequency (RF) radiation on the brain. Because mitochondrial DNA (mtDNA) defects are closely associated with various nervous system diseases and mtDNA is highly susceptible to oxidative stress, the purpose of this study was to determine whether radiofrequency radiation can cause oxidative damage to mtDNA. In this study, we exposed primary cultured cortical neurons to pulsed RF electromagnetic fields at a frequency of 1800 MHz modulated by 217 Hz at an average special absorption rate (SAR) of 2 W/kg. At 24h after exposure, we found that RF

radiation induced a significant increase in the levels of 8-hydroxyguanine (8-OHdG), a common biomarker of DNA oxidative damage, in the mitochondria of neurons. Consistent with this finding, the copy number of mtDNA and the levels of mitochondrial RNA (mtRNA) transcripts showed an obvious reduction after RF exposure. Each of these mtDNA disturbances could be reversed by pretreatment with melatonin, which is known to be an efficient in the brain. Together, these results suggested that 1800 MHz RF radiation could cause oxidative damage to mtDNA in primary cultured neurons. Oxidative damage to mtDNA may account for the neurotoxicity of RF radiation in the brain.

(E) Xu S, Chen G, Chen C, Sun C, Zhang D, Murbach M, Kuster N, Zeng Q, Xu Z. Cell type-dependent induction of DNA damage by 1800 MHz radiofrequency electromagnetic fields does not result in significant cellular dysfunctions. PLoS One. 8(1):e54906, 2013. (VT, AE, GT, CS)

BACKGROUND: Although IARC clarifies radiofrequency electromagnetic fields (RF-EMF) as possible human carcinogen, the debate on its health impact continues due to the inconsistent results. Genotoxic effect has been considered as a golden standard to determine if an environmental factor is a carcinogen, but the currently available data for RF-EMF remain controversial. As an environmental stimulus, the effect of RF-EMF on cellular DNA may be subtle. Therefore, more sensitive method and systematic research strategy are warranted to evaluate its genotoxicity. **OBJECTIVES:** To determine whether RF-EMF does induce DNA damage and if the effect is cell-type dependent by adopting a more sensitive method γH2AX foci formation; and to investigate the biological consequences if RF-EMF does increase γH2AX foci formation. **METHODS:** Six different types of cells were intermittently exposed to GSM 1800 MHz RF-EMF at a specific absorption rate of 3.0 W/kg for 1 h or 24 h, then subjected to immunostaining with anti-γH2AX antibody. The biological consequences in γH2AX-elevated cell type were further explored with comet and TUNEL assays, flow cytometry, and cell growth assay. **RESULTS:** Exposure to RF-EMF for 24 h significantly induced γH2AX foci formation in Chinese hamster lung cells and Human skin fibroblasts (HSFs), but not the other cells. However, RF-EMF-elevated γH2AX foci formation in HSF cells did not result in detectable DNA fragmentation, sustainable cell cycle arrest, cell proliferation or viability change. RF-EMF exposure slightly but not significantly increased the cellular ROS level. **CONCLUSIONS:** RF-EMF induces DNA damage in a cell type-dependent manner, but the elevated γH2AX foci formation in HSF cells does not result in significant cellular dysfunctions.

(E) Yadav AS, Sharma MK. Increased frequency of micronucleated exfoliated cells among humans exposed in vivo to mobile telephone radiations. Mutat Res.650(2):175-180, 2008. (HU, LE, GT)

The health concerns have been raised following the enormous increase in the use of wireless mobile telephones throughout the world. This investigation had been taken, with the motive to find out whether mobile phone radiations cause any in vivo effects on the frequency of micronucleated exfoliated cells in the exposed subjects. A total of 109 subjects including 85 regular mobile phone users (exposed) and 24 non-users (controls) had participated in this study. Exfoliated cells were obtained by swabbing the buccal-mucosa from exposed as well as sex-age-matched controls. One thousand exfoliated cells were screened from each individual for nuclear

anomalies including micronuclei (MN), karyolysis (KL), karyorrhexis (KH), broken egg (BE) and binucleated (BN) cells. The average daily duration of exposure to mobile phone radiations is 61.26 min with an overall average duration of exposure in term of years is 2.35 years in exposed subjects along with the 9.84+/-0.745 micronucleated cells (MNCs) and 10.72+/-0.889 total micronuclei (TMN) as compared to zero duration of exposure along with average 3.75+/-0.774 MNC and 4.00+/-0.808 TMN in controls. The means are significantly different in case of MNC and TMN at 0.01% level of significance. The mean of KL in controls is 13.17+/-2.750 and in exposed subjects is 13.06+/-1.793. The value of means of KH in exposed subjects (1.84+/-0.432) is slightly higher than in controls (1.42+/-0.737). Mean frequency of broken egg is found to be more in exposed subjects (0.65+/-0.276) as compared to controls (0.50+/-0.217). Frequency of presence of more than one nucleus in a cell (binucleated) is also higher in exposed (2.72+/-0.374) in comparison to controls (0.67+/-0.231). Although there is a slight increase in mean frequency of KH, BE and BN in exposed subjects but the difference is not found statistically significant. Correlation between 0-1, 1-2, 2-3 and 3-4 years of exposure and the frequency of MNC and TMN has been calculated and found to be positively correlated.

Yahyazadeh A, Deniz ÖG, Kaplan AA, Altun G, Yurt KK, Davis D. The genomic effects of cell phone exposure on the reproductive system. Environ Res. 167:684-693, 2018. (review)

Humans are exposed to increasing levels of electromagnetic fields (EMF) at various frequencies as technology advances. In this context, improving understanding of the biological effects of EMF remains an important, high priority issue. Although a number of studies in this issue and elsewhere have focused on the mechanisms of the oxidative stress caused by EMF, the precise understanding of the processes involved remains to be elucidated. Due to unclear results among the studies, the issue of EMF exposure in the literature should be evaluated at the genomic level on the reproductive system. Based on this requirement, a detail review of recently published studies is necessary. The main objectives of this study are to show differences between negative and positive effect of EMF on the reproductive system of animal and human. Extensive review of literature has been made based on well known data bases like Web of Science, PubMed, MEDLINE, Google Scholar, Science Direct, Scopus. This paper reviews the current literature and is intended to contribute to a better understanding of the genotoxic effects of EMF emitted from mobile phones and wireless systems on the human reproductive system, especially on fertility. The current literature reveals that mobile phones can affect cellular functions via non-thermal effects. Although the cellular targets of global system for mobile communications (GSM)-modulated EMF are associated with the cell membrane, the subject is still controversial. Studies regarding the genotoxic effects of EMF have generally focused on DNA damage. Possible mechanisms are related to ROS formation due to oxidative stress. EMF increases ROS production by enhancing the activity of nicotinamide adenine dinucleotide (NADH) oxidase in the cell membrane. Further detailed studies are needed to elucidate DNA damage mechanisms and apoptotic pathways during oogenesis and spermatogenesis in germ cells exposed to EMF.

(E) Yakymenko I, Burlaka A, Tsybulin I, Brieieva I, Buchynska L, Tsehmistrenko I, Chekhun F. Oxidative and mutagenic effects of low intensity GSM 1800 MHz microwave radiation. Exp Oncol. 40(4):282-287,2018. (VO, LE, GT, OX)

AIM: Despite a significant number of epidemiological studies on potential carcinogenicity of microwave radiation (MWR) from wireless devices and a bulk of experimental studies on oxidative and mutagenic effects of low intensity MWR, the discussion on potential carcinogenicity of low intensity MWR is going on. This study aims to assess oxidative and mutagenic effects of low intensity MWR from a typical commercial model of a modern smartphone. **MATERIALS AND METHODS:** The model of developing quail embryos has been used for the assessment of oxidative and mutagenic effects of Global System for Mobile communication (GSM) 1800 MHz MWR from a commercial model of smartphone. The embryos were exposed in ovo to 0.32 μW/cm2, discontinuously - 48 s - On, 12 s - Off, during 5 days before and 14 days through the incubation period. **RESULTS:** The exposure of quail embryos before and during the incubation period to low intensity GSM 1800 MHz has resulted in expressive statistically significant oxidative effects in embryonic cells, including a 2-fold increase in superoxide generation rate and 85% increase in nitrogen oxide generation rate, damages of DNA integrity and oxidative damages of DNA (up to twice increased levels of 8-oxo-dG in cells of 1-day old chicks from the exposed embryos). Finally, the exposure resulted in a significant, almost twice, increase of embryo mortality.
CONCLUSION: The exposure of model biological system to low intensity GSM 1800 MHz MWR resulted in significant oxidative and mutagenic effects in exposed cells, and thus should be recognized as a significant risk factor for living cells.

(E)Yan JG, Agresti M, Bruce T, Yan YH, Granlund A, Matloub HS. Effects of cellular phone emissions on sperm motility in rats. Fertil Steril.88(4):957-964, 2007. (CE, LE, GE)

OBJECTIVE: To evaluate the effects of cellular phone emissions on rat sperm cells. DESIGN: Classic experimental. SETTING: Animal research laboratory. SUBJECTS: Sixteen 3-month-old male Sprague-Dawley rats, weighing 250-300 g. INTERVENTION(S): Rats in the experimental group were exposed to two 3-hour periods of daily cellular phone emissions for 18 weeks; sperm samples were then collected for evaluation. MAIN OUTCOME MEASURE(S): Evaluation of sperm motility, sperm cell morphology, total sperm cell number, and mRNA levels for two cell surface adhesion proteins. RESULT(S): Rats exposed to 6 hours of daily cellular phone emissions for 18 weeks exhibited a significantly higher incidence of sperm cell death than control group rats through chi-squared analysis. In addition, abnormal clumping of sperm cells was present in rats exposed to cellular phone emissions and was not present in control group rats. CONCLUSION(S): These results suggest that carrying cell phones near reproductive organs could negatively affect male fertility.

(E) Yan JG, Agresti M, Zhang LL, Yan Y, Matloub HS. Upregulation of specific mRNA levels in rat brain after cell phone exposure. Electromagn Biol Med. 27(2):147-154, 2008. (VO, LE, GE)

Adult Sprague-Dawley rats were exposed to regular cell phones for 6 h per day for 126 days (18 weeks). RT-PCR was used to investigate the changes in levels of mRNA synthesis of several injury-associated proteins. Calcium ATPase, Neural Cell Adhesion Molecule, Neural Growth

Factor, and Vascular Endothelial Growth Factor were evaluated. The results showed statistically significant mRNA up-regulation of these proteins in the brains of rats exposed to cell phone radiation. These results indicate that relative chronic exposure to cell phone microwave radiation may result in cumulative injuries that could eventually lead to clinically significant neurological damage.

(E) Yang X-S, He G-L, Hao Y-T, Xiao Y, Chen C-H, Zhang G-B, Yu Z-P. Exposure to 2.45 GHz electromagnetic fields elicits an HSP-related stress response in rat hippocampus. Brain Res Bull 88(4):371-378, 2012. (VO, AE, GE)

The issue of possible neurobiological effects of the electromagnetic field (EMF) exposure is highly controversial. To determine whether electromagnetic field exposure could act as an environmental stimulus capable of producing stress responses, we employed the hippocampus, a sensitive target of electromagnetic radiation, to assess the changes in its stress-related gene and protein expression after EMF exposure. Adult male Sprague-Dawley rats with body restrained were exposed to a 2.45 GHz EMF at a specific absorption rate (SAR) of 6 W/kg or sham conditions. cDNA microarray was performed to examine the changes of gene expression involved in the biological effects of electromagnetic radiation. Of 2048 candidate genes, 23 upregulated and 18 downregulated genes were identified. Of these differential expression genes, two heat shock proteins (HSP), HSP27 and HSP70, are notable because expression levels of both proteins are increased in the rat hippocampus. Result from immunocytochemistry revealed that EMF caused intensive staining for HSP27 and HSP70 in the hippocampus, especially in the pyramidal neurons of cornu ammonis 3 (CA3) and granular cells of dentate gyrus (DG). The gene and protein expression profiles of HSP27 and HSP70 were further confirmed by reverse transcription polymerase chain reaction (RT-PCR) and Western blot. Our data provide direct evidence that exposure to electromagnetic fields elicits a stress response in the rat hippocampus.

(E) Yao K, Wang KJ, Sun ZH, Tan J, Xu W, Zhu LJ, Lu de Q. Low power microwave radiation inhibits the proliferation of rabbit lens epithelial cells by upregulating P27Kip1 expression. Mol Vis. 10:138-143, 2004. (VT, AE, GE)

PURPOSE: The goal of this study was to examine the effects of low power microwave radiation (<10 mW/cm2) on the proliferation of cultured rabbit lens epithelial cells (RLEC). METHODS: Cultured RLEC were exposed to continuous microwave radiation at a frequency of 2,450 MHz and power densities of 0.10, 0.25, 0.50, 1.00, and 2.00 mW/cm2 for 8 h. Cell morphologic changes were observed under a phase-contrast microscope. Cell viability was measured using the MTT assay and cell cycle analysis was measured using flow cytometry. After exposure to 2.00 mW/cm2 microwave radiation for 4, 6, and 8 h, the expression of cell cycle-regulatory proteins, P21WAF1 and P27Kip1, was examined using western blot analysis. Finally, the levels of P21WAF1 and P27Kip1 mRNA were analyzed by reverse transcription-polymerase chain reaction (RT-PCR). RESULTS: After 8 h of radiation treatment, cells treated with 0.50, 1.00, and 2.00 mW/cm2 microwave radiation exhibited decreased cell viability, increased cell condensation and an inhibition of DNA synthesis. RLEC showed significant G0/G1 arrest. No obvious changes could be detected in the 0.10 and 0.25 mW/cm2 microwave treatment groups.

Protein expression of P27Kip1 was markedly increased after microwave radiation. However, the mRNA levels were unchanged. On the other hand, there were no detectable differences in P21WAF1 protein expression and mRNA levels between microwave treatment and control groups. CONCLUSIONS: This study suggests that low power microwave radiation higher than 0.50 mW/cm2 can inhibit lens epithelial cell proliferation, and increase the expression of P27Kip1. These effects may account for the decline of lens epithelial proliferation after exposure to microwave radiation.

(E) Yao K, Wu W, Wang K, Ni S, Ye P, Yu Y, Ye J, Sun L. Electromagnetic noise inhibits radiofrequency radiation-induced DNA damage and reactive oxygen species increase in human lens epithelial cells. Mol Vis 14:964-969, 2008. (VT, AE, GT, IX, OX)

PURPOSE: The goal of this study was to investigate whether superposing of electromagnetic noise could block or attenuate DNA damage and intracellular reactive oxygen species (ROS) increase of cultured human lens epithelial cells (HLECs) induced by acute exposure to 1.8 GHz radiofrequency field (RF) of the Global System for Mobile Communications (GSM). METHODS: An sXc-1800 RF exposure system was used to produce a GSM signal at 1.8 GHz (217 Hz amplitude-modulated) with the specific absorption rate (SAR) of 1, 2, 3, and 4 W/kg. After 2 h of intermittent exposure, the ROS level was assessed by the fluorescent probe, 2',7'-dichlorodihydrofluorescein diacetate (DCFH-DA). DNA damage to HLECs was examined by alkaline comet assay and the phosphorylated form of histone variant H2AX (gammaH2AX) foci formation assay. RESULTS: After exposure to 1.8 GHz RF for 2 h, HLECs exhibited significant intracellular ROS increase in the 2, 3, and 4 W/kg groups. RF radiation at the SAR of 3 W/kg and 4 W/kg could induce significant DNA damage, examined by alkaline comet assay, which was used to detect mainly single strand breaks (SSBs), while no statistical difference in double strand breaks (DSBs), evaluated by gammaH2AX foci, was found between RF exposure (SAR: 3 and 4 W/kg) and sham exposure groups. When RF was superposed with 2 muT electromagnetic noise could block RF-induced ROS increase and DNA damage. CONCLUSIONS: DNA damage induced by 1.8 GHz radiofrequency field for 2 h, which was mainly SSBs, may be associated with the increased ROS production. Electromagnetic noise could block RF-induced ROS formation and DNA damage.

(E) Ye W, Wang F, Zhang W, Fang N, Zhao W, Wang J. Effect of Mobile Phone Radiation on Cardiovascular Development of Chick Embryo. Anat Histol Embryol. 45(3):197-208, 2016. (VO, LE, GT, DE)

The biological effects on cardiovascular development of chicken embryos were examined after radiation exposure using mobile phone (900 MHz; specific absorption rate˜1.07 W/kg) intermittently 3 h per day during incubation. Samples were selected by morphological and histological methods. The results showed the rate of embryonic mortality and cardiac deformity increased significantly in exposed group ($P < 0.05$). No any histological pathological changes were observed on Day 5-7 (D5-D7) of incubation. A higher distribution of lipid droplets was unexpectedly present in myocardial tissue from the exposure groups on D10-D13. Soon afterwards, myofilament disruption, atrioventricular valve focal necrosis, mitochondria vacuolization and atrial natriuretic peptide (ANP) decrease appeared on D15-D21 of incubation.

Comet assay data showed the haemocyte mean tail in the exposed group was significantly larger than that of the control ($P < 0.01$). The arterial vascular wall of exposed group was thicker ($P < 0.05$) than that of the control on D13, which was reversed to normal in later stages. Our findings suggest that long-term exposure of MPR may induce myocardium pathological changes, DNA damage and increased mortality; however, there was little effect on vascular development.

(NE) Yildirim MS, Yildirim A, Zamani AG, Okudan N. Effect of mobile phone station on micronucleus frequency and chromosomal aberrations in human blood cells. Genet Couns. 21(2):243-251, 2010. (HU, LE, GT)

The use of mobile telephones has rapidly increased worldwide as well as the number of mobile phone base stations that lead to rise low level radiofrequency emissions which may in turn have possible harm for human health. The national radiation protection board has published the known effects of radio waves exposure on humans living close to mobile phone base stations. However, several studies have claimed that the base station has detrimental effects on different tissues. In this study, we aimed to evaluate the effects of mobile phone base stations on the micronucleus (MN) frequency and chromosomal aberrations on blood in people who were living around mobile phone base stations and healthy controls. Frequency of MN and chromosomal aberrations in study and control groups was 8.96 +/- 3.51 and 6.97 +/- 1.52 (p: 0.16); 0.36 +/- 0.31 and 0.75 +/- 0.61 (p: 0.07), respectively. Our results show that there was not a significant difference of MN frequency and chromosomal aberrations between the two study groups. The results claim that cellular phones and their base stations do not produce important carcinogenic changes.

(E)Yu G, Zhu Y, Song C, Chen L, Tang Z, Wu T. The ZIP9-centered androgen pathway compensates for the 2605 MHz radiofrequency electromagnetic radiation-mediated reduction in resistance to H2O2 damage in Sertoli cells of adult rats. Ecotoxicol Environ Saf 254:114733, 2023. (VO, LE, GE)

The direct biological effects of radiofrequency electromagnetic radiation (RF-EMR) from wireless communication equipment on the testes are still unclear. Our previous study proved that long-term exposure to 2605 MHz RF-EMR gradually damage spermatogenesis and resulted in time-dependent reproductive toxicity by directly disrupting blood-testis barrier circulation. Although short-term exposure did not cause readily observable damage to fertility, whether it caused specific biological effects and how these effects contributed to the time-dependent reproductive toxicity of RF-EMR were currently unknown. Studies on this issue are important for elucidating the time-dependent reproductive toxicity of RF-EMR. The present study established a 2605 MHz RF-EMR (SAR=1.05 W/Kg) scrotal exposure model with rats and extracted primary Sertoli cells for exposure to investigate the direct biological effects of short-term RF-EMR exposure on the testis. The results showed that short-term RF-EMR exposure did not decrease sperm quality and spermatogenesis, but it increased the levels of testicular testosterone (T) and zinc transporter 9 (ZIP9) in Sertoli cells of rats. In vitro, 2605 MHz RF-EMR exposure did not increase the apoptosis rate of Sertoli cells, but it increased the apoptosis rate and MDA of Sertoli cells exposed to H2O2. T reversed these changes and increased ZIP9 level in Sertoli cells, whereas inhibiting ZIP9 expression significantly suppressed these T-

mediated protective effects. Moreover, T increased the levels of phosphorylated inositol-requiring enzyme 1 (P-IRE1), phosphorylated protein kinase R (PKR)-like endoplasmic reticulum kinase (P-PERK), phosphorylated eukaryotic initiation factor 2a (P-eIF2a) and phosphorylated activating transcription factor 6 (P-ATF6) in Sertoli cells, and these effects were reversed by ZIP9 inhibition. With prolonged exposure time, testicular ZIP9 was gradually downregulated, and testicular MDA increased. ZIP9 level was negatively correlated with MDA level in the testes of exposed rats. Thus, although short-term exposure to 2605 MHz RF-EMR (SAR=1.05 W/kg) did not significantly disturb spermatogenesis, it suppressed the ability of Sertoli cells to resist external insults, which was rescued by enhancing the ZIP9-centered androgen pathway in the short term. Increasing the unfolded protein response might be an important downstream mechanism involved. These results promote a better understanding of the time-dependent reproductive toxicity of 2605 MHz RF-EMR.

(E) Zalata, A., A. Z. El-Samanoudy, D. Shaalan, Y. El-Baiomy, and T. Mostafa. In vitro effect of cell phone radiation on motility, DNA fragmentation and clusterin gene expression of sperm. Int J Fertil Steril. 9(1):129-136, 2015. (VT, AE, GT, GE, RP)

Background: Use of cellular phones that emits radiofrequency electromagnetic field (RF-EMF) has been increased exponentially and became a part of everyday life. This study aimed to investigate the effects of RF-EMF radiation emitted from cellular phones on sperm motility variables, sperm DNA fragmentation and clusterin (CLU) gene expression. Materials and Methods: 124 semen samples were grouped into; normozoospermia (N, n=26), asthenozoospermia (A, n=32), asthenoteratozoospermia (AT, n=31) and oligoasthenoteratozoospermia (OAT, n=35). Semen samples were divided into two aliquots; samples not exposed to cell phone and samples exposed to cell phone radiation (850 MHz, maximum power < 1 watt; SAR 1.46 W/kg at 10 cm distance) for 1 hr. Before and immediately after exposure both aliquots were subjected to assessment of sperm motility, acrosin activity, sperm DNA fragmentation and CLU gene expression. Statistical differences were analyzed using paired t-student test for comparisons where P< 0.05 was set as significant. Results: There was significant decrease in sperm motility, sperm linear velocity, sperm linearity index, sperm acrosin activity and significant increase in sperm DNA fragmentation percent, CLU gene expression and CLU protein levels in the exposed semen samples to RF-EMF compared with non- exposed samples in OAT > AT > A > N groups (P<0.05). Conclusions: Cell phone emissions have a negative impact on exposed sperm motility indices, sperm acrosin activity, sperm DNA fragmentation and CLU gene expression especially in OAT cases.

(NE) Zeni, O., Schiavoni, A. S., Sannino, A., Antolini, A., Forigo, D., Bersani, F. and Scarfi, M. R. Lack of Genotoxic Effects (Micronucleus Induction) in Human Lymphocytes Exposed In Vitro to 900 MHz Electromagnetic Fields. Radiat. Res. 160, 152-158, 2003. (VT, AE, GT)

In the present study, we investigated the induction of genotoxic effects in human peripheral blood lymphocytes after exposure to electromagnetic fields used in mobile communication systems (frequency 900 MHz). For this purpose, the incidence of micronuclei was evaluated by applying the cytokinesis-block micronucleus assay. Cytotoxicity was also investigated using the cytokinesis-block proliferation index. The experiments were performed on peripheral blood

from 20 healthy donors, and several conditions were tested by varying the duration of exposure, the specific absorption rate (SAR), and the signal [continuous-wave (CW) or GSM (Global System of Mobile Communication) modulated signal]. The following exposures were carried out: (1) CW intermittent exposure (SAR = 1.6 W/kg) for 6 min followed by a 3-h pause (14 on/off cycles); (2) GSM signal, intermittent exposure as described in (1); (3) GSM signal, intermittent exposure as described in (1) 24 h before stimulation with phytohemagglutinin (8 on/off cycles); (4) GSM signal, intermittent exposure (SAR = 0.2 W/kg) 1 h per day for 3 days. The SARs were estimated numerically. No statistically significant differences were detected in any case in terms of either micronucleus frequency or cell cycle kinetics.

(NE) Zeni O, Romano M, Perrotta A, Lioi MB, Barbieri R, d'Ambrosio G, Massa R, Scarfi MR. Evaluation of genotoxic effects in human peripheral blood leukocytes following an acute in vitro exposure to 900 MHz radiofrequency fields. Bioelectromagnetics. 26(4):258-265, 2005. (VT, AE, GT)

Human peripheral blood leukocytes from healthy volunteers have been employed to investigate the induction of genotoxic effects following 2 h exposure to 900 MHz radiofrequency radiation. The GSM signal has been studied at specific absorption rates (SAR) of 0.3 and 1 W/kg. The exposures were carried out in a waveguide system under strictly controlled conditions of both dosimetry and temperature. The same temperature conditions (37.0 ± 0.1 degrees C) were realized in a second waveguide, employed to perform sham exposures. The induction of DNA damage was evaluated in leukocytes by applying the alkaline single cell gel electrophoresis (SCGE)/comet assay, while structural chromosome aberrations and sister chromatid exchanges were evaluated in lymphocytes stimulated with phytohemagglutinin. Alterations in kinetics of cell proliferation were determined by calculating the mitotic index. Positive controls were also provided by using methyl methanesulfonate (MMS) for comet assay and mitomycin-C (MMC), for chromosome aberration, or sister chromatid exchange tests. No statistically significant differences were detected in exposed samples in comparison with sham exposed ones for all the parameters investigated. On the contrary, the positive controls gave a statistically significant increase in DNA damage in all cases, as expected. Thus the results obtained in our experimental conditions do not support the hypothesis that 900 MHz radiofrequency field exposure induces DNA damage in human peripheral blood leukocytes in this range of SAR.

(NE) Zeni O, Gallaerano GP, Perrotta A, Romano, Sannino A, Sarti M, et al. Cytogenetic observations in human peripheral blood leukocytes following in vitro exposure to THz radiation: A pilot study. Health Phy. 92: 349-357, 2007. (VT, AE, GT)

Emerging technologies are considering the possible use of Terahertz radiation in different fields ranging from telecommunications to biology and biomedicine. The study of the potential effects of Terahertz radiation on biological systems is therefore an important issue in order to safely develop a variety of applications. This paper describes a pilot study devoted to determine if Terahertz radiation could induce genotoxic effects in human peripheral blood leukocytes. For this purpose, human whole blood samples from healthy donors were exposed for 20 min to Terahertz radiation. Since, to our knowledge, this is the first study devoted to the evaluation of

possible genotoxic effects of such radiation, different electromagnetic conditions were considered. In particular, the frequencies of 120 and 130 GHz were chosen: the first one was tested at a specific absorption rate (SAR) of 0.4 mW g-1, while the second one was tested at SAR levels of 0.24, 1.4, and 2 mW g-1. Chromosomal damage was evaluated by means of the cytokinesis block micronucleus technique, which also gives information on cell cycle kinetics. Moreover, human whole blood samples exposed to 130 GHz at SAR levels of 1.4 and 2 mW g-1 were also tested for primary DNA damage by applying the alkaline comet assay immediately after exposure. The results obtained indicate that THz exposure, in the explored electromagnetic conditions, is not able to induce either genotoxicity or alteration of cell cycle kinetics in human blood cells from healthy subjects.

(NE) Zeni O, Schiavoni A, Perrotta A, Forigo D, Deplano M, Scarfi MR. Evaluation of genotoxic effects in human leukocytes after in vitro exposure to 1950 MHz UMTS radiofrequency field. Bioelectromagnetics 29:177-184, 2008. (VT, AE, GT)

In the present study the third generation wireless technology of the Universal Mobile Telecommunication System (UMTS) signal was investigated for the induction of genotoxic effects in human leukocytes. Peripheral blood from six healthy donors was used and, for each donor, intermittent exposures (6 min RF on, 2 h RF off) at the frequency of 1950 MHz were conducted at a specific absorption rate of 2.2 W/kg. The exposures were performed in a transverse electro magnetic (TEM) cell hosted in an incubator under strictly controlled conditions of temperature and dosimetry. Following long duration intermittent RF exposures (from 24 to 68 h) in different stages of the cell cycle, micronucleus formation was evaluated by applying the cytokinesis block micronucleus assay, which also provides information on cell division kinetics. Primary DNA damage (strand breaks/alkali labile sites) was also investigated following 24 h of intermittent RF exposures, by applying the alkaline single cell gel electrophoresis (SCG)/comet assay. Positive controls were included by treating cell cultures with Mitomycin-C and methylmethanesulfonate for micronucleus and comet assays, respectively. The results obtained indicate that intermittent exposures of human lymphocytes in different stages of cell cycle do not induce either an increase in micronucleated cells, or change in cell cycle kinetics; moreover, 24 h intermittent exposures also fail to affect DNA structure of human leukocytes soon after the exposures, likely indicating that repairable DNA damage was not induced.

(E) Zeni O, Sannino A, Romeo S, Massaa R, Sarti M, Reddy AB, et al. Induction of an adaptive response in human blood lymphocytes exposed to radiofrequency fields: Influence of the universal mobile telecommunication system (UMTS) signal and the specific absorption rate. Mutat Res. 747: 29-35, 2012a. (VT, AE. GT, IX)

The induction of an adaptive response (AR) was examined in human peripheral blood lymphocytes exposed to non-ionizing radiofrequency fields (RF). Cells from nine healthy human volunteers were stimulated for 24h with phytohaemagglutinin and then exposed for 20h to an adaptive dose (AD) of a 1950MHz RF UMTS (universal mobile telecommunication system) signal used for mobile communications, at different specific absorption rates (SAR) of 1.25, 0.6, 0.3, and 0.15W/kg. This was followed by treatment of the cells at 48h with a challenge dose

(CD) of 100ng/ml mitomycin C (MMC). Lymphocytes were collected at the end of the 72h total culture period. The cytokinesis-block method was used to record the frequency of micronuclei (MN) as genotoxicity end-point. When lymphocytes from six donors were pre-exposed to RF at 0.3W/kg SAR and then treated with MMC, these cells showed a significant reduction in the frequency of MN, compared with the cells treated with MMC alone; this result is indicative of induction of AR. The results from our earlier study indicated that lymphocytes that were stimulated for 24h, exposed for 20h to a 900MHz RF GSM (global system for mobile communication) signal at 1.25W/kg SAR and then treated with 100ng/ml MMC, also exhibited AR. These overall data suggest that the induction of AR depends on RF frequency, type of the signal and SAR. Further characterization of RF-induced AR is in progress.

(NE) Zeni O, Sannino A, Sarti M, Romeo S, Massa R,Scarfi MR. Radiofrequency radiation at 1950 MHz (UMTS) does not affect key cellular endpoints in neuron-like PC12 Cells. Bioelectromagnetics. 33: 497-507, 2012b.(VT, AE, GT)

In this study, rat pheochromocytoma (PC12) cells were exposed, as a model of neuron-like cells, to 1950 MHz radiofrequency (RF) radiation with a signal used by the 3G wireless technology of the Universal Mobile Telecommunications System (UMTS) to assess possible adverse effects. RF exposure for 24 h at a specific absorption rate (SAR) of 10 W/kg was carried out in a waveguide system under accurately controlled environmental and dosimetric parameters. DNA integrity, cell viability, and apoptosis were investigated as cellular endpoints relevant for carcinogenesis and other diseases of the central nervous system. Very sensitive biological assays were employed to assess the effects immediately after RF exposure and 24 h later, as demonstrated by the cellular response elicited in PC12 cells using positive control treatments provided for each assay. In our experimental conditions, 24 h of RF exposure at a carrier frequency and modulation scheme typical of a UMTS signal was not able to elicit any effect in the selected cellular endpoints in undifferentiated PC12 cells, despite the application of a higher SAR value than those applied in the majority of the studies reported in the literature.

(E) Zeni O, Romeo S, Sannino A, Palumbo R, Scarfì MR. Evidence of bystander effect induced by radiofrequency radiation in a human neuroblastoma cell line. Environ Res 196:110935, 2021. (VT, AE, GT)

In previous studies we demonstrated that radiofrequency (RF) electromagnetic fields (EMF) is able to reduce DNA damage induced by a subsequent treatment with genotoxic agents, resembling the adaptive response, a phenomenon well known in radiobiology. In this study we report on the capability of the culture medium from SH-SY5Y neuroblastoma cells exposed to 1950 MHz to elicit, in recipient non-exposed cells, a reduction of menadione-induced DNA damage ($P < 0.05$; comet assay), indicating the capability of non-ionizing radiation to elicit a bystander effect. A comparable reduction was also detected in cultures directly exposed to the same EMF conditions ($P < 0.05$), confirming the adaptive response. In the same exposure conditions, we also evidenced an increase of heat shock protein 70 (hsp70) in culture medium of cells exposed to RF with respect to sham exposed ones ($P < 0.05$; western blot analysis), while no differences were detected in the intracellular content of hsp70. On the whole, our results evidence a protective effect of RF against menadione-induced DNA damage in directly and non-

directly exposed cells, and suggest hsp70 pathway to be investigated as one of the potential candidate underpinning the interaction between RF exposure and biological systems.

(NE) Zhadobov M, Sauleau R, Le Coq L, Debure L, Thouroude D, Michel D, Le Dréan Y. Low-power millimeter wave radiations do not alter stress-sensitive gene expression of chaperone proteins. Bioelectromagnetics 28(3):188-196, 2007. (VT, AE, GE)

This article reports experimental results on the influence of low-power millimeter wave (MMW) radiation at 60 GHz on a set of stress-sensitive gene expression of molecular chaperones, namely clusterin (CLU) and HSP70, in a human brain cell line. Selection of the exposure frequency is determined by its near-future applications for the new broadband civil wireless communication systems including wireless local area networks (WLAN) for domestic and professional uses. Frequencies around 60 GHz are strongly attenuated in the earth's atmosphere and such radiations represent a new environmental factor. An exposure system operating in V-band (50-75 GHz) was developed for cell exposure. U-251 MG glial cell line was sham-exposed or exposed to MMW radiation for different durations (1-33 h) and two different power densities (5.4 microW/cm(2) or 0.54 mW/cm(2)). As gene expression is a multiple-step process, we analyzed chaperone proteins induction at different levels. First, using luciferase reporter gene, we investigated potential effect of MMWs on the activation of transcription factors (TFs) and gene promoter activity. Next, using RT-PCR and Western blot assays, we verified whether MMW exposure could alter RNA accumulation, translation, or protein stability. Experimental data demonstrated the absence of significant modifications in gene transcription, mRNA, and protein amount for the considered stress-sensitive genes for the exposure durations and power densities investigated. The main results of this study suggest that low-power 60 GHz radiation does not modify stress-sensitive gene expression of chaperone proteins.

(E) Zhang DY, Xu ZP, Chiang H, Lu DQ, Zeng QL. [Effects of GSM 1800 MHz radiofrequency electromagnetic fields on DNA damage in Chinese hamster lung cells.] Zhonghua Yu Fang Yi XueZaZhi 40:149-152, 2006. [Article in Chinese] (VT, AE, GT)

OBJECTIVE: To study the effects of GSM 1800 MHz radiofrequency electromagnetic fields (RF EMF) on DNA damage in Chinese hamster lung (CHL) cells. METHODS: The cells were intermittently exposed or sham-exposed to GSM 1800 MHz RF EMF (5 minutes on/10 minutes off) at a special absorption rate (SAR) of 3.0 W/kg for 1 hour or 24 hours. Meanwhile, cells exposed to 2-acetaminofluorene, a DNA damage agent, at a final concentration of 20 mg/L for 2 hours were used as positive control. After exposure, cells were fixed by using 4% paraformaldehyde and processed for phosphorylated form of H2AX (gammaH2AX) immunofluorescence measurement. The primary antibody used for immunofluorescence was mouse monoclonal antibody against gammaH2AX and the secondary antibody was fluorescein isothiocyanate (FITC)-conjugated goat anti-mouse IgG. Nuclei were counterstained with 4, 6-diamidino-2-phenylindole (DAPI). The gammaH2AX foci and nuclei were visualized with an Olympus AX70 fluorescent microscope. Image Pro-Plus software was used to count the gammaH2AX foci in each cell. For each exposure condition, at least 50 cells were selected to detect gammaH2AX foci. Cells were classified as positive when more than five foci were detected. The percentage of gammaH2AX foci positive cells was adopted as the index of DNA damage. RESULTS: The percentage of gammaH2AX foci positive cell of 1800 MHz RF EMF

exposure for 24 hours (37.9 +/- 8.6)% or 2-acetylaminofluorene exposure (50.9 +/- 9.4)% was significantly higher compared with the sham-exposure (28.0 +/- 8.4)%. However, there was no significant difference between the sham-exposure and RF EMF exposure for 1 hour (31.8 +/- 8.7)%. CONCLUSION: 1800 MHz RF EMF (SAR, 3.0 W/kg) for 24 hours might induce DNA damage in CHL cells.

(E) Zhang MB, He JL, Jin LF, Lu DQ. Study of low-intensity 2450-MHz microwave exposure enhancing the genotoxic effects of mitomycin C using micronucleus test and comet assay in vitro. Biomed Environ Sci 15(4):283-290, 2002. (VT. AE, GT, IX)

OBJECTIVE: To determine the interaction between 2450-MHz microwaves (MW) radiation and mitomycin C (MMC). METHODS: The synergistic genotoxic effects of low-intensity 2450-MHz microwave and MMC on human lymphocytes were studied using single cell gel electrophoresis (SCGE) assay (comet assay) and cytokinesis-blocked micronucleus (CBMN) test in vitro. The whole blood cells from a male donor and a female donor were either only exposed to 2450-MHz microwaves (5.0 mW/cm2) for 2 h or only exposed to MMC (0.0125 microgram/mL, 0.025 microgram/mL and 0.1 microgram/mL) for 24 h; and the samples were exposed to MMC for 24 h after exposure to MW for 2 h. RESULTS: In the comet assay, the comet lengths (29.1 microns and 25.9 microns) of MW were not significantly longer than those (26.3 microns and 24.1 microns) of controls (P > 0.05). The comet lengths (57.4 microns, 68.9 microns, 91.4 microns, 150.6 microns, 71.7 microns, 100.1 microns, 145.1 microns) of 4 MMC groups were significantly longer than those of controls (P < 0.01). The comet lengths (59.1 microns, 92.3 microns, 124.5 microns, 182.7 microns and 57.4 microns, 85.5 microns, 137.5 microns, 178.3 microns) of 4 MW plus MMC groups were significantly longer than those of controls too (P < 0.01). The comet lengths of MW plus MMC groups were significantly longer than those of the corresponding MMC doses (P < 0.05 or P < 0.01) when the doses of MMC were > or = 0.025 microgram/mL. In the CBMN, the micronucleated cell (MNC) rates of MW were 5@1000 and 6@1000, which showed no difference compared with those (4@1000 and 4@1000) of controls (P > 0.05). The MNC rates of 4 MMC groups were 8@1000, 9@1000, 14@1000, 23@1000 and 8@1000, 8@1000, 16@1000, 30@1000 respectively. When the doses of MMC were > or = 0.05 microgram/mL, MNC rates of MMC were higher than those of controls (P < 0.05). MNC rates of 4 MW plus MMC groups were 12@1000, 13@1000, 20@1000, 32@1000 and 8@1000, 9@1000, 23@1000, 40@1000. When the doses of MMC were > or = 0.05 microgram/mL, MNC rates of MW plus MMC groups were much higher than those of controls (P < 0.01). MNC rates of 4 MW plus MMC groups were not significantly higher than those of the corresponding MMC doses. CONCLUSION: The low-intensity 2450-MHz microwave radiation cannot induce DNA and chromosome damage, but can increase DNA damage effect induced by MMC in comet assay.

(E) Zhang SZ, Yao GD, Lu DQ, Chiang H, Xu ZP. [Effect of 1.8 GHz radiofrequency electromagnetic fields on gene expression of rat neurons]. Zhonghua Lao Dong Wei Sheng Zhi Ye Bing Za Zhi. 26(8):449-452, 2008. [Article in Chinese] (VT, AE, GE, WS)

OBJECTIVE: To investigate the changes of gene expression in rat neuron induced by 1.8 GHz radiofrequency electromagnetic fields (RF EMF) to screen for RF EMF-responsive genes and the effect of different exposure times and modes on the gene expression in neuron. METHODS:

Total RNA was extracted immediately and purified from the primary culture of neurons after intermittent exposed or sham-exposed to a frequency of 1.8 GHz RF EMF for 24 hours at an average special absorption rate (SAR) of 2 W/kg. Affymetrix Rat Neurobiology U34 array was applied to investigate the changes of gene expression in rat neuron. Differentially expressed genes (Egr-1, Mbp and Plp) were further confirmed by semi-quantitative revere transcription polymerase chain reaction (RT PCR). The expression levels of Egr-1, Mbp and Plp were observed at different exposure times (6, 24 h) and modes (intermittent and continuous exposure). RESULTS: Among 1200 candidate genes, 24 up-regulated and 10 down-regulated genes were found by using Affymetrix microarray suite software 5.0 which are associated with multiple cellular functions (cytoskeleton, signal transduction pathway, metabolism, etc.) after functional classification. Under 24 h and 6 h intermittent exposure, Egr-1 and Plp in experiment groups showed statistic significance ($P < 0.05$) compared with the control groups, while expression of Mbp did not change significantly ($P > 0.05$). After 24 h continuous exposure, Egr-1 and Mbp in experiment groups showed statistic significance ($P < 0.05$) compared with the control group, while expression of Plp did not change significantly ($P > 0.05$). Under the same exposure mode 6 h, expression of all the 3 genes did not change significantly. Different times (6, 24 h) and modes (intermittent and continuous exposure) of exposure exerted remarkable different influences on the expression of Egr-1, Mbp, Plp genes ($P < 0.01$). CONCLUSION: The changes of many genes transcription were involved in the effect of 1.8 GHz RF EMF on rat neurons; Down-regulation of Egr-1 and up-regulation of Mbp, Plp indicated the negative effects of RF EMF on neurons; The effect of RF intermittent exposure on gene expression was more obvious than that of continuous exposure; The effect of 24 h RF exposure (both intermittent and continuous) on gene expression was more obvious than that of 6 h (both intermittent and continuous).

(E)Zhao L, Yao C, Wang H, Dong J, Zhang J, Xu X, Wang H, Yao B, Ren K, Sun L, Peng R. Immune Responses to Multi-Frequencies of 1.5 GHz and 4.3 GHz Microwave Exposure in Rats: Transcriptomic and Proteomic Analysis. Int J Mol Sci. 23(13):6949, 2022. (VT, AE. GE)

With the rapidly increasing application of microwave technologies, the anxiety and speculation about microwave induced potential health hazards has been attracting more and more attention. In our daily life, people are exposed to complex environments with multi-frequency microwaves, especially L band and C band microwaves, which are commonly used in communications. In this study, we exposed rats to 1.5 GHz (L10), 4.3 GHz (C10) or multi-frequency (LC10) microwaves at an average power density of 10 mW/cm^2. Both single and multi-frequency microwaves induced slight pathological changes in the thymus and spleen. Additionally, the white blood cells (WBCs) and lymphocytes in peripheral blood were decreased at 6 h and 7 d after exposure, suggesting immune suppressive responses were induced. Among lymphocytes, the B lymphocytes were increased while the T lymphocytes were decreased at 7 d after exposure in the C10 and LC10 groups, but not in the L10 group. Moreover, multi-frequency microwaves regulated the B and T lymphocytes more strongly than the C band microwave. The results of transcriptomics and proteomics showed that both single and multi-frequency microwaves regulated numerous genes associated with immune regulation and cellular metabolism in peripheral blood and in the spleen. However, multi-frequency microwaves altered the expression of many more genes and proteins. Moreover, multi-frequency microwaves down-regulated T lymphocytes' development, differentiation and activation-associated genes, while they up-

regulated B lymphocytes' activation-related genes. In conclusion, multi-frequency microwaves of 1.5 GHz and 4.3 GHz produced immune suppressive responses via regulating immune regulation and cellular metabolism-associated genes. Our findings provide meaningful information for exploring potential mechanisms underlying multi-frequency induced immune suppression.

(E) Zhao R, Zhang S, Xu Z, Ju L, Lu D, Yao G. Studying gene expression profile of rat neuron exposed to 1800 MHz radiofrequency electromagnetic fields with cDNA microassay. Toxicology 235:167-175, 2007. (VT, AE, GE)

A widespread use of mobile phone (MP) evokes a growing concern for their possible adverse effects on human, especially the brain. Gene expression is a unique way of characterizing how cells and organism adapt to changes in the external environment, so the aim of this investigation was to determine whether 1800 MHz radiofrequency electromagnetic fields (RF EMF) can influence the gene expression of neuron. Affymetrix Rat Neurobiology U34 array was applied to investigate the changes of gene expression in rat neuron after exposed to the pulsed RF EMF at a frequency of 1800 MHz modulated by 217 Hz which is commonly used in MP. Among 1200 candidate genes, 24 up-regulated genes and 10 down-regulated genes were identified after 24-h intermittent exposure at an average special absorption rate (SAR) of 2 W/kg, which are associated with multiple cellular functions (cytoskeleton, signal transduction pathway, metabolism, etc.) after functional classification. The results were further confirmed by quantitative real-time polymerase chain reaction (RT PCR). The present results indicated that the gene expression of rat neuron could be altered by exposure to RF EMF under our experimental conditions.

(E) Zhao TY, Zou SP, Knapp PE. Exposure to cell phone radiation up-regulates apoptosis genes in primary cultures of neurons and astrocytes. Neurosci Lett. 412(1):34-38, 2007. (VT, AE, GE, CS)

The health effects of cell phone radiation exposure are a growing public concern. This study investigated whether expression of genes related to cell death pathways are dysregulated in primary cultured neurons and astrocytes by exposure to a working Global System for Mobile Communication (GSM) cell phone rated at a frequency of 1900MHz. Primary cultures were exposed to cell phone emissions for 2h. We used array analysis and real-time RT-PCR to show up-regulation of caspase-2, caspase-6 and Asc (apoptosis associated speck-like protein containing a card) gene expression in neurons and astrocytes. Up-regulation occurred in both "on" and "stand-by" modes in neurons, but only in "on" mode in astrocytes. Additionally, astrocytes showed up-regulation of the Bax gene. The effects are specific since up-regulation was not seen for other genes associated with apoptosis, such as caspase-9 in either neurons or astrocytes, or Bax in neurons. The results show that even relatively short-term exposure to cell phone radiofrequency emissions can up-regulate elements of apoptotic pathways in cells derived from the brain, and that neurons appear to be more sensitive to this effect than astrocytes.

(E)Zhao Z-g, Zhang J-f, Yan P, Ma Y-m. [Relationship between millimeter wave
(NE) Zhijian C, Xiaoxue L, Yezhen L, Deqiang L, Shijie C, Lifen J, Jianlin L, Jiliang H. Influence of 1.8-GHz (GSM) radiofrequency radiation (RFR) on DNA damage and repair

induced by X-rays in human leukocytes in vitro. Mutat Res. 677(1-2):100-104, 2009. (VT, AE, GT, IX)

In the present study, the in vitro comet assay was used to determine whether 1.8-GHz radiofrequency radiation (RFR) can influence DNA repair in human leukocytes exposed to X-rays. The specific energy absorption rate (SAR) of 2 W/kg (the current European safety limit) was applied. The leukocytes from four young healthy donors were intermittently exposed to RFR for 24 h (fields on for 5 min, fields off for 10 min), and then irradiated with X-rays at doses of 0.25, 0.5, 1.0 and 2.0 Gy. DNA damage to human leukocytes was detected using the comet assay at 0, 15, 45, 90, 150 and 240 min after exposure to X-rays. Using the comet assay, the percent of DNA in the tail (% tail DNA) served as the indicator of DNA damage; the DNA repair percentage (DRP) served as the indicator of the DNA repair speed. The results demonstrated that (1) the DNA repair speeds of human leukocytes after X-ray exposure exhibited individual differences among the four donors; (2) the intermittent exposures of 1.8-GHz RFR at the SAR of 2 W/kg for 24 h did not directly induce DNA damage or exhibit synergistic effects with X-rays on human leukocytes.

(NE) Zhijian C, Xiaoxue L, Yezhen L, Shijie C, Lifen J, Jianlin L, Deqiang L, Jiliang H. Impact of 1.8-GHz radiofrequency radiation (RFR) on DNA damage and repair induced by doxorubicin in human B-cell lymphoblastoid cells. Mutat Res. 695(1-2):16-21, 2010. (VT, AE, GT, IX)

In the present in vitro study, a comet assay was used to determine whether 1.8-GHz radiofrequency radiation (RFR, SAR of 2W/kg) can influence DNA repair in human B-cell lymphoblastoid cells exposed to doxorubicin (DOX) at the doses of 0microg/ml, 0.05microg/ml, 0.075microg/ml, 0.10microg/ml, 0.15microg/ml and 0.20microg/ml. The combinative exposures to RFR with DOX were divided into five categories. DNA damage was detected at 0h, 6h, 12h, 18h and 24h after exposure to DOX via the comet assay, and the percent of DNA in the tail (% tail DNA) served as the indicator of DNA damage. The results demonstrated that (1) RFR could not directly induce DNA damage of human B-cell lymphoblastoid cells; (2) DOX could significantly induce DNA damage of human B-cell lymphoblastoid cells with the dose-effect relationship, and there were special repair characteristics of DNA damage induced by DOX; (3) E-E-E type (exposure to RFR for 2h, then simultaneous exposure to RFR and DOX, and exposure to RFR for 6h, 12h, 18h and 24h after exposure to DOX) combinative exposure could obviously influence DNA repair at 6h and 12h after exposure to DOX for four DOX doses (0.075microg/ml, 0.10microg/ml, 0.15microg/ml and 0.20microg/ml) in human B-cell lymphoblastoid cells.

(NE) Ziemann C, Brockmeyer H, Reddy SB, Vijayalaxmi, Prihoda TJ, Kuster N, Tillmann T, Dasenbrock C. Absence of genotoxic potential of 902 MHz (GSM) and 1747 MHz (DCS) wireless communication signals: In vivo two-year bioassay in B6C3F1 mice. Int J Radiat Biol. 85(5):454-464, 2009. (VO, LE, GT)

PURPOSE: The aim of the present investigation was to determine the incidence of micronuclei in peripheral blood erythrocytes of B6C3F1 mice that had been chronically exposed to radiofrequencies (RF) used for mobile communication. MATERIALS AND METHODS: 'Ferris

wheels' were used to expose tube-restrained male and female mice to simulated environmental RF signals of the Global System for Mobile Communications (GSM, 902 MHz) or Digital Cellular System (DCS, 1747 MHz). RF signals were applied to the mice for 2 hours/day on 5 days/week for two years, at maximal whole-body-averaged specific absorption rates of 0.4, 1.3, and 4.0 W/kg body weight. Concurrent sham-exposed mice, cage controls, and positive controls injected with mitomycin C were included in this investigation. At necropsy, peripheral blood smears were prepared, and coded slides were stained using May-Grunwald-Giemsa or acridine orange. The incidence of micronuclei was recorded for each mouse in 2000 polychromatic and 2000 normochromatic erythrocytes. RESULTS: There were no significant differences in the frequency of micronuclei between RF-exposed, sham-exposed, and cage control mice, irrespective of the staining/counting method used. Micronuclei were, however, significantly increased in polychromatic erythrocytes of the positive control mice. CONCLUSIONS: In conclusion, the data did not indicate RF-induced genotoxicity in mice after two years of exposure.

(E) Zong C, Ji Y, He Q, Zhu S, Qin F, Tong J, et al. Adaptive response in mice exposed to 900 MHz radiofrequency fields: Bleomycin-induced DNA and oxidative damage/repair. Int J Radiat Biol. 91: 270-276, 2015. (VO, LE, GT, IX, OX, LI)

Purpose: To determine whether mice exposed to radiofrequency fields (RF) and then injected with a radiomimetic drug, bleomycin (BLM), exhibit adaptive response and provide some mechanistic evidence for such response. **Materials and methods:** Adult mice were exposed to 900 MHz RF at 120 μW/cm(2) power density for 4 hours/day for 7 days. Immediately after the last exposure, some mice were sacrificed while the others were injected with BLM 4 h later. In each animal: (i) The primary DNA damage and BLM-induced damage as well as its repair kinetics were determined in blood leukocytes; and (ii) the oxidative damage was determined from malondialdehyde (MDA) levels and the antioxidant status was assessed from superoxide dismutase (SOD) levels in plasma, liver and lung tissues. **Results:** There were no indications for increased DNA and oxidative damages in mice exposed to RF alone in contrast to those treated with BLM alone. Mice exposed to RF+ BLM showed significantly: (a) reduced BLM-induced DNA damage and that remained after each 30, 60, 90, 120 and 150 min repair time, and (b) decreased levels of MDA in plasma and liver, and increased SOD level in the lung. **Conclusions:** The overall data suggested that RF exposure was capable of inducing adaptive response and mitigated BLM- induced DNA and oxidative damages by activating certain cellular processes.

(E) Zothansiama, Zosangzuali M, Lalramdinpuii M, Jagetia GC. Impact of radiofrequency radiation on DNA damage and antioxidants in peripheral blood lymphocytes of humans residing in the vicinity of mobile phone base stations. Electromagn Biol Med. 36(3):295-305, 2017. (HU, LE, GT, OX)

Radiofrequency radiations (RFRs) emitted by mobile phone base stations have raised concerns on its adverse impact on humans residing in the vicinity of mobile phone base stations. Therefore, the present study was envisaged to evaluate the effect of RFR on the DNA damage and antioxidant status in cultured human peripheral blood

lymphocytes (HPBLs) of individuals residing in the vicinity of mobile phone base stations and comparing it with healthy controls. The study groups matched for various demographic data including age, gender, dietary pattern, smoking habit, alcohol consumption, duration of mobile phone use and average daily mobile phone use. The RF power density of the exposed individuals was significantly higher ($p < 0.0001$) when compared to the control group. The HPBLs were cultured and the DNA damage was assessed by cytokinesis blocked micronucleus (MN) assay in the binucleate lymphocytes. The analyses of data from the exposed group (n = 40), residing within a perimeter of 80 m of mobile base stations, showed significantly ($p < 0.0001$) higher frequency of micronuclei when compared to the control group, residing 300 m away from the mobile base station/s. The analysis of various antioxidants in the plasma of exposed individuals revealed a significant attrition in glutathione (GSH) concentration ($p < 0.01$), activities of catalase (CAT) ($p < 0.001$) and superoxide dismutase (SOD) ($p < 0.001$) and rise in lipid peroxidation (LOO) when compared to controls. Multiple linear regression analyses revealed a significant association among reduced GSH concentration ($p < 0.05$), CAT ($p < 0.001$) and SOD ($p < 0.001$) activities and elevated MN frequency ($p < 0.001$) and LOO ($p < 0.001$) with increasing RF power density.

(E) Zotti-Martelli L, Peccatori M, Scarpato R, Migliore L, Induction of micronuclei in human lymphocytes exposed in vitro to microwave radiation. *Mutat Res* 472(1-2):51-58, 2000. (VT, AE, GT)

Increasing applications of electromagnetic fields are of great concern with regard to public health. Several in vitro studies have been conducted to detect effects of microwave exposure on the genetic material leading to negative or questionable results. The micronucleus (MN) assay which is proved to be a useful tool for the detection of radiation exposure-induced cytogenetic damage was used in the present study to investigate the genotoxic effect of microwaves in human peripheral blood lymphocytes in vitro exposed in G(0) to electromagnetic fields with different frequencies (2.45 and 7.7GHz) and power density (10, 20 and 30mW/cm(2)) for three times (15, 30 and 60min). The results showed for both radiation frequencies an induction of micronuclei as compared to the control cultures at a power density of 30mW/cm(2) and after an exposure of 30 and 60min. Our study would indicate that microwaves are able to cause cytogenetic damage in human lymphocytes mainly for both high power density and long exposure time.

(E) Zotti-Martelli L, Peccatori M, Maggini V, Ballardin M, Barale R. Individual responsiveness to induction of micronuclei in human lymphocytes after exposure in vitro to 1800-MHz microwave radiation. Mutat Res. 582(1-2):42-52, 2005. (VT, AE, GT)

The widespread application of microwaves is of great concern in view of possible consequences for human health. Many in vitro studies have been carried out to detect possible effects on DNA and chromatin structure following exposure to microwave radiation. The aim of this study is to assess the capability of microwaves, at different power densities and exposure times, to induce genotoxic effects as evaluated by the in vitro micronucleus (MN) assay on peripheral blood lymphocytes from nine different healthy donors, and to investigate also the possible inter-individual response variability. Whole blood samples were exposed for 60, 120 and 180

min to continuous microwave radiation with a frequency of 1800 MHz and power densities of 5, 10 and 20 mW/cm(2). Reproducibility was tested by repeating the experiment 3 months later. Multivariate analysis showed that lymphocyte proliferation indices were significantly different among donors ($p<0.004$) and between experiments ($p<0.01$), whereas the applied power density and the exposure time did not have any effect on them. Both spontaneous and induced MN frequencies varied in a highly significant way among donors ($p<0.009$) and between experiments ($p<0.002$), and a statistically significant increase of MN, although rather low, was observed dependent on exposure time ($p=0.0004$) and applied power density ($p=0.0166$). A considerable decrease in spontaneous and induced MN frequencies was measured in the second experiment. The results show that microwaves are able to induce MN in short-time exposures to medium power density fields. Our data analysis highlights a wide inter-individual variability in the response, which was confirmed to be a characteristic reproducible trait by means of the second experiment.

(NE) Zuo WQ, Hu YJ, Yang Y, Zhao XY, Zhang YY, Kong W, Kong WJ. Sensitivity of spiral ganglion neurons to damage caused by mobile phone electromagnetic radiation will increase in lipopolysaccharide-induced inflammation in vitro model. J Neuroinflammation. 12(1):105,2015. (VT, AE, GT, IX)

BACKGROUND: With the increasing popularity of mobile phones, the potential hazards of radiofrequency electromagnetic radiation (RF-EMR) on the auditory system remain unclear. Apart from RF-EMR, humans are also exposed to various physical and chemical factors. We established a lipopolysaccharide (LPS)-induced inflammation in vitro model to investigate whether the possible sensitivity of spiral ganglion neurons to damage caused by mobile phone electromagnetic radiation (at specific absorption rates: 2, 4 W/kg) will increase. METHODS: Spiral ganglion neurons (SGN) were obtained from neonatal (1- to 3-day-old) Sprague Dawley® (SD) rats. After the SGN were treated with different concentrations (0, 20, 40, 50, 100, 200, and 400 μg/ml) of LPS, the Cell Counting Kit-8 (CCK-8) and alkaline comet assay were used to quantify cellular activity and DNA damage, respectively. The SGN were treated with the moderate LPS concentrations before RF-EMR exposure. After 24 h intermittent exposure at an absorption rate of 2 and 4 W/kg, DNA damage was examined by alkaline comet assay, ultrastructure changes were detected by transmission electron microscopy, and expression of the autophagy markers LC3-II and Beclin1 were examined by immunofluorescence and confocal laser scanning microscopy. Reactive oxygen species (ROS) production was quantified by the dichlorofluorescin-diacetate assay. RESULTS: LPS (100 μg/ml) induced DNA damage and suppressed cellular activity ($P < 0.05$). LPS (40 μg/ml) did not exhibit cellular activity changes or DNA damage ($P > 0.05$); therefore, 40 μg/ml was used to pretreat the concentration before exposure to RF-EMR. RF-EMR could not directly induce DNA damage. However, the 4 W/kg combined with LPS (40 μg/ml) group showed mitochondria vacuoles, karyopyknosis, presence of lysosomes and autophagosome, and increasing expression of LC3-II and Beclin1. The ROS values significantly increased in the 4 W/kg exposure, 4 W/kg combined with LPS (40 μg/ml) exposure, and H_2O_2 groups ($P < 0.05$, 0.01). CONCLUSIONS: Short-term exposure to radiofrequency electromagnetic radiation could not directly induce DNA damage in normal spiral ganglion neurons, but it could cause the changes of cellular ultrastructure at special SAR

4.0 W/kg when cells are in fragile or micro-damaged condition. It seems that the sensitivity of SGN to damage caused by mobile phone electromagnetic radiation will increase in a lipopolysaccharide-induced inflammation in vitro model.

Genetic effects of radiofrequency electromagnetic radiation

authors in blue = study involved exposure to wireless devices. LI= effects observed at low SAR (< 0.4 W/kg); GE = gene expression study; * = no effect

	Exposure conditions	Results
*Agarwal et al. (2009)	Human semen sample to mobile phone radiation in talk mode for 1 h	No significant DNA damage, increase in reactive oxygen species; decrease in sperm motility and viability.
Aitken et al. (2005)	Mice to 900-MHz RFR for 7 days at 12 h/day; SAR 0.09 W/kg	Significant damage to Mitochondrial genome and nuclear β-globin locus in epididymal spermatozoa.
Akdag et al. (2016) (LI)	Male Wistar-Albino rats to 2400 MHz RFR from a Wi-Fi signal generator for a year; SAR 0.000141 (min)- 0.007127 (max) W/kg	No significant change in DNA single strand breaks (Comet assay) in brain, kidney, liver, and skin tissues, increased in testes.
Akdag et al. (2018)	Men who used mobile phone for different durations per day; peak head SAR 0.45-0.79 W/kg	Increased DNA single strand breaks (Comet assay) in ear canal hair follicle cells; a dose-response relationship was observed.
Akhavan-Sigari et al. (2014) (GE)	Resected Glioblastoma multiforme (GBM) from human subjects	Increased mutant type of p53 expression in the peripheral zone of GBM in patient who use cell phone form ≥3h/day; the increase was significantly correlated with shorter overall survival time.
Albaqami et al. (2020) (GE)	Arabidopsis (rockcress) seedlings exposed to 7 MHz RFR, 2 μT (rms); 3 h under blue light activation	Increased expression of cryptochrome-regulated genes (IAA, PIN1, and PIN3)
Alkis et al. (2019a) (LI)	Rats exposed to 900 MHz (brain SAR 0.0845 W/kg), 1800 MHz (0.04563 W/kg), and 2100 MHz (0.03957 W/kg) RFR 2 h/day for 6 months	Increased DNA single strand break (Comet assay), oxidative DNA damage, and oxidative stress in brain frontal lobe.
Alkis et al. (2019b) (LI)	Rats exposed to 900 MHz, 1800 MHz, and 2100 MHz RFR 2 h/day	Increased DNA single strand beak (Comet assay), oxidative DNA damage and oxidative stress in testicular tissue.

	for 6 months; maximum SAR over the rat 0.017 W/kg	
Alkis et al. (2021)	Liver of rats exposed to 1800 MHz (0.62 W/kg) or 2100 MHz (0.2 W/kg) RFR 2 h/day for7 months.	Increased 8-hydroxydeoxyguanosine (8-OHdG) (Comet assay)
*Al-Serori et al. (2017)	Human U87 (wild-type) and U251 (mutated) glioblastoma cells exposed to intermittent (5 min ON/10 min OFF) UMTS 1750 MHz signal for 16 h, SAR 0.25, 0.5, and 1 W/kg	No effect on micronucleus frequency. Apoptosis was induced in U231 cells.
Al-Serori et al. (2018)	Ten human cell types exposed to intermittent (5 min ON/10 min OFF) UMTS 1750 MHz signal for 16 h, SAR 0.25, 0.5, and 1 W/kg	Increased in single strand breaks (Comet assay) in U87 p52- proficient glioblastoma cells grew under serum free condition; no effect on double strand breaks (gH2AX foci); nucleotide excision repair induced.
*Antonopoulos et al. (1997)	Human blood samples exposed to 380 MHz (17.65 Hz modulation, 0.08 W/kg); 900 MHZ (217 Hz modulation, 0.208 W/kg); or 1700 MHz (217 Hz modulation, 1.7 W/kg) for 48-68 h	No significant effect on cell cycle progression and frequency of sister-chromatin exchange in lymphocytes.
Aravinda et al. (2022)	Buccal mucosa cells of children used mobile phone 1-2 h, 3-6 h, or >6 h per day	Children with >6 h /day usage showed chromosomal aberrations
Atasoy et al. (2013) (LI)	Male Wister rats exposed to 2437 MHz (Wi-Fi) RFR; 24 h/day for 20 weeks; maximum SAR 0.091 W/kg	Increased oxidative DNA damage and decreased catalase and glutathione activities in blood and testes.
Atlı Şekeroğlu et al (2013)	Immature (whole body SAR 0.38-0.78 W/kg) and mature (0.31-0.52 W/kg)	Increased bone marrow cell chromosome aberration, micronucleus frequency, mitotic index and ratio of polychromatic erythrocytes.

	rats exposed to 900 MHz RFR 2 h/day for 45 days	Cytogenetic damage in immature rats was significantly higher than in the mature rats. No recovery on day 15 post-exposure.
Avendaño et al. (2012)	Human sperm exposed to an internet-connected laptop for 4 h	Increased DNA fragmentation and motility.
Azimipour et al. (2020) (GE)	Pre-antral ovary follicles of mouse exposed to radiation from a Sony Ericsson K800 for 1 h (no dosimetry data available)	Increased the matrix metalloproteinases MMP-2 gene and decreased MMP-9 gene expression; decreased expression of tissue inhibitors of metalloproteinases (TIMPs) genes, *TIMP-1* and -2
Balakrishnan et al. (2014) (GE)	Serum of mobile phone frequent and infrequent users	Increased HSP70 gene expression with phone use
Balode (1996)	Blood samples from female Latvian Brown cows lived close to and in front of the Skrundra Radar and from a control area	Significantly higher micronucleus concentration was found in the erythrocytes of the exposed cows.
Banerjee et al. (2016)	Buccal mucosal cells from subjects who used their mobile phone less than five years and less than three hours a week (low), and those who used more than five years and more than 10 hours a week comprised of the second group.	Micronucleus frequency in buccal mucosal cells was found to be significantly increased in longer cellular phone users.
Baohong et al. (2005)	Human lymphocytes exposed in vitro to 1800 MHz RFR (SAR 3 W/kg) for two hours and also co-treated with various mutagens	DNA strand break assayed (Comet assay) at 0 and 21 h after treatment. No effect when cells were exposed to RFR alone. RFR co-exposure enhanced the DMA damage induced by mitomycin C and 4-nitroquinoline-1-oxide.
Baohong et al. (2007)	Human lymphocytes exposed in vitro to 1800 MHz RFR (SAR 3 W/kg) for 0. 1.5, and 4 h. Cells were also co-treated with ultraviolet ray C	DNA damage as assayed by the Comet assay showed no significant effect with RFR alone. RFR co-exposure reduced DNA damage induced by ultraviolet C.
Banerjee et al. (2016)	Buccal mucosal cells of	Increased micronucleated cells in high users

	low (less than five years and less than three hours a week) and high (more than five years and more than 10 hours a week) mobile phone users	
Beaubois et al. (2007) (GE)	tomato plants (Lycopersicon esculentum) exposed 90 MHz RFR at 5 V/m for 10 min	Increased wound-inducible transcript bZIP
Bektas et al (2020)	Pregnant women who used cell phone and Wi-Fi; placenta and cord blood samples were analyzed	Samples from cell phone users showed increased oxidative DNA damage and oxidative stress; Wi-Fi users showed increased oxidative DNA damage but no oxidative stress; more DNA single strand breaks (Comet assay) in cell phone users than in control (did not use cell phone nor WiFI) and Wi-Fi users; Wi-Fi and cell phone uses were synergistic.
Belyaev et al. (1992)	X-irradiated E. coli cells exposed to 51.62-51.84 GHz and 41.25-41.50 GHz millimeter-wave RFR	Power density of 1 mW/cm^2 was sufficient to suppress X-radiation-induced repair of genome conformational state.
Belyaev et al. (2005) (LI)	Lymphocytes from human subjects exposed to GSM 915 MHz RFR for 2 h ; SAR 0.037 W/kg;	Increased condensation of chromatin; no significant difference between responses of blood samples of healthy and electro-hypersensitive subjects.
Belyaev et al. (2006) (LI)	Rats exposed to GSM 915 MHz RFR for 2 h, SAR 0.4 W/kg	Affected gene expression in brain cells; no significant effect on chromatin conformation and double strand DNA breaks.
Belyaev et al. (2009) (LI)	Human lymphocytes exposed to UMTS cell phone signal (1947.4 MHz, 5 MHz band width) for 1 h; SAR 0.04 W/kg	Chromatin affected and inhibition of DNA double-strand break co-localizing 53BPI/gamma-H2AX DNA repair foci; lymphocytes from electro-hypersensitive subjects responded differently to UMTS and GSM signals in the formation of DNA repair foci than in healthy subjects.
*Bisht et al. (2002)	Mouse embryo sarcoma fibroblast C3H 10T½ cells exposed to FDMA (835.62 MHz; SAR 3.2 or 5.1 W/kg) and CDMA	No significant effect on micronucleus formation.

	(847.74 MHz; SAR 3.2 or 4.8 W/kg) RFR for 3, 8, 16 or 24h	
Bourdineaud et al. (2017) (LI) (GE)	Eisenia fetida earthworms exposed to 900 MHz for 2 h; SAR 0.00013-0.00933 W/kg	DNA genotoxic effect persisted for at least 24 h; gene expressions up regulated for HSP70 (heat shock protein), MEKKI (signal transduction); oxidative stress; and chemical and immune defenses.
*Bourthoumieu et al. (2010)	Human amniotic cells exposed to GSM-900 MHz RFR for 24 h; SAR 0.25 W/kg	No significant genotoxic effect was observed at 0 and 24 h after exposure by visual examination of chromosomal rearrangement.
*Bourthoumieu et al. (2011)	Human amniotic cells exposed to GSM-900 MHz RFR for 24 h; SAR 0.25, 1,2, and 4 W/kg	No significant change in the rate of aneuploidy of chromosomes 11 and 17 was found.
*Bourthoumieu et al. (2013) (GE)	Human amniotic cells exposed to GSM-900 MHz RFR for 24 h; SAR 0.25, 1,2, and 4 W/kg	No significant change in the expression and activation of the p53 protein was found. (p53 can cause cell cycle arrest and allow time for DNA repair or apoptosis.)
Brech et al. (2019)	Canine and human blood exposed to 123.9 KHz and 250.8 KHz RFR at 0.79 or 0.1 mT for 1-5 h or 20-24 h	Increased DNA single strand breaks (Comet assay) after 20 h of exposure.
Burlaka et al. (2013)	Male Wister rats exposed to 2450 MHz RFR for 2 h a day. 7 days a week for 2, 8, 15, or 30 days at 5-10 mW/cm^2.	Increased micronucleus formation was found in bone marrow erythropoietic cells after 15-day exposure; erythrocyte count, haemoglobin and haematocrit were increased in peripheral blood after 8 and 15 days of exposure.
Busljeta et al. (2004)	Bone marrow from rats exposed to 2450 MHz RFR at 5-10 mW/cm^2, 2 h/day up to 30 days.	Increased micronucleated cells
Buttiglione et al. (2007) (GE)	Human SH-SY5Y neuroblastoma cells exposed to modulated 900 MHz RFR for 24 h; SAR 1 W/kg	Increased Egr-1 gene expression paralleled with activation of the MAPK subtypes ERK1/2 and SAPK/JNK, and decrease in mRNA of Bcl-2 and surviving genes. RFR has anti-proliferative effect and causes cell cycle arrest at G2-M.
Cam and Seyhan (2012)	Hair root cells of human subjects after 15-30 min use of a 900-MHz GSM	Increased in DNA single strand breaks (Comet assay) was observed; more damages resulted after 30 min than after 15 min use.

	cell phone	
Campisi et al. (2010)	Rat neocortical astroglial to 50 Hz-modulated or CW 900 MHz RFR for 5, 10, or 20 min; incident power density 0.0265 mW/cm^2	Significant increases in DNA fragmentation and reactive oxygen species were observed at 20 min only after exposure to the modulated RFR.
Cantu et al. (2023) (GE) (LI)	Human keratinocytes exposed to 900 MHz RFR for 1 h at <0.01 W/kg	Six common DNA targets that were both differentially methylated and differentially expressed
Cappucci et al. (2022) (GE) (LI)	Head, ovary, and testis of Drosophila melanogaster exposed to 2437 MHz RFR (WiFi) fom embryo to adult stage at 0.0608 W/kg	Increased gene expression of hsp70, induced gene instability, and increased DNA damage.
Cervellati et al. (2013) (GE)	Human placenta trophoblast-derived HTR-8/SVneo cells exposed to 1.8 GHz GSM RFR amplitude modulated by rectangular pulses of 217 Hz for 1 h; SAR 2 W/kg	Increased connexin Cx40 and Cx43 mRNA expression; decreased Integrin alpha1 and β 1 mRNA levels but enhanced Int alpha5 mRNA expression.
Chandel et al. (2019a) (LI)	Onion roots (Allium cepa L.) exposed to 2350 MHz RFR for 1, 2, or 4 h, SAR 0.313 W/kg	Increased in mitotic index and chromosomal aberration; significant increase in DNA single strand breaks (Comet assay) at 2 and 4 h.
Chandel et al. (2019b) (LI)	Onion roots (Allium cepa L.) exposed to 2100 MHz RFR for 1 or 4 h, SAR 0.282 W/kg	Increased mitotic index, chromosomal aberration, and DNA single-strand breaks (Comet assay) after 4 h of exposure.
*Chang et al. (2005)	Escherichia coli and Salmonella typhimurium exposed to 835 MHz RFR for 48h; SAR 4 W/kg	835-MHz RFR neither affected the reverse mutation frequency nor accelerated DNA degradation in vitro. (Some interaction effects with mutagens were observed.)
Chaturvedi et al. (2011) (LI)	Male mice exposed to 2450 MHz RFR, 2 h/day for 30 days; SAR 0.03561 W/kg	Increased DNA single strand breaks (Comet assay) in brain cells.
*Chauhan et al. (2006a) (GE)	Human lymphoblastoma cells (TK6) exposed to pulsed-modulated,	No evidence of a general stress response with proto-oncogene and heat-shock protein gene transcriptions.

	intermittent (5 min ON, 10 min OFF) 1900-MHz RFR for 6 h; SAR 1 or 6 W/kg	
*Chauhan et al. (2006b)	Human –derived immune cell-lines HL-60 and MM6 cells exposed to pulsed-modulated, intermittent (5 min ON, 10 min OFF) 1900-MHz RFR for 6 h; SAR 1 or 10 W/kg	No evidence of detectable change in stress-related gene expression.
*Chauhan et al. (2007) (GE)	Human glioblastoma-derived cell-line (U87MG) and human monocyte-derived cell-line (MM6) exposed to pulsed-modulated, intermittent (5 min ON, 10 min OFF) 1900-MHz RFR for 24 and 6 h; SAR 0.1-10 W/kg	No evidence that the RFR exposure altered late onset gene expression in either cultured cell-lines.
Chavdoula et al. (2010)	Drosophila melanogaster flies exposed to GSM-900 MHz and DCS-1800 MHz cell phone radiation; 6 min per day for 5 days	Decreased insect's reproductive capacity with fragmented DNA (apoptosis) in the egg chamber.
*Chemeris et al. (2004)	Frog (Xenopus laevis) erythrocytes exposed to high peak power pulsed RFR (8.8 GHz, 180 ns pulse width, peak power 65 kW, repetition rate 50 Hz) for 40 min; SAR 1.6 kW/kg (peak SAR 300 MW/kg)	Increased DNA single strand breaks (Comet assay) was caused by temperature rise.
*Chemeris et al. (2006)	human whole-blood leukocytes and isolated lymphocytes exposed to 8.8 GHz RFR (180 ns pulse width, 50 Hz pulse repetition frequency) at 1.6 kW/kg (peak 300 MW/kg) for 40 min	No effect on DNA damage (Comet assay)

Chen C et al. (2021) (GE)	Neural stem cells-derived neurons and retinoic acid-induced Neuro-2A cells exposed to 1800 MHz GSM-talk mode signal (5 min ON/10 min OFF) for 48 h at 4W/kg	Gene expression of Eph receptors 5 (EPHA5), which is required for neurite outgrowth, was inhibited. (Decreased neurite outgrowth from cells)
Chen G et al. (2012) (GE)	Saccharomyces cerevisiae yeast cells exposed to 1800 MHz RFR for 6 h; SAR 4.7 W/kg	Expression of several genes.
Cheon et al. (2023) (GE)	SK-MEL-3 melanoma cells exposed to 1.6 pulsed THz RFR (1 kHz repetition) for 1-48 h at 14.7 mW/cm^2	downregulated genes involved in cancer and apoptosis pathways (FOS, JUN, and CXCL8), DNA demethylation involved.
*Choi et al. (2020)	Human adipose tissue-derived stem cells (ASCs), Huh7 and Hep3B liver cancer stem cells (CSCs), HeLa and SH-SY5Y cancer cells, and normal fibroblast IMR-90 cells exposed to WCDMA-signal 1.7-GHz RFR for 72 h, SAR 1 and 2 W/kg	No significant effect on double strand breaks; increased intracellular reactive oxygen species and deceased proliferation.
*Ciaravino et al. (1991)	Chinese hamster ovary cells exposed to 2450-MHz pulsed RFR (SAR 33.8 W/kg) simultaneously with adriamycin for 2 h	RFR did not affect changes in cell progression and number of sister chromatid exchanges induced by Adriamycin.
Costantini et al. (2022) (GE)	Human karotinocytes (HaCat) cells exposed to 27.1 MHz pulse-modulated at 600 Hz RFR for 2 x 30 min, 6 h, or 24 h (No dosimetry data.)	Affected gene expression of cytokines (IL4 and IL6), and matrix metalloproteinases related to wound healing depending on exposure conditions.
Czyz et al. (2004) (GE)	Pluripotent embryonic stem (ES) cells (wild-type and deficient for the	Upregulation of mRNA levels of the heat shock protein, hsp70 and a low and transient increase of c-jun, c-myc, and p21 levels in

	tumor suppressor p53 exposed to pulsed 1710 MHz RFR at GSM-217 (1.5 W/kg) or GSM-talk (0.4 W/kg) mode for 6-48 h	p53-deficient, but not in wild-type cells.
d'Ambrosio et al. (1995)	Human blood exposed to 9 GHz (CW or 50-Hz amplitude-modulated) RFR; 72 h, 90 W/kg	Increased micronucleus in amplitude-modulated RFR exposed blood.
d'Ambrosio et al. (2002)	Human blood cultures exposed to 1748 MHz RFR (continuous –wave or phase modulated (GMSK)) for 15 min: SAR ~5 W/kg	Micronucleus frequency in lymphocytes was increased only after exposure to phase-modulated RFR.
*Danese et al. (2017)	Human blood exposed to a 900-MHz activated mobile phone for 30 min	No change in γ-H2AX foci in DNA of lymphocytes.
Dasdag et al. (2015a) (LI) (GE)	Brain of rats exposed to 900 MHz RFR 3h/day. 7 day/week for 12 months; 0.0369 W/kg	Decreased rno-miR107; no effect on levels of other miRNA studied.
Dasdag et al. (2015b) (LI) (GE)	Brain of rats exposed to 2450-MHz RFR 24 h/day for 1 year; 0.004 W/kg	Decreased expression of microRNA miR-106b-5p and miR-107
Dasdag et al. (2019) (LI) (GE)	Brain of rats exposed to 900 MHz FR 3 h/day (7 days a week) for 12 months, 0.198 W/kg	Increased expression of rno-miR-145-5p (no effect on several other miRNA).
Dasdag et al. (2022) (LI) (GE)	Brain of rats exposed to 2400 MHz RFR emitted from a Wi-Fi generator feed to an antenna for 24 h/day for one year; 0.000328 W/kg	increased rno-miR-181a-5p and membrane lipids
Dasgupta et al. (2022) (GE)	Zebrafish exposed to 3.5 GHz RFR from 6 h post-fertilization to 48 h; 8.27 W/kg	A modest transcriptomic disruption at 48 h post-fertilization, with 28 differentially expressed genes; biochemical pathways related to metabolism were significantly perturbed
*Dawe et al. (2008) (GE)	transgenic hsp16-1::lacZ strain of Caenorhabditis elegans exposed to 1800	No effect on hsp16-1 heat-shock gene expression.

	MHz RFR (continuous-wave or talk-pulsed) for 2.5 h at 1.8 W/kg	
De Amicis et al. (2015)	Human fetal fibroblasts exposed to THz radiation (0.1-0.15 THz) for 20 min; SAR 15-20 W/kg	Increased total number of micronucleus and centromere positive micronuclei that could lead to chromosome loss. No significant effect on DNA strand breaks (Comet assay), phosphorylation of H2AX histone and apoptosis.
De Iuliis et al. (2009)	Human spermatozoa exposed to 1800-MHz RFR; SAR 0.4 – 27.5 W/kg for 16 h	Increased oxidative DNA damage and fragmentation (apoptosis) and reactive oxygen species; sperm motility and vitality were reduced.
*de Oliveira et al. (2017)	Human buccal cells from cell phone users; Averaged years of use 11.4 yrs; mean duration of daily use 2.8 min	Cells ipsilateral to cell phone use did not have a statistically significantly higher micronucleus frequency, compared to cells contralateral to exposure.
Del Re et al. (2019) (GE)	Human HeLa, BE2C and SH-SY5Y cells exposed to 900-MHz 217-Hz pulse-modulated RFR for 48 h; SAR 1 W/kg	Increased transcription of repetitive DNA, type of transcription depended on cell type. (Alteration of repetitive DNA transcription can be induced by environmental stress conditions, causing human pathological effects.)
Del Vecchio et al. (2009) (GE)	Murine SN56 cholinergic cell line (48 and 72 h) and rat primary cortical neurons (24, 72, 120 h) exposed to GSM-modulate 900 MHz RFR; SAR 1 W/kg	Increased expression of beta-thymosin (cytoskeleton regulating factor) m-RNA, and reduced neurite generation.
Demsia et al. (2004)	Bone marrow of rats exposed to 910 MHz RFR 2h/day for 30 days, 0.42 W/kg	Increased micronuclei in polychromatic erythrocytes and polymorphonuclear cells. The induction of micronucleus in female rats was less than that in male rats.
Deshmukh et al. (2013) (LI)	Male Fischer rats exposed to 900 MHz (0.0005953 W/kg), 1800 MHz (0.0005835 W/kg), and 2450 MHz (0.0006672 W/kg) RFR for 2 h/day, 5 days/week for 30 days.	Increased DNA single strand breaks (Comet assay) in brain tissues.
Deshmukh et al. (2015) (LI)	Male Fischer rats exposed to 900 MHz	Increased DNA single strand breaks (Comet assay) in brain tissues; elevated heat-shock

	(0.0005953 W/kg), 1800 MHz (0.0005835 W/kg), and 2450 MHz (0.0006672 W/kg) RFR for 2 h/day, 5 days/week for 180 days.	protein-70 level.
Deshmukh et al. (2016) (LI)	Male Fischer rats exposed to 900 MHz (0.0005953 W/kg), 1800 MHz (0.0005835 W/kg), and 2450 MHz (0.0006672 W/kg) RFR for 2 h/day, 5 days/week for 90 days.	Increased DNA single strand breaks (Comet assay) in brain tissues; elevated heat-shock protein-70 level.
Diem et al. (2005)	Human diploid fibroblasts and cultured rat granulosa cells exposed to 1800 MHz intermittent (5 min On/10 min Off) or continuous–wave; SAR 1.2 or 2 W/kg	Increased in DNA single and double strand breaks (Comet assay) in both cell types after 16 h exposure. Intermittent wave showed a higher effect than continuous wave.
Ding et al (2022) (GE)	Mouse embryo Balb/c-3T3 cells exposed to 1800 MHz RFR; 4 h/day for 40 or 60 days at 8 W/kg	Changes in gene expression, and induction of miRNA (malignant transformation of the cells observed; lipid metabolism was the pivotal biological process and pathway.)
D'Silva et al. (2017)	Liver of chicken embryos (in eggs) exposed to mobile phones emitting 2G or 3G signals; 72 min per day up to 12 days of incubation	Increased DNA single strand break (Comet assay)
D'Silva et al. (2021)	Brain of chicken embryos (in eggs) exposed to mobile phones emitting 2G or 3G signals; 72 min per day up to 12 days of incubation	Increased DNA single strand break (Comet assay)
Duan et al (2015)	Mouse spermatocyte-derived GC-2 cells exposed to intermittent (5 min On/10 min Off)	Increased oxidative DNA damage at 4 W/kg (Comet assay).

	1800 MHz RFR (from a GSM cell phone in talk mode) for 24 h; SAR 1, 2, or 4 W/kg	
*Durdik et al. (2019)	Umbilical cord blood (UCB) cells exposed to a GSM 900 (1-17 h, 0.004 or 0.04 W/kg) or UMTS-1947.4 MHz (3 h, 0.04 /kg) cell phone signals fed to a TEM cell	No changes in DNA single and double strand breaks (Comet assay), and apoptosis; increased reactive oxygen species was observed.
Eker et al. (2018) (LI) (GE)	Female Wistar albino rats exposed to 1800-MHz RFR for 2h/day for 8 weeks; SAR 0.06 W/kg	Caspase-3 and p38MAPK gene expressions increased in eye tissues.
Engelmann et al. (2008) (GE)	Cell suspension cultures of Arabidopsis thaliana exposed to 1900 MHz UMTS-modulated RFR for 24 h; SAR peak 2 W/kg, average 0.75 W/kg	Significant changes in transcription of 10 genes.
Ergun et al. (2022)	C3H cancer fibroblast cells exposed to 2100 MHz RFR for 2 h at 2 W/kg	Increased DNA methyltransferase Activities (epigenetic effect)
Esmekaya et al. (2011) (LI)	Human peripheral blood lymphocytes exposed to 1800 MHz GSM- (217 HZ) modulated RFR for 6, 8, 24, or 48 h; SAR 0.21 W/kg	Chromatin changes and increase in sister chromatin exchange.
*Falzone et al. (2010)	Human spermatozoa exposed to pulse-modulated 900-MHz RFR for 1 or 24 h; SAR: 2.0 and 5.7 W/kg	No significant effects on DNA fragmentation, reactive oxygen species, and capase-3 activity.
Ferreira et al. (2006)	Pregnant rats exposed to a cell phone at 834 MHz for 8.5 h/day from conception to birth: SAR 0.55-1.23 W/kg	Increased erythrocyte micronucleus frequency but no significant effects in oxidative parameters in blood and liver of newborn pups.
*Finnie et al. (2006)	Pregnant mice exposed to 900-MHz RFR (modulated at 217 Hz	No significant effect on c-fos expression in brain of offspring.

	with pulse-width of 0.6 ms) for 60 min per day from day 1-19 of gestation; SAR 4 W/kg	
Fragopoulou et al. (2018)	C57BL/6 adult male mice exposed to 2 hr to GSM 1800-MHz RFR (from a phone) for 2 h at an average power density of 0.0049-0.081 mW/cm^2	In the hippocampus, the expression of 178 genes changed significantly, revealing an impact on genes involved in critical biological processes, such as cell cycle, DNA replication and repair, cell death, cell signaling, nervous system development and function, immune system response, lipid metabolism, and carcinogenesis.
Franchini et al. (2018a)	Human fetal and adult fibroblasts exposed to 25 GHz RFR for 20 min; SAR 20 W/kg	Increased total number of micronuclei and centromere positive micronuclei in exposed samples. No significant effect on DNA single strand break (Comet assay).
Franchini et al. (2018b)	Human adult fibroblasts exposed to 0.15 THz (150 GHz) RFR (4 μs pulses at 25 Hz) for 20 min; SAR 15-20 W/kg	Increased centromere-positive micronuclei frequencies and chromosomal nondisjunction events, indicating induction of aneuploidy and not by DNA breakage.
Franzellitti et al. (2008) (GE)	Human trohoblastes HTR-8/SVneo exposed to 1800 MHz continuous-wave, GSM-217-Hz, and GSM-Talk signals for 4-24 h, time averaged SAR 2 W/kg	Levels of the inducible HSP70C transcript were significantly enhanced after 24 h exposure to GSM-217Hz signals and reduced after 4 and 16 h exposure to GSM-Talk signals. No effect on inducible HSP70A, HSP70B and the constitutive HSC70 transcripts.
Franzellitti et al. (2010)	Human trophoblast HTR-8/SVneo cells exposed to1800 MHz continuous –wave. GSM (217 Hz modulated) and GSM intermittent (5 min on/10 min off) RFR for 4. 16, or 24 h: SAR 2 W/kg	GSM signals increased DNA single strand breaks (Comet assay) after 16 and 24 h exposure; recovered within 2 h post-exposure; continuous-wave RFR was without effect.
*Fritze et al. (1997)	Rats expose to GSM 90 MHz RFR for 4 h, brain average SAR 0.3- 1.5 W/kg	No effect on C-jun and GFAP expression in brain.
Fucic et al. (1992)	Lymphocytes from humans occupationally exposed to RFR; 1250-1350 MHz, 10 $\mu W/cm^2$-	Showed preferentially clastogenic effect measured by micronucleus. Effect on genetic material similar to both of a chemical agent and of ionizing radiation.

	20 mW/cm^2	
Furtado-Filho et al. (2014)	Rats of different ages (0-30 days) exposed 950 MHz RFR for 0.5 h/day for 51 days (21 days of gestation and 6-30 days old): SAR pregnant rat 0.01-0.03 W/kg; neonate 0.88 W/kg, 6-day old 0.51 W/kg, 15-day old 0.18 W/kg, 30-day old 0.06 W/kg.	Decreased DNA single strand breaks (Comet assay) in liver of 15-day old and increased breaks in 30-day old rats, no oxidative stress detected.
*Furtado-Filho et al. (2015)	At exposed to 950 MHz RFR. 0.5 h/day to 27 days (throughout pregnancy and 6 days postnatal); SAR 0.44-0.35 W/kg, neonatal rat 1.32 W/kg, 6-day old 1.14 W/kg	Right cerebral cortex showed an increase in DNA single strand breaks (Comet assay), but no significant effect in the left cerebral cortex in RFR-exposed 6-day old rats. No oxidative effects observed.
Gadhia et al. (2003)	Blood samples of cell phone and non-cell phone users	Increased dicentric chromosomes and sister chromatid exchange in lymphocytes of cell phone users.
Gajski and Garaj-Vrhovac (2009)	Blood samples from Wistar rats exposed to GSM-modulated 915 MHz RFR for 30 min, SAR 0.6 W/kg	Increased basal (single strand) and oxidative DNA damage (Comet assay) in lymphocytes.
Gandhi and Anita (2005)	Blood from cell phone users (most for 2-5 yrs)	Increased DNA single strand breaks (Comet assay) and micronucleus found in cell phone users.
Gandhi and Singh (2005)	Blood and buccal cells from cell phone users (3-4,5 yrs); controls never used cell phone	Increased micronucleated buccal cells and chromosomal aberration in peripheral lymphocytes.
Gandhi et al. (2015a)	People lived within 300 m of a cell phone base station (average power density= 1.149 mW/cm^2) for an average of 7.45 yrs, controls average power density = 0.0045 mW/cm^2.	Increased DNA single strand breaks (Comet assay) in peripheral blood leukocytes. Daily cell phone usage, location of residence, and power density are significant predictor of DNA damage.
Gandhi et al. (2015b)	Buccal cells of mobile phones users (for 3-5 y)	Various measurements showed chromosomal damages and decreased genetic repair in

	and non-mobile phones users	mobile phone users.
Gapeyev et al. (2014)	Mouse whole blood leukocytes exposed to 42.2 GHz RFR modulated at 1 Hz for 20 min at 0.1 mW/cm^2 and then exposed to x-ray (4 Gy)	RFR reduced DNA damage (Comet assay) caused by x-ray (H_2O_2 may play a role).
Garaj-Vrhovac et al. (1990)	V79 Chinese hamster cells exposed to 7.7 GHz RFR for 15, 30, or 60 min; power density 30 mW/cm^2	Inhibited [^{3}H]thymidine into DNA with stoppage of cell cycle at S phase; chromosome aberration observed.
Garaj-Vrhovac et al. (1991)	V79 Chinese hamster fibroblast cells exposed to 7.7 GHz RFR for 15, 30, or 60 min; power density 0.5 mW/cm^2	Increased chromosome aberration (dicentric and ring chromosomes) and micronucleus.
Garaj-Vrhovac et al. (1992)	Human whole blood samples exposed to 7.7 GHz RFR for 10, 30, or 60 min; power density 0.5, 10, or 30 mW/cm^2	Increased chromosome aberration (dicentric and ring chromosomes) and micronucleus in lymphocytes.
Garaj-Vrhovac et al. (1993)	Lymphocyte from radar station personnel who were engaged in repairing radar devices (1250-1350 MHz) a couple of days earlier	Increased number of chromosome breaks, acentric fragments, dicentric and polycentric chromosomes with accompanying fragments, ring chromosomes and chromatid interchange. Some effects persisted at 30 weeks after exposure.
Garaj-Vrhovac. (1999)	Peripheral blood lymphocytes of workers on radar equipment and antenna system service, 1250-1350 MHz; power density 10 µW/cm^2 -20 mW/cm^2; average employment duration 13.3 yrs	Exposed subjects showed an increase in the number of micronucleus and number of micronucleus per cell; disturbance of cells in the cell cycle.
Garaj-Vrhovac and Orescanin (2009a)	Peripheral blood lymphocytes of workers on radar equipment and antenna system service, 1250-1350 MHz; power density 10 µW/cm^2 -20	Increased DNA single strand breaks (Comet assay) and bleomycin-induced chromatid breakage.

	mW/cm^2; average employment duration 13.3 yrs	
Garaj-Vrhovac et al. (2009b)	Wistar rats exposed to 915 MHz RFR 1 h/day for two weeks, SAR 0.6 W/kg	Increased basal DNA single strand break and oxidative DNA damages (Comet assay) in blood lymphocytes.
Garaj-Vrhovac et al. (2011)	Workers occupationally exposed to marine radar pulsed RFR (3, 5.5, and 9.4 GHz)	Increased DNA single strand break (Comet assay) and micronucleus in blood lymphocytes; increased oxidative stress.
*Garson et al. (1991)	Blood samples of radio-linemen occupationally exposed to 400 kHz – 20 GHz	No increase in chromosomal damage in lymphocytes.
Ghandehari et al. (2021)	Buccal mucosa cells of human subjects	Increased amount of cell phone usage had a significant correlation with a higher frequency of micronucleus containing cells and a higher frequency of micronucleus in each cell in the buccal mucosa.
Ghatei et al. (2017) (GE)	Mice exposed pre- and postnatally to radiation from a cellular phone jammer (900 and 1800 MHz)	At 8-10 weeks old, in the cerebellum, no effect on expression level of bcl-2 and p53 genes, but gene expression level of *bax* was decreased and gene expression level of *p21* was increased.
*Gläser et al. (2016)	Human hematopoietic stem cells and HL-60 cells exposed to GSM (900 MHz), UMTS (1,950 MHz) and LTE (2,535 MHz) RFR for 4, 20 or 66 h at 0-4 W/kg	No effect on DNA damage and DNA repair. (Exposure of HSC to the GSM modulation for 4 h caused a small but statistically significant decrease in DNA damage.)
Gökçek-Saraç et al. (2021) (GE)	Hippocampus of rats exposed to UMTS 2100 MHz signal, 2 h/day for 7 days; 0.41 or 1.3 W/kg	Decreased mRNA expressions of acetylcholinesterase, choline acetyltransferase and vesicular acetyl-choline transporter
*Görlitz et al. (2005)	Mice exposed to GSM-900 or DCS1800 RFR; 2 h/day for 1 week (3.7-33.2 W/kg) or 6 weeks (2.8-24.9 W/kg)	No effect on the number of micronuclei in erythrocytes of the bone marrow or peripheral blood, in keratinocytes, or in spleen lymphocytes.
Gorpinchenko et al. (2014)	Human sperms exposed to a cell phone in stand-by/talk mode for 5 h	Increased DNA fragmentation (apoptosis) and decreased motility in spermatozoa.
Goswami et al. (1999)	Murine embryonic	Increased proto-oncogene expression Fos

(GE)	fibroblasts exposed to 835.62 MHz FMCW or 847.74 MHz CDMA RFR at 0.6 W/kg	expression.
Gulati et al. (2016)	Blood and buccal cells of people lived close (<400 meters) to a cell tower; 1800 MHz, Maximum power density (at 150 meters) 1.22 μW/cm^2, some subjects lived in the area for more than 9 yrs	Increased DNA single strand breaks (Comet assay) in lymphocytes and micronucleus in buccal cells. Female subjects had significantly higher effects than males.
Gulati et al. (2020) (GE)	Human lymphocytes exposed to 3G mobile phone radiation (1923, 1947.47, and 1977 MHz); 1 or 3 h	Inhibition of a bulk RNA expression and induction of DNA damage in dependence on UMTS frequency channel with maximal effect at 1977.0 MHz.
Guler et al (2010)	Pregnant and non-pregnant New Zealand white rabbit exposed to GSM 1800-MHz RFR for 15 min/day for 7 days(15th to 22nd days of gestation); power density 0.052 mW/cm^2	Increased oxidative DNA damage and lipid peroxidation in brain tissues in adult rabbits, no significant effect in newborn rats
Güler et al. (2012)	New Zealand white rabbits exposed to GSM 180-MHz RFR for 15 min/day in utero between 15th to 22nd days of gestation and at 1-month old 15 min/day 7 days for female and 14 days for male; SAR 1.8 W/kg	Increased DNA oxidative damage in liver of female rabbits (not in male) and increased lipid peroxidation in liver of both male and female rabbits.
Gunes et al. (2021)	Drosophila melanogaster exposed to 900 MHz, 1800 MHz, and 2100 MHz RFR; 2, 4, or 6 h/day for 2 days'\; 35.2-41 V/m	Number of mutant clones was statistically increased in all applications except for the 6-h application at 1800 MHz.
*Gurbuz et al. (2010)	Female Wistar rats exposed to GSM 1800-MHz RFR 20 min/day, 5 days/week for 1 month;	No significant effect on micronucleus frequency in bladder cells.

	power density 0.0054 mW/cm^2	
*Gurbuz et al. (2014)	Male Wistar rats exposed to 1800- or 2100-MHz RFR 30 min/day, 6 days/week for 1 or 2 months: SAR 0.23 W/kg	No significant effect on micronucleus frequency in bladder cells.
*Gurbuz et al. (2015)	Exfoliated bladder cells of non-diabetic and diabetic rats exposed to 2100 MHz RFR at 0.24 W/kg	No effect on micronucleus formation
*Gurisik et al. (2006) (GE)	Human cell lines (one of neuronal (SK-N-SH) and the other of monocytoid (U937) origin) exposed to a 900 MHz (217 pulses) RFR for 2 h at 0.2 W/kg	No effect on gene expression.
Gürler (2014)	Wistar rats exposed to 2450 MHz RFR 1 h/day for 30 consecutive days; power density 0.0036 mW/cm^2	Increased oxidative DNA damage in brain and blood, and oxidative protein products in blood.
Gustavino et al. (2016)	Secondary roots of Vicia faba (broad bean) seedlings exposed to continuous-wave 915-MHz RFRfor 2 h; SAR 0.4-1.5 W/kg	Increased micronucleus frequency up to 7-fold.
Habauzit et al. (2014) (GE)	Human keratinocytes exposed to 60.4 GHz RFR for 3 hr, incident power desnity of 20 mW/cm^2: SAR 594 W/kg (average), 1233 W/kg (peak)	7 gene expressions showed specific electromagnetic effect under hyperthermia condition (i.e., not mimicked by heat-shock controls).
* Habauzit et al. (2020) (GE)	Male hairless ats exposed to 94 GHz RFR 3 h/day, 3 days/week for 5 months, incident power density 10 mW/cm2	No significant modification of gene expression in skin cells.
Haider et al. (1994)	Plant cutting bearing young flower buds exposed for 30 h to	Increased micronucleus was found in all conditions (compared to lab controls).

	short-wave 10-21 MHz RFR on both sides of a slewable curtain antenna (0.424-7.67 mW/cm^2), at 15 m (2.15 mW/cm^2) and 30 m (1.3 mW/cm^2) from a cage antenna; and 200 m from a broadcasting station (0.00027-0.0024 mW/cm^2)	
Hamann et al. (2006) (GE)	Rat sciatic nerve in mid thigh, or to the L4 anterior primary ramus just distal to the intervertebral foramen exposed to 20 ms 500 KHz pulses at 2 Hz	Up-regulation of transcription factor 3, ATF3, as an indicator of cellular "stress". (Effect appeared to be selective on neurons whose axons are the small diameter C and A delta nociceptive fibers.)
Hanci et al. (2013)	Pregnant rats exposed 1 h/day on days 13-21 of pregnancy to 900-MHz RFR at power density 0.0265 mW/cm^2.	Testicular tissue of 21-day old offspring showed increased DNA oxidative damage, apoptotic index, and lipid peroxidation.
*Hansteen et al. (2009a)	Human lymphocytes exposed to18 GHz or pulsed 16.5 GHz RFR for 53 h	No significant effect on chromosomal aberration frequency.
*Hansteen et al. (2009b)	Human lymphocytes exposed to 2.3 GHz continuous-wave or pulsed (200 Hz, 50% duty cycle) RFR	No significant effect on chromosomal aberration frequency.
Hao et al. (2010) (GE)	Murine N9 microglial cells were exposed to pulsed 2450-MHz RFRfor 20 min, SAR 6.2 W/kg	Significant induced phosphorylation of STAT3, increased transcription levels of the inflammation-associated genes, iNOS and TNF-alpha, which are reported to contain STAT-binding elements in their promoter region. (STAT3 is a transcription activator that mediates the expression of a variety of genes in response to cell stimuli, and thus plays a key role in many cellular processes such as cell growth and apoptosis.)
Hao et al. (2022) (GE)	Rats exposed to 2856 MHz RFR for15 min/day for 3 days, 10.5 W/kg; PC12 cells exposed to	Decreased expression of microRNA miR-30a-5p that regulates genes involved in autophagy.

	2856 MHz RFR for15 min, 19 W/kg	
Harvey and French (2000) (GE)	Humen mast cell HMC-1 exposed to 864.3 MHz RFR for three 20-min/day for 7 days at 7 W/kg	Up-regulation of proto-oncogene c-kit, the transcription factor Nucleoside diphosphate kinase B, the apoptosis-associated gene DAD-1,and some stress response genes.
Hassanzadeh-Taheri et al. (2022)	Human sperm exposed to a mobile phone (2.5 cm away) for 1 h	Increased DNA fragmentation
He et al. (2016) (GE) (LI)	Mouse bone marrow stromal cells exposed to 900 MHz RFR for 3 hours/day for 5 days at peak and average SARs of 4.1×10^{-4} and 2.5×10^{-4} W/kg,	Increased mRNA of poly(ADP-ribose) polymerase-1, a nuclear enzyme which plays an important role in the repair of damaged DNA.
He et al. (2017) (GE) (LI)	Same as above and included e\cells exposed to gamma radiation	Increased mRNA of poly(ADP-ribose) polymerase-1, cells treated with RFR and gamma radiation showed less genetic damage an faster kinetics of repair.
Hekmat et al. (2013) (LI)	Calf thymus exposed to 940 MHz RFR for 45 min; SAR 0.04 W/kg	Altered DNA structure at 0 and 2 h after exposure; conformational changes and disaggregation caused by increment in surface charge and size of DNA.
*Hintzsche and Stopper (2010)	Oral cavity mucosa cells from human subjects who used cell phones for different durations weekly (0, <3 h, and > 3h)	No significant change in micronucleus frequency in mucosa cells with cell phone use.
*Hintzsche et al. (2012a)	Human HaCaT cells and A(L) human-hamster hybrid cells exposed to continuous-wave or GSM-modulated 900 MHz RFR for 30 min or 22 h; power density 0.0066-2.15 mW/cm^2	No significant effect on micronucleus frequency.
*Hintzsche et al. (2012b)	HaCaT and HDF cells exposed to 0.106 THz RFR for 2, 8, or 24 h at 0 - 0.88 mW/cm^2	No effect on micronuclear formation and DNA breaks (Comet assay)
*Hirose et al. (2006)	Human glioblastoma A172 cells exposed to	No significant changes in induction of p53-dependent apoptosis, DNA damage, or other

	2.1425 GHz W-CDMA radiation at SARs of 0.08, 0.25, and 0.8 W/kg, and continuous-wave radiation at 0.08 W/kg for 24 or 48 h; and human IMR-90 fibroblasts from fetal lungs exposed to both W-CDMA and continuous-wave RFR at a SAR of 0.08 W/kg for 28 h	stress response
*Hirose et al. (2007) (GE)	Human glioblastoma A172 cells exposed to W-CDMA radiation at SARs of 0.08 and 0.8 W/kg for 2-48 h, and continuous-wave 2.1425 GHz RFR at 0.08 W/kg for 24 h, and human IMR-90 fibroblasts from fetal lungs were exposed to W-CDMA at 0.08 and 0.8 W/kg for 2 or 28 h, and continuous-wave at 0.08 mW/kg for 28 h.	No significant induction of phosphorylation of hsp27 or expression of hsp gene family.
*Hook et al. (2004)	Human Molt-4 T lymphoblastoid cells exposed to 847.74 MHz code-division multiple-access (CDMA) (SAR 3.2 W/kg), 835.62 MHz frequency-division multiple-access (FDMA) (3.2 W/kg), 813.56 MHz iDEN(R) (iDEN) (0.0024 or 0.024 W/KG), and 836.55 MHz time-division multiple-access (TDMA) (0.0026 or 0.026 W/kg) for up to 24 h	No significant changes in DNA single strand breaks (Comet assay) and apoptosis.
*Hou et al. (2015)	Mouse embryonic fibroblasts (NIH/3T3)	No significant effect on the number of γH2AX. There were increases in free radicals

	exposed to 1800-MHz GSM-talk mode RFR (5 min on/10 min off), for 0.5 to 8 h at an average specific absorption rate of 2 W/kg.	and apoptosis.)
Houston et al. (2019)	Male mice exposed to 906 MHz RFR for 12 h/day for 1, 3, or 5 weeks; SAR 2.2 W/kg	Increased DNA oxidative and fragmentation (Comet assay) in spermatozoa across all exposure periods, increased mitochondrial reactive oxygen species.
*Huang et al. (2008a) (GE)	Jurkat human T lymphoma cells exposed for 24 h to 1763 MHz RFR; SAR 10 W/kg	Alterations in cell proliferation, cell cycle progression, DNA integrity (Comet assay) or global gene expression were not detected.
*Huang et al. (2008b) (GE)	HEI-OC1 immortalized mouse auditory hair cells exposed to 1763 MHz (CDMA) RFR for 24 or 48 h; SAR 20 W/kg	No significant effects on cell cycle distribution, DNA damage (Comet assay), stress response and gene expression
Ibitayo et al. (2017)	Brain of rats exposed to a 2.5 GHz WiFi device (10 cm away) for 30, 45, or 60 consecutive days.	Increased DNA damage.
Jeong et al. (2015) (GE)	Hippocampus of transgenic (Tg)-5xFAD mice exposed to 1950 MHz RFR 2 h/day, 5 day/week for 8 months, 5 W/kg	5 genes (Tshz2, Gm12695, St3gal1, Isx and Tll1), which are involved in amyloid β were significantly altered. Effects not observed in normal mice.
*Jeong et al. (2018)	14-month old C57BL/6 mice exposed to 1950 MHz RFR for 2 h/day, 5 day/week, for 8 months; SAR 5 W/kg	No significant effects on levels of oxidative stress, oxidative DNA damage, apoptosis, astrocyte, or microglia markers in brain tissues.
Jeong et al. (2020) (GE)	Hippocampus of two groups of C57BL/6 mice, aged 2 and 12 months, exposed to 1,950-MHz RFR 2 hr/day, 5 days/week for 8 months; 5 W/kg	15 genes involved in neurogenesis, were altered in both groups; increased gene expression of Epha8 and Wnt6 in the hippocampi of the older group.
Ji et al (2004)	Human subjects used cell phones for 4 h.	DNA single strand breaks (Comet assay) increased in peripheral blood cells (T-cells, B-cells, granulocytes).

Ji et al. (2016)	Mouse bone-marrow stromal cells (BMSC) exposed to 900-MHz RFR for 4 h/day for 5 days; power density 0.12 mW/cm^2; some cells were also irradiated with 1.5 Gy γ-radiation after RFR exposure	RFR followed by γ-radiation exposure significantly decreased number of DNA strand breaks (Comet assay) and resulted in faster kinetics of repair of DNA strand breaks compared to γ-radiation alone. Thus, data suggest that RFR preexposure protected cells from damage induced by γ-radiation.
Jiang et al. (2012)	Mice were pre-exposed to a 900-MHz RFR for 4 h/day for 1, 3, 5, 7, and 14 days; power density 0.12 mW/cm2 and then subjected to an acute dose of 3 Gy γ-radiation	DNA single strand breaks (Comet assay) in blood leukocytes from mice pre-exposed to RFR for 3, 5, 7, and 14 days showed progressively decreased damage and was significantly different from those exposed to γ-radiation alone.
Jiang et al. (2013)	Immature erythrocytes in peripheral blood and bone marrow in mice exposed to900 MHz RFR 4 h/day for 7 days at 0.12 mW/cm^2 and hen expsoed to 3 Gy gamma ray	Pre-exposure of mice to RFR decreased the induction of micronucleus in erythrocytes in the blood and bone marrow caused by ionizing radiation.
Jin et al. (2021) (GE)	murine melanoma cell line B16 and the human keratinocyte cell line HaCaT exposed to EMF-long term evolution radiation (LTE, 1.762 GHz) for 4 h at 8 W/kg; mice exposed to the radiation 8 h/day, 5 day/week for 4 weeks, 6 W/kg	No effect on γ-H2AX (Comet assay) (but p53 gene was up-regulated in cells; reduced the γ-H2AX level in the skin tissue of exposed mice. (p53 plays a role in the protective effect of EMF-LTE against DNA double strand breaks.)
*Juutilainen et al. (2007)	Female CBA/S mice were exposed for 78 weeks (1.5 h/day, 5 day/week) to either a continuous 902.5-MHz signal similar to that emitted by analog NMT (Nordic Mobile Telephone) phones at a whole-body SAR of 1.5	No significant effects of RFR on micronucleus frequency in polychromatic or normochromatic erythrocytes.

	W/kg, or to a pulsed 902.4-MHz signal similar to that of digital GSM phones at 0.35 W/kg and also 4 Gy of X-ray on the first three weeks; female transgenic mice (line K2) and their non-transgenic littermates were exposed for 52 weeks (1.5 h/day, 5 day/week) to two digital mobile phone signals, GSM and DAMPS at SAR 0.5 W/kg, and repeated ultraviolet radiation	
*Kadeh et al. (2023)	Buccal cells from mobile phone users (Period of use: <5 years to > 10 year; duration of use per week:<1h to > 5h.	No difference in micronucleus frequency between buccal cells in oral mucosa of preferred phone-use side from non-preferred side.
*Kao et al. (2013) (GE)	Db/db diabetic mice with wound exposed to pulsed 27.12 MHz RFR	No effect on mRNA expression of vascular endothelial growth factor and basic fibroblast growth factor. However, wounds showed dermal cell proliferation and increased collagen synthesis.
Karaca et al. (2012) (GE)	Mouse brain cells exposed to a 10.715 GHz RFR for 6 h/day for three days, SAR 0.725 W/kg	Increased micronucleus apoptosis and necrosis, and decreased expression of the STAT3 genes.
Keles and Süt (2021)	Pregnant rats exposed to 900 MHz RFR 1 h/day from day 13.5 until birth; 0.0269 mW/cm^2	Offspring RFR-exposed group had low levels of H3K27me3 (epigenetic component) staining in ependymal cells and motor neurons in the spinal cord
*Kerbacher et sl. (1990)	Chinese Hamster Ovary cells exposed for 2 h to pulsed 2450 MHz RFR; SAR 33.8 W/kg	No significant effect on chromosome aberration; no interactions with Mitomycin C and Adriamycin.
Kesari and Behari (2009) (LI)	Male Wistar rats exposed to 50-GHz RFR 2 h/day for 45 days; SAR 0.0008 W/kg	Increased in brain tissue DNA double strand breaks (Comet assay); decreased antioxidant enzymes superoxide dismutase and glutathione peroxidase, and increased catalase activity.
Kesari et al. (2010)	Male Wistar rats exposed	Increased in brain tissue DNA double strand

(LI)	to 2.45-GHz RFR 2 h/day for 35 days; SAR 0.11 W/kg	breaks (Comet assay); decreased antioxidant enzymes superoxide dismutase and glutathione peroxidase, and increased catalase activity.
Kesari et al. (2011)	Rats exposed to 900 MHz RFR from a mobile phone, 2 h/day for 35 days, 0.9 W/kg	Decreased micronuclear formation in blood cells.
Kesari et al. (2014) (LI)	Male Wistar rats exposed to a 3D cell phone. 2 h/day for 60 days; SAR 0.26 W/kg	Increased DNA double strand breaks (comet assay), micronuclei, Caspase 3 and apoptosis in brain cells; activation of hsp27/p38MAPK stress pathway.
*Khalil et al. (2011)	Blood, brain, and spleen from mice exposed to 900 MHz RFR 0 min/day for 30 days, 1 W/kg	No effect on level of 8-oxo-7, 8-dihydro-2'-deoxyguanosine
Khalil et al. (2012)	Male Sprague-Dawley rats exposed for 2 h to 1800-MHz GSM signal, SAR 1 W/kg	Urine samples collected 0.5, 1, 2, and 4 h from the beginning of exposure showed elevated 8-oxo-7, 8-dihydro-2'-deoxyguanosine (from repair of oxidative DNA damage) level.
*Khalil et al. (2014)	Saliva from human subjects 15 or 30 min usage of mobile phone (1800 MHz, 1.09 W/kg)	No effect on level of 8-oxo-7,8-dihydro-2'-deoxyguanosine
Khodamoradi et al. (2022)	Blood of rats exposed to WiFi radiation (2450 MHz, for 2, 24, or 72 h) at 4.2 nW/m^2 and 100 µCi of gamma radiation	RFR synergistically (at 72 h exposure) with gamma radiation to increase DNA double strand break (gamma-H2AX foci).
Kim JY et al. (2008)	Mouse lymphoma cells and Chinese hamster lung cells exposed to 835-MHz RFR for 48 h; SAR 4W/kg	RFR increased clastogen-induced DNA single strand breaks (Comet assay).
*Kim HS et al. (2021)	Blood and placenta of pregnant rats exposed to 915 MHz radiofrequency identification signal for 8 h/day from gestational Day 1 to 19; 4 W/kg	No effect on expression of the gene of placental 11β-hydroxysteroid dehydrogenase type 2 (11β-HSD2) (an enzyme that converts cortisol into its receptor-inactive congener cortisone.) (There was an increase in cortisol level in the blood).
Kim JH et al. (2021) (GE)	SH-SY5Y neuronal cells exposed to 1760 MHz RFR 4 h/day for 4 days;	Transcriptional activation of p53

	4 /kg	
*Komatsubara et al. (2005)	Mouse m5S cells exposed for 2 h to 2450 MHz CW RFR (SAR 5,10, 20, 50 and 100 W/kg) or pulsed RFR (SAR mean 100W/kg, peak 900 W/kg)	No chromosomal aberration observed.
Korenstein-Ilan et al. (2008)	Dividing human lymphocytes exposed to 0.1 THz RFR for 1, 2, or 24 h at 0.031 mW/cm^2	Induced genomic instability in various chromosomes.
Koyama et al (2003)	Chinese hamster ovary K1 cells exposed to 2450 MHz RFR for 18 h; SAR up to 100 W/kg	Increased micronucleus formation at 78 W/kg or higher. Potentiated the effect of bleomycin.
Koyama et al (2004)	Chinese hamster ovary K1 cells exposed to 2450 MHz RFR for 2 h; SAR 5-200 W/kg	Increased micronucleus formation above 50 W/kg (May be related to temperature rise).
*Koyama et al (2016a)	Human corneal epithelial (HCE-T) cells exposed to 0.12 THz RFR for 24 h at 5 mW/cm^2	No effect on micronucleus formation
*Koyama et al (2016b)	Human corneal epithelial (HCE-T) and human lens epithelial (SRA01/04) exposed to 60 GHz RFR (GHz) for 24 h at 1 mW/cm^2.	No effect on micronucleus frequency and DNA strand break (Comet assay).
Kucukbagriacik et al. (2022) (GE) (LI)	Liver and blood of mice exposed to 900 MHz GSM RFR; 4 h/day for 7 days; 0.339 W/kg	Increased DNA repair mechanism genes expression.
*Kuribayashi et al. (2005) (GE)	Brain of rats exposed to 1439 MHz RFR, 90 min/day for 1 - 2 weeks, 2 or 6 W/kg	No effect on expression of three blood-brain barrier-related genes.
Kumar A. et al. (2020)	Allim cepa (onion) root meristematic cells exposed to 900- (0.0902 W/kg) and 1800-MHz (0.169 W/kg) RFR for 0.5, 1, 2, and 4 h	Increased chromosomal aberrations and increased DNA single strand breaks (Comet assay).
*Kumar G. et al.	Long bone (femur and	No significant effect on DNA single-strand

(2011)	tibia) of male Sprague – Dawley rats exposed to 900-MHz continuous-wave RFR for 30 min; SAR 2 W/kg	breaks (Comet assay) in bone marrow lymphocytes. (Assayed at 72 h after exposure.)
*Kumar G. et al. (2015)	Long bone (femur and tibia) of male Sprague – Dawley rats exposed to 900 and 1800 MHz continuous-wave and pulsed RFR; 900-MHz CW at 2 and 10 W/kg for 90 min and 1800-MHz CW and PW at 2.5 and 12.4 W/kg for 120 min	No significant effect on DNA single-strand breaks (Comet assay) in bone marrow lymphoblasts. (Assayed at 1 h after exposure.)
Kumar R et al. (2021) (LI)	Hippocampus of rats exposed to 900 MHz, 1800 MHz or2450 MHz RFR at 5.84×10^{-4} W/kg, 5.94×10^{-4} W/kg and 6.4×10^{-4} W/kg respectively, for 2 h/day for 1-month, 3-month or 6-month periods	Alteration in epigenetic modulation was observed in the hippocampus. Global DNA methylation was decreased, and histone methylation was increased.
Kumar S. et al. (2010) (LI)	Male Wistar rats exposed to 10-GHz RFR 2 h a day for 45 days, SAR 0.014 W/kg	Increased micronucleus and reactive oxygen species in blood cells.
Kumar S. et al. (2013) (LI)	Blood of rats exposed to 10 GHz RFR 2 h/day for 45 days; 0.014 W/kg.	Increased micronucleus frequency and DNA (deoxyribonucleic acid) strand break (Comet assay).
Kumar S. et al. (2014) (LI)	Male Wistar rats exposed to 1910.6 MHz RFR from a cell phone in "talk mode' for 60 days (2 h/day, 6 days a week); SAR 0.28 (Max.) and 0.0226 (Min.)	Increased DNA single strand breaks (Comet assay) and lipid peroxidation in spermatozoa,
Kundu et al. (2021) (GE)	40-day-old Swarnaprabha rice plants exposed to 1837.50 MHz RFR for 2.5 h at 2.75 mW/m2	Upregulation in calmodulin and phytochrome B gene expressions
*Lagroye et al. (2004a)	Sprague-Dawley rats exposed to pulsed 2450-	No significant change in DNA single strand breaks (Comet assay) (with or without

	MHz RFR for 2 h; SAR 1.2 W/kg	proteinase-k treatment of samples-for detection of DNA-protein crosslinks) in brain cells.
*Lagroye et al. (2004b)	Clonal mouse embryo C3H 10T(1/2) cells exposed 2450-MHz continuous-wave RFR for 2 h; SAR 1.9 W/kg	No significant change in DNA single strand breaks (Comet assay) (with or without proteinase-k treatment of samples.)
Lai and Singh (1995)	Male Sprague-Dawley rats exposed to pulsed or continuous-wave 2450-MHz RFR for 2 h; SAR 0.6 and 1.2 W/kg	Increased DNA single strand breaks (Comet assay) in brain cells was observed at 4 h after exposure to pulsed RFR and at 0 and 4 h after continuous-wave exposure.
Lai and Singh (1996)	Male Sprague-Dawley rats exposed to pulsed or continuous-wave 2450-MHz RFR for 2 h; SAR 1.2 W/kg	Increased DNA single- and double-strand breaks (Comet assay) in brain cells was observed at 4 h after exposure to pulsed or continuous-wave RFR.
Lai and Singh (1997)	Male Sprague-Dawley rats exposed to pulsed 2450-MHz RFR for 2 h; SAR 1.2 W/kg	Increased DNA single- and double-strand breaks (Comet assay) in brain cells at 4 h after exposure. Effects blocked by melatonin or the spin-trap compound N-tert-butyl-alpha-phenylnitrone. (Free radicals are involved in the effects).
Lai and Singh (2005)	Male Sprague-Dawley rats exposed to continuous-wave 2450-MHz RFR for 2 h; SAR 0.6 W/kg	Increased DNA single- and double-strand breaks (Comet assay) in brain cells at 4 h after exposure. Effects blocked by a temporally incoherent magnetic field.
Lai et al. (1997)	Male Sprague-Dawley rats exposed to pulsed 2450-MHz RFR for 2 h; SAR 1.2 W/kg	Increased DNA double-strand breaks (Comet assay) in brain cells at 4 h after exposure. Effect blocked by naltrexone. (Involvement of endogenous opioids in the effects).
Lakshmi et al. (2010)	Human subjects professionally used VDTs	No effect on DNA single strand breaks (Comet assay) and micronucleus frequency in blood cells of subjects exposed for 2 years; increased in long-term (>10 years) users.
Lameth et al. (2020) (GE)	Healthy rats, rats undergoing an acute neuroinflammation triggered by a lipopolysaccharide (LPS) treatment, and transgenic hSOD1^{G93A} rats that modeled a	Cortical cell gene modulations triggered by GSM-RFR in the course of an acute neuroinflammation and indicate that GSM-induced gene responses can differ according to pathologies affecting the CNS.

	presymptomatic phase of human amyotrophic lateral sclerosis (ALS) exposed head only to a GSM-1800 MHz RFR for 2 h, SAR 3.22 W/kg.	
*Lamkowski et al. (2018) (GE)	Human peripheral blood cells exposed to 900 MHz RFR for 30, 60, and 90 min; SAR 9.3 W/kg	No significant effect on gene expression.
*Lamkowski et al. (2021) (GE)	Human blood exposed to 900 MHz RFR for 30-90 min, 7-10 W/kg; experiment was repeated after 2 years with the same 5 human subjects	Some changes in microRNA expression was detected in the first experiment, which was not replicated in the second experiment.
Lawler et al. (2022) (GE)	Primary human dermal fibroblasts exposed to 60 GHz RFR, 2-4 days at 2.6 mW/cm^2, 46.8 J/cm^{-2} /day	Genomic and transcriptomic alterations observed.
Le Quément et al. (2012) (GE)	Primary human skin cells exposed to a 60.4-GHz RFR for 1, 6, or 24 h, SAR 42.4 W/kg.	Expression of 130 transcripts were found to be potentially modulated. PCR confirmed 5 genes as differentially expressed after 6 h of exposure.
Lee K-S et al. (2008) (GE)	Drosophila exposed to 833 MHz RFR for 12 or 18 h, 1.6 or 4 W/kg	1.6 W/kg activated an anti-apoptotic gene, whereas SAR 4.0 W/kg activated expression of apoptotic genes.
Lee S et al. (2005) (GE)	HL-60 cells exposed to pulsed 2450 MHz RFR (155 μs, duty cycle 7.5%) for 2 or 6 h; 10 W/kg	Number of gene expressions depended on exposure duration (apoptosis-related genes were among the upregulated ones and the cell cycle genes among the downregulated ones.)
*Lerchl et al. (2020)	Pregnant mice exposed to 1960 MHz UMTS signal from 7-14 post-conception days; 0.04 or 0.4 W/kg; then injected with ethylnitrosourea, at 24, 36, and 72 h post-injection, brain, liver, and lung each fetus were examined	No effect on DNA adenyl adduct formation.
Li H et al. (2022)	PC12 cells and 293T	Transcriptional activity of 5-HT_{1A} receptor

(GE)	cells exposed to 2856 MHz RFR, 5 min 3X at intervals of 5 min, 19 W/kg	promoter containing rs198585630 C allele was higher than that of 5-HT_{1A} receptor promoter containing T allele (polymorphism). In vivo experiment with rats showed cognitive deficits and inhibition of brain electrical activity
*Li L et al. (2001)	Murine C3H 10T(1/2) fibroblasts exposed to 847.74 MHz code-division multiple access (CDMA) and 835.62 frequency-division multiple access (FDMA) RFR for 2, 4, or 24 h; SAR 3.2 - 5.1 W/kg	No significant effect on DNA single strand breaks (Comet assay).
Li M et al. (2018)	Mouse spermatocyte-derived cells (GC-2) were exposed to 1800-MHz RFR for 24 h, SAR 1, 2 or 4 W/kg	No effect on DNA double strand streak, increased DNA single strand breaks (Comet assay); free radicals involved.
Li R et al. (2018)	Mouse spermatocyte-derived cells (GC-2) were exposed to 180 MHz RFR for 24 h, 4 W/kg	Increased DNA damage (Comet assay)
*Li Y et al. (2021) (GE)	Primary hippocampal neurons from C57BL/6 mice and Neuro2a cells exposed to 1,800 MHz RFR for 48 h; 4 W/kg	Rap1 (Ras-related protein 1, a protein involved in cell proliferation and survival) activity was inhibited after 48 h of exposure with no detectable alteration in either gene or protein expression of Rap1.
Li ZQ et al. (2020)	Pregnant rats exposed to 1800 or 2400 MHz RFR beginning on the 21st day of pregnancy; hippocampus of offspring examined at 7 weeks postnatal	Altered mRNA expression of NMDA receptors. (Down and up-regulation dependent on the subtypes of receptor.)
Lin et al. (2016) (LI) (GE)	Budding yeast exposed to 2000 MHz RFR for 96 h at 0.12 W/kg	Upregulate the expression of genes involved in glucose transportation and the tricarboxylic acid (TCA) cycle, but not the glycolysis pathway.
Lin et al. (2016) (LI) (GE)	Budding yeast exposed to 2000 MHz RFR for 96 h, SAR 0.12 W/kg	Upregulation of the expression of genes involved in glucose transportation and the tricarboxylic acid (TCA) cycle.

Lin et al. (2017) (GE)	Mouse Leydig (TM3) cells exposed to 1950 MHz GSM RFR for 24 h at 3 W/kg	Decreased P450scc gene expression. (No effect on steroidogenic acute regulatory protein (StAR) expression.)
Liu C et al. (2013a)	Mouse spermatocyte-derived GC-2 cells exposed to 1800-MHz Global System for Mobile Communication (GSM) signals (5 min on and 10 min off) for 24 h; SAR 1, 2, or 4 W/kg	Increased DNA single strand breaks (Comet assay) and DNA adduct 8-oxoguanine at SAR of 4 W/kg; increased reactive oxygen species generation.
Liu C et al. (2013b)	Mouse spermatocyte-derived GC-2 cell line was exposed to a commercial mobile phone handset once every 20 minutes in standby, listen, dialed or dialing modes for 24 h; power density 0.0059-0.0122 mW/cm^2	Increased DNA single strand breaks (Comet assay) (attenuated by melatonin).
Lixia et al. (2006) (GE)	Human lens epithelial cells exposed to GSM-1.8 GHz RFR for 2 h, SAR 1, 2, 3 W/kg	Increased DNA single strand breaks (Comet assay) at 3 W/kg at 30 min post-exposure; Increased mRNA and protein expression of hsp70.
López-Martín et al. (2009) (LI)	Picrotoxin-pretreated male Sprague-Dawley rats exposed to 900-MHz GSM-modulated or unmodulated RFR for 2 h, SAR modulated RFR 0.03 W/kg average–peak 0.14 W/kg in brain: unmodulated RFR average 0.26 W/kg- peak 1.4 w/kg in brain	Increased c-fos expression in brain areas.
Luukkonen et al. (2009)	Human SH-SY5Y neuroblastoma cells exposed to 872-MHz (CW and GSM) RFR for 1 h; SAR 5 W/kg	CW RFR increased DNA single strand breaks (Comet assay) and reactive oxygen species in cells treated with menadione (a chemical that induces intracellular ROS production and DNA damage) compared to cells treated with menadione alone. GSM-modulated RFR had no significant effect.
*Luukkonen et al.	Human SH-SY5Y	CW and modulated RFR had no significant

(2010)	neuroblastoma cells exposed to 872-MHz (CW and GSM) RFR for 3 h (DNA damage) and 1 h (reactive oxygen species) ; SAR 5 W/kg	effect on DNA single strand breaks (Comet assay) and reactive oxygen species production in cells treated with ferrous chloride,
Maes et al (1993)	Human peripheral blood lymphocytes exposed to pulsed 2450-MHz RFR for 30 or 120 min, SAR 75 W/kg	Increase in the frequency of chromosome aberrations (including dicentric chromosomes and acentric fragments) and micronuclei.
Maes et al (1996)	Human whole blood samples exposed to GSM 954- MHz emitting antenna for 2 h, SAR 1.5 W/kg, some samples also incubated with mitomycin C after exposure	Synergistic effect between RFR and mitomycin C was observed the frequencies of sister chromatid exchanges in metaphase figures.
*Maes et al (1997)	Human whole blood cells exposed to 935.2 MHz RFR alone and in combination with mitomycin C for 2 h; SAR 0.3-0.4 W/kg	No significant effects of RFR on chromosome aberration, sister chromatid exchange, and DNA single strand breaks (Comet assay). No synergistic effect with mitomycin C.
*Maes et al (2000)	Human lymphocytes exposed to 455.7 MHz RFR from antenna of a car phone for 2 h; SAR 6.5 W/kg	No significant effects of RFR on chromosome aberration and sister chromatid exchange. No synergistic effect with mitomycin C.
*Maes et al (2001)	Human lymphocytes exposed to 900-MHz RFR for 2 h, SAR 0-10 W/kg	No significant effects of RFR on chromosome aberration and sister chromatid exchange. No synergistic effect with mitomycin C.
*Maes et al (2006)	Peripheral blood lymphocytes from subjects who were professionally exposed to cell phone RFR	No evidence of RFR-induced genetic effects: DNA single strand breaks (Comet assay), chromosome aberration, and sister chromatid exchange.
*Malini (2017)	Blood and semen of Mobile phone users (classified into 1-5, 6-10, and above 10 h/day usage groups)	No effect on DNA damage (DNA ladder assay)

*Malyapa et al. (1997a)	U87MG and C3H 10T1/2 cells exposed to 2450-MHz continuous-wave RFR for 2 h; SAR 0.7 and 1.9 W/kg	No significant effects on DNA single strand breaks (Comet assay).
*Malyapa et al. (1997b)	Mouse C3H 10T1/2 fibroblasts and human glioblastoma U87MG cells exposed to 835.62 MHz (FMCW) and 847.74 MHz (CDMA) RFR up to 24 h; SAR 0.6 W/kg	No significant effects on DNA single strand breaks (Comet assay).
*Malyapa et al. (1998)	Male Sprague-Dawley rats exposed to 2450 MHz continuous-wave (CW) RFR for 2 h; SAR 1.2 W/kg	No significant effects on DNA single strand breaks (Comet assay) in cerebral cortex or hippocampus.
Manti et al. (2017) (LI) (GE)	Four days-old adult female flies (Drosophila melanogaster) exposed to GSM-1800 talk mode RFR emitted by a commercial cellular phone for 30 min; SAR 0.15 W/kg	168 genes were differentially expressed associated with multiple and critical biological processes, such as basic metabolism and cellular subroutines related to stress response and apoptotic death. Free radicals may be involved.
Manti et al. (2008)	Human peripheral blood lymphocytes exposed a UMTS 1.95 GHz signal for 24 h; SAR 0.5 and 2.0 W/kg; some samples also exposed to x-ray	X-ray induced chromosome exchange per cell was increased by RFR exposure. (RFR may either influence the repair of X-ray-induced DNA breaks or alter the cell death pathways of the damage response.)
Marinelli et al. (2004) (LI) (GE)	acute T-lymphoblastoid leukemia cells exposed to 900 MHz RFR for 2-48 h, SAR 0.0035 W/kg	Increased DNA damage (DNA ladder) and activation of genes involved in pro-survival signaling.
Markova et al. (2005) (LI)	Human lymphocytes exposed to 905 and 915 MHz GSM signals for 1 h, SAR 0.037 W/kg	RFR from GSM cell phone affected chromatin conformation and 53BP1/gamma-H2AX foci similar to heat shock. No significant difference between lymphocytes from healthy and electro-hypersensitive subjects.
Markova et al. (2010) (LI)	Human diploid VH-10 fibroblasts and human adipose-tissue derived	915 MHz and 1947.4 MHz signals inhibited tumor suppressor TP53 binding protein 1 (53BP1) foci that are typically formed at the

	mesenchymal stem cells exposed to GSM (905 MHz or 915 MHz) or UMTS (1947.4 MHz, middle channel) RFR for 1, 2, or 3 hr; SAR 0.037-0.039 W/kg	sites of DNA double strand break location in both cell types. 905 MHz RFR did not inhibit 53BP1 foci in differentiated cells but in stem cells. (Inability to form DNA repair foci has been correlated to radiosensitivity, genomic instability, and other repair deficits.)
Martin et al. (2020) (GE)	Four types of human keratinocytes exposed to 60 GHz RFR for 3 h at 594 W/kg	With four keratinocyte cell types, three different expression patterns (downregulation, upregulation, and no effect) were observed, despite their exposure having been the same in all regards.
Mashevich et al. (2003)	Human peripheral blood lymphocytes exposed to830 MHz RFR for 72 hr, SAR 1.6-8.8 W/kg	A linear increase in chromosome 17 aneuploidy (loss and gain of chromosome) and abnormal chromosome 17 replication were observed as a function of the SAR value, demonstrating that this radiation has a genotoxic effect.
Mazor et al. (2008)	Human lymphocytes exposed to continuous-wave800 MHz for 72 hr; SAR 2,9 and 4,1 W/kg	Increased levels of aneuploidy depending on the chromosome studied as well as on the level of exposure. In chromosomes 1 and 10, there was increased aneuploidy at the higher SAR, while for chromosomes 11 and 17, the increases were observed only for the lower SAR.
*McNamee et al. (2002a)	Human blood cultures exposed to continuous-wave 1900 MHz RFR for 2 h; SAR 0-10 W/kg	No effect on DNA single strand breaks (Comet assay) in leukocytes.
*McNamee et al. (2002b)	Human blood cultures exposed to pulsed 1900 MHz RFR for 2 h; SAR 0-10 W/kg	No effect on DNA single strand breaks (Comet assay) and micronucleus formation in leukocytes.
*McNamee et al. (2003)	Human blood cultures exposed to continuous-wave or pulsed 1900 MHz RFR for 24 h; SAR 0-10 W/kg	No effect on DNA single strand breaks (Comet assay) and micronucleus formation in leukocytes.
*McNamee et al. (2016) (GE)	Male C57BL/6 mice exposed to pulse-modulated or continuous-wave 1900 MHz RFR for 4 h/day for 5 consecutive days; whole body averae SAR	No differentially expressed gene expressions were identified in various regions of the brain.

	~0.2 W/kg and ~1.4 W/kg.	
Meena et al. (2014) (LI)	Sperm of rats exposed to 2450 MHz RFR, 2 h/day for 5 days, 0.14 W/kg	Increased DNA damage (Comet assay) attenuated by melatonin.
Megha et al. (2015a) (LI) (GE)	Fischer rats exposed to 900 and 1800 MHz RFR for 30 days (2 h/day, 5 days/week), SAR 0.00059 and 0.00058 W/kg	Reduced levels of neurotransmitters dopamine, norepinephrine, epinephrine, and serotonin, and downregulation of mRNA of tyrosine hydroxylase and tryptophan hydroxylase (synthesizing enzymes for the transmitters) in the hippocampus.
Megha et al. (2015b) (LI)	Hippocampus of rats exposed to 900 (0.00059 W/kg), 1800 (0.00058 W/kg), or 2450 (0.00066 W/kg) MHz RFR; for 60 days (2 h/day, 5 days/week);	Increased DNA single strand breaks (Comet assay)
*Meltz et al. (1990)	Mouse leukemic cells exposed to pulsed 2450 MHz RFR for 4 h, SAR 40 W/kg	No evidence in any mutagenic action by the RFR exposure alone or interaction with proflavin, a DNA-intercalating drug.
Migdal et al. (2023) (GE) (LI)	Bees exposed to 900 MHz RFR for 0.25, 1, or 3 h at 0.05, 0.3, or 1.4 W/kg	Increased expression of Hsp70 and Hsp 90 genes (at 0.05 W/kg for 3 h)
Mildažienė et al. (2019) (GE)	Sunflower seeds exposed to 5.28 MHz RFR for 5, 10, 15 min, 12.7 kV/m	RFR exposure induced a long-term effect on gene expression in leaves, mostly stimulating expression of proteins involved in photosynthetic processes and their regulation.
Millenbaugh et al. (2008)	Skin of rats exposed to 35 GHz RFR at 75 mW/cm^2 until colonic temperature reached 41-42^{0}C	Changes were detected in 56 genes at 6 h and 58 genes at 24 h after exposure. Genes associated with regulation of transcription, protein folding, oxidative stress, immune response, and tissue matrix turnover were affected at both times. (Changes could be thermally related.)
*Miyakoshi et al. (2002)	Human brain tumor-derived MO54 cells were exposed to 2450 MHz RFR for 2 h at 50 W/kg or 100 W/kg	No effect on DNA strand breaks (Comet assay)

*Miyakoshi et al. (2019)	Human corneal epithelial cells exposed to 5.8-GHz electromagnetic fields for 24 h, 1 mW/cm^2	No effect on DNA strand breaks (Comet assay)
Moghadasi et al. (2021)	Blood of pregnant mice exposed to 900 MHz RFR for 8 h/day for 10 days, 0.045 µw /cm2	Increased 8-hydroxy-2' -deoxyguanosine
Mokarram et al. (2017)	Colon tissue of rats exposed to 900 MHz RFR from a GSM mobile phone for 4 h	Altered the pattern of estrogen receptor α gene methylation
*Nakatani-Enomoto et al. (2016)	human spermatozoa expsoed to 1950 W-CDMA-like RFR for 1 h at 2 or 6 W/kg	No effect on 8-hydroxy-2'-deoxyguanosine
Narasimhan and Huh (1991)	Lambdaphage DNA exposed to short pulses of RFR	Observed conformational anomalies in DNA probably resulting from single strand breaks and localized strand separations induced by RFR.
Nikolova et al. (2005) (GE)	Mouse embryonic neural progenitor stem cells exposed to 1710-MHz GSM RFR for 6 or 48 h; SAR 1.5 W/kg	Exposure for 6 h, but not for 48 h, resulted in a low and transient increase of DNA double-strand breaks and the transcript level of genes related to apoptosis and cell cycle control.
Nittby et al. (2008) (LI)	Fischer 344 rats exposed to 1800 MHz GSM RFR for 6 h; SAR whole body average 0.013 W/kg, head 0.03 W/kg	Expression in cortex and hippocampus of genes connected with membrane functions.
Nylund and Leszczynski (2006) (GE)	Human endothelial cell lines EA.hy926 and EA.hy926v1exposed to 900-MHz GSM RFR for 1 h; SAR 2.8 W/kg	Gene and protein expression were altered dependent on the cell type.
Odaci et al. (2016) (LI)	Pregnant Sprague - Dawley rats exposed to 900 MHz RFR 1 h each day during days 13 - 21 of pregnancy; SAR whole body average 0.024 W/kg	Testis and epididymis of offspring showed higher DNA oxidation and lipid peroxidation at 60 days postnatal.
Ohtani et al. (2016) (GE)	Sprague-Dawley rats exposed to wideband code division multiple	Exposure at 4 W/kg (at 6 h/day) increased core temperature and upregulation of some stress markers, heat-shock proteins and heat-

	access 2140 MHz RFR for 6 h or 3 or 6 h/day for 4 days, SAR 4 or 0.4 W/kg	shock transcription factors family, in the cerebral cortex and cerebellum.
*Ono et al. (2004)	Pregnant lacZ-transgenic mice exposed intermittently (10 sec On, 50 sec OFF) 16 h/day to 2450-MHz RFR from embryonic days of 0 to 15; SAR whole body average 0.71 W/kg	No significant effects on mutation frequencies at the lacZ gene in spleen, liver, brain, and testis in offspring. The RFR is not mutagenic in utero.
Ozgur et al. (2014)	Hepatocarcinoma cells exposed to intermittent (15 min ON, 15 min OFF) GSM 900- and 1800-MHz RFR for 1, 2, 3, or 4 h; SAR 2 W/kg	Cells showed irregular nuclei pattern and DNA damage (apoptosis).
Pacini et al. (2002) (GE)	Human skin fibroblasts exposed to GSM 904.2-MHz RFR for 1 h (from a cell phone); SAR 0.6 W/kg	Increased the expression of mitogenic signal transduction genes (e.g., MAP kinase 3, G2/mitotic-specific cyclin G1), cell growth inhibitors (e.g., transforming growth factor-beta), and genes controlling apoptosis (e.g., bax).
Panagopoulos et al. (2007)	Flies (Drosophila melanogaster) exposed to either GSM 900-MHz or DCS 1800-MHz signals from a digital cell phone, for few minutes per day during the first 6 days of their adult life.	Degeneration of large numbers of egg chambers after DNA fragmentation (apoptosis) of their constituent cells, induced by both types of mobile telephony radiation.
Panagopoulos (2019)	Human peripheral blood lymphocytes exposed to UMTS signal (1900-2200 MHz) using a cell phone for 15 min	Chromatid-type aberrations (gaps and breaks) observed.
Panagopoulos (2020)	Human peripheral blood lymphocytes in G2/M phase exposed to signal from a UMTS mobile phone for 15 min	Induction of chromatid-type aberrations
Pandey et al. (2017)	Swiss albino mice exposed to 900-MHz RFR for 4 or 8 h per day for	RFR exposure-induced oxidative stress causes DNA single-strand breaks (Comet assay) in germ cells, with altered cell cycle progression

	35 days; SAR 0.0054-0.0516 W/kg	leading to low sperm count in mice (depolarization of mitochondrial membranes resulting in destabilized cellular redox homeostasis). Larger effect with longer exposure time, and recovery at 35 days post-exposure.
Pandey and Giri (2018) (LI)	Swiss albino mice exposed to GSM 900-MHz RFR 3h twice/day for 35 days, SAR 0.0516-0.0054W/kg	Increased DNA single strand breaks (Comet assay) and free radicals in testis and germ cells, effects attenuated by melatonin
*Paparini et al. (2008) (GE)	Mice exposed to GSM 1800-MHz signal for 1 h; SAR whole body average 1.1 W/kg, brain 0.2 W/kg	No significant modulation in gene expression in whole brain.
Paulraj and Behari (2006)	35-day old male Wistar rats exposed 2 h/day for 35 days to 2450 MHz or 16.6 GHz RFR; SAR 1.0 and 2.01 W/kg , respectively.	Increased in DNA single strand breaks (Comet assay) in brain cells for both frequencies.
Pesnya and Romanovsky (2013)	Onion (Allium cepa) exposed to GSM 900-MHz RFR from a cell phone for 1 h/day or 9 h/day for 3 days; incident power density 0.05 μW/cm^2	Increased the mitotic index, the frequency of mitotic and chromosome abnormalities, and the micronucleus frequency in an exposure-duration manner.
Phillips et al. (1998) (LI)	Human Molt-4 T-lymphoblastoid cells exposed to pulsed signals at cellular telephone frequencies of 813.5625 MHz (iDEN signal) and 836.55 MHz (TDMA signal) for 2or 21 h. SAR 0.0024 and 0.024 W/Kg for iDEN and 0.0026 and 0.026 W/kg for TDMA)	Changes in DNA single strand breaks (increase and decrease depending on exposure parameters) (Comet assay) were observed.
Piccinetti et al. (2018) (GE) (LI)	zebrafish embryos exposed to 100 MHz RFR from immediately after	At the 48 h post-fertilization stage- an increased transcription of oxidative stress genes and reduced growth.

	fertilization to 72 h post-fertilization, SAR about three orders of magnitude lower than ICNIRP basic restrictions of 0.08 W/kg.	
Pooam etal. (2022) (GE)	Human HEK293 cells exposed to 1800 MHz RFR; 15 min (no dosimetry data.)	Increased gene expression of antioxidant enzymes (SOD, GPx, CAT)
Porcher et al. (2023) (GE) (LI)	Leaves of Arabidopsis thaliana exposed to 2450 MHz RFR for 30 min at 0.21 W/kg	Increased accumulation of transcripts of stress-related genes (TCH1 and ZAT12 transcription factor) and genes involved in ROS metabolism (RBOHF and APX1)
*Port et al. (2003) (GE)	Human leukaemia cells (HL-60) exposed to pulsed (1 Hz) 400 MHz RFR for 6 min;50 kV/m-25 times higher than the ICNIRP reference levels for occupational exposure	No significant effects on apoptosis, micronucleation, abnormal morphologies, and gene expression assayed at 9, 24, 48, and 72 h post-exposure.
Pyrpasopoulou et al. (2004) (GE) (LI)	Kidney of offspring of rats exposed to 9400 MHz RFR on 1-7 post-coitum days at 0.0005 W/kg	Increased bone morphogenetic proteins (BMP-4) and their receptors (BMPR-IA), and decreased BMPR-II gene expressions.
Qin et al. (2014) (LI) (GE)	Rats exposed to 1800 MHz RFR for 2 h/day for 32 days at 0.0405 W/kg.	Altered mRNA expression of cytochrome P450 and steroidogenic acute regulatory protein.
Qin et al. (2018) (LI)	Male mice exposed to 1800-MHz RFR 2 h/day for 32 days, SAR 0.0553 W/kg	Inhibition of testosterone synthesis might be mediated through CaMKI/RORα signaling pathway.
Qin et al. (2019) (LI) (GE)	Mouse Leydig cells exposed to 1800 MHz RFR; 1, 2, or 4 h; 0.116 W/kg	Down-regulation of testosterone synthase genes and clock genes.
Qin et al. (2021) (GE)	Testis of four-week-old mice exposed to 1800 MHz RFR, 1 h at 7 am and another hour at 7 pm each day for 21 days;	Expression of non-coding RNAs play crucial roles in the RFR exposure damage to the developing pubertal testis.

	0.50 W/kg	
*Qutob et al. (2006) (GE)	Human U87MG glioblastoma cells exposed to pulse-modulated 1900 MHz RFR for 4 h; SAR 0.1, 1.0, and10 W/kg	No significant effect on gene expression.
Racuciu (2009)	Zea mays root tips exposed to continuous-wave 900 MHz RFR for 1 – 36 h; SAR < 1 W/kg)	Increased mitotic index and chromosomal aberration frequency linear with increased exposure time.
Rago et al. (2013)	Human subjects with different daily durations of cell phone use (no use, < 2 h, 2-4 h, > 4 h) and "trouser users" and "shirt users"	>4 h and "trouser users" had higher sperm DNA fragmentations.
Rammal et al. (2014)	Lycospersicon esculentum (tomato) exposed to 1250 MHz RFR at 6 V/m for 10 days	Increased expression of mRNA of stress proteins
*Regalbuto et al. (2020) (GE)	Human fibroblasts exposed to continuous or pulsed 2450 MHz RFR for 2 h, 0.7 W/kg.	No effect on gene expression profile.
Remondini et al. (2006) (GE)	Six human cell types exposed to 900 and 1800 MHz RFR; three exposure systems were used, exposure time 1, 24, or 44 h, SAR 1 - 2.5 W/kg (Details in Table 1 of paper.)	Some but not all human cells reacted to RFR with an increase in expression of genes encoding ribosomal proteins and therefore up-regulating the cellular metabolism.
Romano-Spica et al. (2000) (GE)	Jurkat T-lymphoblastoid and Leydig TM3 cells exposed to 50 MHz 16-Hz modulated (a calcium resonance frequency) RFR for 0.5-6 h 60 V/m	overexpression of the ets1 mRNA (a transcription factor). No effect with non-modulated field.
*Ros-Lior et al. (2012)	Cells collected from cheeks of human	Compared control cheek area with the side cell phone was placed, no significant genotoxic effect was found (DNA damage

	subjects	and cytokinetic defects, proliferative potential, and cell death).
*Roti-Roti et al (2001)	C3H 10T(1/2) cells exposed to 835.62 MHz FDMA or 847.74 MHz CDMA for 7 days and then one-dose X-ray followed by RFR for 42 days; SAR 0.6 W/kg	No significant effect of RFR on neoplastic transformation (induced by X-ray) was observed.
Roux et al. (2006) (GE)	Tomato plants exposed to a 900-MHz RFR, 2-10 min, 5-40 V/m	Stress-related transcripts (calmodulin, protease inhibitor and chloroplast mRNA-binding protein) increased 15 min after the end of exposure (10 min), dropped to close to initial levels by 30 min, and then increased again at 60 min.
Roux et al. (2008) (GE)	Tomato plants exposed to a 900-MHz RFR for 10 min at 0.0066 mW/cm^2	Induction of stress gene expression; similar to wound responses suggesting that the radiation is perceived by plants as an injurious stimulus.
*Roux et al. (2011) (GE)	Human keratinocytes exposed to 900 MHz RFR for 10 or 30 min at 0.0026 - 0.073 W/kg	No effect on gene expression (47000 human genes).
Sagripanti and Swicord (1986)	Purified DNA solution exposed to 2.55-GHz RFR for 20min; SAR_{min} and SAR_{max} ranges: 0, 2-8-5 and 21-85 W/kg,	Structural changes in DNA suggested that exposure to RFR can cause single as well as double-strand breaks in DNA in solution.
Sagripanti et al. (1987)	Purified plasmid DNA exposed to RFR in the frequency range from 2.00 to 8.75 GHz for 20 min; SAR 0, 8.5, or 85 W/kg	Induced dose- and exposure-duration-dependent DNA single and double strand beaks depends on the presence of small amounts of cuprous ions.
Sahin et al. (2016)	Brain of rats exposed to 3-G 2100 MHz RFR 6 h/day (5 days/week) for 10 or 40 days, 0.4 W/kg	Oxidative DNA damage increased after 5 days and decreased after 40 days of exposure.
Said-Salman et al. (2019) (GE)	Escherichia coli K-12 DH5α exposed to 2.4 GHz RFR for 5 h	Expression of 101 genes were differentially affects (up- and down-regulation).
Saka et al. (2023) (GE)	Primary cortical neurons from neonatal rat cerebral cortex exposed to 2100 MHz RFR for 2	Increased expression of Bax and p53 genes, decreased expression of Bc12 gene

	h at 1.6 W/kg	
*Sakuma et al. (2006)	Human glioblastoma A172 cells exposed to W-CDMA 2.1426 GHz radiation at SARs of 80, 250, and 800 mW/kg and CW radiation at 0.08 W/kg for 2 and 24 h; normal human IMR-90 fibroblasts from fetal lungs exposed to W-CDMA and CW radiations at 0.08 W/kg for 2 and 24 h.	No significant effect on DNA single strand breaks (Comet assay).
*Sakurai et al. (2011) (GE)	Human glial cell line, SVGp12, exposed to continuous-wave 2450 MHz RFR for 1, 4, and 24 h; SAR 1, 5, and 10 W/kg	No effect on gene expression.
Salameh et al. (2022) (GE)	Liver of rats exposed pre-and postnatally (24 h/day) to GSM 900 MHz RFR; 0.783 W/kg at liver	Reduction (PND1 and PND9) in catalase amounts and mRNA expression, SOD1 mRNA expression increased at PND1, and decreased at PND9 and 21
*Salmen et al. (2018)	S. aureus, S. epidermidis, and P. aeruginosa. Exposed to exposed to 900 and 1800 MHz RFR for 2 h using a cell phone	No significant effects on DNA, growth rate and antibiotic susceptibility
*Sannino et al. (2006)	Human peripheral blood leukocytes exposed to 1950 MHz UMTS signal for 24 h; 0.5 or 2 W/kg	No effect on DNA strand breaks (Comet assay)
*Sannino et al. (2009)	Human dermal fibroblasts from a healthy subject and from a subject affected by Turner's syndrome exposed to GSM 900 MHz.RFR for 24 h; SAR 1 W/kg	No effect on DNA single strand breaks (Comet assay)
Sannino et al. (2011)	Human lymphocytes exposed to 900 MHz RFR for 20 h at 1.25 W/kg and then	Adaptive response on micronucleus formation by mitomycin C occurred when RFR was applied in S phase of the cell cycle.

	challenged with mitomycin C	
Sannino et al. (2014) (LI)	Human lymphocytes exposed to 1950 MHz RFR for 20 h at 0.3 W/kg and then challenged with 1.0 or 1.5 Gy X-irradiation	A decrease in the number of micronuclei in lymphocytes exposed to RFR + x-ray as compared with those subjected to x-ray alone
Sannino et al. (2017) (LI)	Chinese hamster lung fibroblast cells (V79) exposed to UMTS 1950 MHz RFR for 20 h at 0.15-1.25 W/kg	increased micronucleus frequency in cells exposed to 0.15 and 0.3 W/kg (no effect at 0.6 and 1.25 W/kg). RFR decreased mitomycin C-induced micronucleus.
Sarimov et al. (2004) (LI)	Human lymphocytes exposed to GSM 895-915 MHz signals for 30 min; SAR 0.0054 W/kg	Condensation of chromatin was observed. (Stronger effect at 1 h exposure.)
Sarkar et al. (1994)	Mice exposed to 2450 MHz RFR 2 h/day for 120, 150, and 200 days; SAR 1.18 W/kg	Rearrangements of DNA segments were observed in brain and testis.
Scarfi et al. (1996)	bovine *(Bos taurus* L.) peripheral blood lymphocytes exposed to 9 GHz RFR for 10 min at 70 W/kg	Increased micronucleus formation.
*Scarfi et al. (2003)	Human peripheral blood lymphocytes exposed to 120-140 GHz RFR for 20 min at 1 mW	No effect on direct chromosomal damage (micronucleus) and cell cycle kinetics
*Scarfi et al. (2006)	Human lymphocytes exposed to GSM 900 MHz RFR for 24 h, SAR 1, 5, and 10 W/kg).	The results provided no evidence for the existence of genotoxic (micronucleus) or cytotoxic effects
* Schuermann et al. (2020)	Human MRC-5 lung fibroblasts, human osteosarcoma cells, HTR-8/SVneo human trophoblasts, and GFP-tagged XRcc1 cells exposed to intermittent (5/10 min ON/FF) or continuous 1950 MHz, 2450 MHz (GSM or unmodulated) RFR for 1-	No significant effect on DNA single strand breaks (Comet assay).

	24 h; SAR 0.5-4.9 W/kg.	
Schwarz et al. (2008)	Human fibroblasts and lymphocytes exposed to UMTS 1950 MHz RFR for 4-48 h; SAR 0.05 to 2 W/kg	Increased DNA single strand breaks (Comet assay) and micronucleus were observed in fibroblasts but not in lymphocytes either unstimulated or stimulated with phytohemegglutinin.
Sekeroğlu V et al. (2012)	Immature (2 week old) and mature (10 weeks old) Wistar rats exposed to continuous–wave 1800 MHz RFR for 2 h/days for 45 days; SAR 0.38-0.78 W/kg (immature rats), 0.31-0.52 W/kg (mature rats)	Bone marrow cells showed chromosome aberrations, micronucleus frequency, mitotic index and ratio of polychromatic erythrocytes (PCEs) in all exposed groups. Immature group showed more effect and less recovery at day 15 post-exposure.
Sekeroglu ZA et al. (2013)	Immature and mature rats exposed to 900 MHz RFR 2 h/day for 45 days; SARs for immature and mature rats were in the range of 0.38-0.78 W/kg, and 0.31-0.52 W/kg, respectively.	Increased chromosome aberrations, micronucleus frequency, mitotic index, and ratio of polychromatic erythrocytes. Effects persisted at 15 days post-exposure.
*Sekijima et al. (2010) (GE)	Human A172 (glioblastoma), H4 (neuroglioma), and IMR-90 (fibroblasts from normal fetal lung) cells exposed to continuous-wave and W-CDMA 2.1425 GHz RFR up to 96 h; SAR 0.08, 0.25, 0.8 W/kg	No significant effects on gene expression and cell proliferation.
Semin et al. (1995)	DNA in glycine and formaldehyde exposed to 10 different 4 to 8 GHz RFR 25 ms pulses, 1-6-Hz repetition rate, 0.4 to 0.7 mW/cm^2 peak power density	3 or 4 Hz pulses and 0.6 mW/cm^2 peak power increased the accumulated damage to the DNA secondary structure. However, changing the pulse repetition rate to 1, 5, 6 Hz, as well as changing the peak power to 0.4 or 0.7 mW/cm^2 had no effect (“window effect”).
*Senturk et al. (2019)	Lymphocytes from patients received radiofrequency treatment on inferior turbinate as	No significant effect on DNA single strand breaks (Comet assay) on Day 15 post-treatment. Increase in oxidative stress was observed.

	they were diagnosed with inferior turbinate hypertrophy	
Shah et al. (2015)	Human lymphocytes exposed to 916 MHz RFR at 20 or 30 dBM for 1-8 h	Increased chromosomal damage observed at higher intensity after 7 h of exposure.
Shahin et al. (2013) (LI)	Female mice (Mus musculus) exposed to continuous-wave 2.45 GHz RFR 2 h/day for 45 days; SAR 0.023 W/kg	Increased DNA strand breaks (Comet assay) observed in the brain. Changes in oxidative mechanisms and oxidative stress were observed in liver, kidney and ovary. Increased embryo implantation/resorption and abnormal pregnancy were observed.
Shahin et al. (2019)	Male Wistar rats exposed to 900 MHz RFR for 2 h/day for 8 weeks, SAR 1.075 W/kg	Increased DNA single strand breaks (Comet assay) in testis and increased oxidative stress.
Sharma and Shukla (2020) (LI)	Male Wistar rats exposed to 900 MHz RFR for 1, 2, or 4 h/day for 90 days; SAR brain 0.231 W/kg	Increased DNA single strand breaks (Comet assay) and increased oxidative stress in brain.
Sharma et al. (2020)	Brain (hippocampus and cortex) of rats exposed to 1800 MHz RFR for 90 days (4 h/day); 0.433 W/kg	Increased DNA single strand breaks Comet assay).
Sharma et al. (2023)	Trigonella foenum-graecum (spice fenugreek) exposed to 900 MHz RFR for 0.5-8 h/day for 7 days, 0.01 W	Induction of chromosomal aberrations in root tip cells.
Shckorbatov et al. (2009)	Human buccal epithelium cells exposed to 35 GHz RFR for 10 seconds, 30 μW/cm^2, 0.75 W/kg	Induced chromatin condensation; depended on polarization
Shckorbatov et al. (2010)	Human fibroblasts exposed to 36.65 GHz RFR for 10 seconds at 1-100 μW/cm^2	Increased heterochromatin granule quantity: right-handed elliptically polarized radiation revealed more biological activity than the left-handed polarized one.
*Shi et al (2014)	Cultured human lens epithelial cells (HLECs) exposed to 90 kHz magnetic field for 2 and 4 h; 93.36 μT	No significant effects on DNA single strand break (Comet assay) and double strand breaks.

*Silva et al. (2016) (LI)	Human thyroid cells exposed to 900 MHz (3 or 6 h, 0.082 W/kg) or 895 MHz (65 h, 0.170 W/kg)	No effect on DNA ploidy
Singh et al. (2023)	Brain (hippocampus and cerebral cortex) of rats exposed to 2115 MHz RFR for 8 h at head SAR 1.51 W/kg	Increased single strand DNA breaks (Comet assay)
Smith-Roe et al. (2020)	Male and female Hsd:Sprague Dawley rats and B6C3F1/N mice exposed from Gestation day 5 or Postnatal day 35, respectively, to code division multiple access (CDMA) or global system for mobile modulations over 18 hr/day, at 10-min intervals for 19 (rats) or 14 (mice) weeks; SAR 1.5, 3, or 6 W/kg (rats, 900 MHz) or 2.5, 5, or 10 W/kg (mice, 1,900 MHz).	Significant increases in DNA single strand breaks (Comet assay) observed in the frontal cortex of male mice (both modulations), leukocytes of female mice (CDMA only), and hippocampus of male rats (CDMA only). No significant increases in micronucleated red blood cells were observed in rats or mice.
Sokolovic et al. (2015) (LI)	Testicular tissue of rats exposed to 900 MHz GSM RFR; 4 h/day for 20, 40, or 60 days, 0.043-0,135 W/kg	Increased DNAse activity (DNA fragmentation).
Son et al. (2023) (GE)	Hippocampus of 5xFAD mice (mouse model of Alzheimer's disease) exposed to 1950 MHz RFR, 2 h/day. 5 days/week for 6 months at 5 W/kg	Decreased expression of genes related to microgliosis and microglial functions, and interleukin-1β.
Soubere Mahamoud et al. (2016) (GE)	Human keratinocyte exposed to a 60.4-GHz RFR at an incident power density of 20 mW/cm^2 for 3 hours	No keratinocyte transcriptome modifications were observed. Co-treatment with a glycolysis inhibitor slightly alters the transcriptome of 6 genes encoding transcription factors or inhibitors of cytokine pathways. Thus, the RFR exposure may affect

		metabolically stressed cells
Souza et al. (2014) (GE)	Exfoliated cells from the oral epithelium from human subjects who spent different time using cell phones (group I, t > 5 h; group II, t > 1 h and ≤ 5 h; and group III, t ≤ 1 h).	Structures that may be associated with gene amplification were significantly greater in the individuals in group I. No significant effects on micronucleus frequency and apoptosis and necrosis were observed.
Spandole-Dinu et al. (2023) (LI)	Brain of mice exposed to 2450 MHz RFR with signal of a WiFi router fed to an antenna for 16 weeks, maximum local SAR in the body 0.01786 W/kg (0.00062-0.0014 W/kg)	Global DNA methylation was lower in exposed mice.
*Speit et al. (2007)	Human fibroblasts (ES1 cells) and Chinese hamster cells (V79) exposed to intermittent (5 min ON/10 min OFF)1800-MHz for 1, 4, 24 h; RFR; SAR 2 W/kg	No significant effects on DNA single strand break (Comet assay) and micronucleus frequency.
*Speit et al. (2013)	Human HL-60 exposed to intermittent (5 min ON/10 min OFF) 1800 MHz RFR for 24 hr; SAR 1.3 W.kg	No significant effects on DNA single strand break (Comet assay) and micronucleus frequency.
Srujana Aravinda et al. (2022)	Buccal mucosa of children with different duration of mobile phone use (Group 1: Usage of 1-2 h a day, Group 2: Usage of 3-6 h a day, and Group 3: Usage of >6 h a day)	Group 3 showed the highest number of cellular and chromosomal aberrations.
*Stronati et al. (2006)	Human blood samples exposed to GSM 935-MHz signal for 24h; SAR 1 and 2 W/kg	Lymphocytes showed no changes in DNA sngle strand breaks (Comet assay), chromosomal aberrations, sister chromatid exchanges, micronuclei frequency and cell cycle. No significant interaction with x-ray.
*Su et al (2017)	Neurogenic A172, U251, and SH-SY5Y cells	No significant DNA damage (gamma H2AX)

	exposed to an intermittently (5 min ON/10 min OFF) 1800 MHz RFR at SAR of 4.0 W/kg for 1, 6, or 24 h.	
*Su et al. (2018)	Primary cultured astrocytes, microglia and cortical neurons were exposed to intermittent (5 min ON/10 min OFF) GSM 1800 MHz RFR for 1, 6 or 24 h; SAR 4.0 W/kg.	The RFR did not elicit DNA double strand breaks (γH2AX foci) but inhibited the phagocytic ability of microglia and the axon branch length and branch number of cortical neurons.
Sueiro-Benavides et al. (2023) (GE)	HL-60 cells exposed to 2450 MHz RFR for 8, 24, or 48 h, 0.406 W/kg	Increased FAS-R gene (related to apoptosis) expression after 24 and 48 h exposure. Interacted with black carbon particles.
Sun C. et al. (2016)	Mouse embryonic fibroblasts (MEFs) with proficient ($Atm^{+/+}$) or deficient ($Atm^{-/-}$) ataxia telangiectasia mutated, which is critical to initiation of DNA repair, to GSM 1800-MHz RFR for 1, 12, 24, or 36 h; SAR 4 W/kg.	Increased DNA single-strand breaks (SSBs) (Comet assay) and activated the SSB repair mechanism. This effect reduced the DNA damage to less than that of the background level after 36 hours of exposure. In the $Atm^{-/-}$ MEFs, the same RF-EMF exposure for 12 h induced both DNA single and double-strand breaks (Comet assay) and activated the two repair processes, which also reduced the DNA damage to less than the control level after prolonged exposure. (Compensatory effects) (Conclusion from interpretation f different results from ($Atm^{+/+}$) and ($Atm^{-/-}$) cells.
Sun, LX et al. (2006a)	Human lens epithelial cells exposed to 217 Hz-modulated 1800 MHz RFR for 2 h; SAR 1, 2, 3, 4 W/kg	No or repairable DNA single strand breaks (Comet assay) was observed after 2 hour irradiation of 1.8 GHz microwave on LECs when SAR </= 3 W/kg. DNA damages caused by 4 W/kg irradiation were irreversible.
Sun, LX et al. (2006b)	Human lens epithelial cells exposed to 217 Hz-modulated 1800 MHz RFR for 2 h; SAR 1, 2, 3, 4 W/kg	No DNA single strand break (Comet assay) was induced using comet assay after 2 hours irradiation of 1. 8 GHz microwave on hLECs at the dose SAR < or = 3.0 W/kg. 4.0 W/kg irradiation caused DNA damage and inhibition of hLECs proliferation.
Sun Y et al. (2017) (GE) (LI)	Human leukemia HL-60 cells exposed to 900 MHz RFR for 4 h/day	Decreased mitochondrial transcription factor A, mtDNA polymerase gamma, mtDNA transcripts and mtDNA copy number;

	for 5 days; average SAR 2.5 x 10^{-4} W/kg	increased 8-hydroxy-2'-dexoyguanosine
Sykes et al. (2001)	pKZ1 mice exposed daily for 30 min to 217-Hz modulated 900 MHz RFR 1, 5, or 25 days; SAR 4 W/kg	After 25 days of exposure, RFR could lead to a perturbation in recombination frequency which may have implications for recombination repair of DNA.
*Takahashi et al. (2002)	Male BigBlue mice (BBM) exposed to 1.5 GHz RFR in the head region for 90 min/day, 5 days/week, for 4 weeks; SAR 0.67 and 2 W/kg	There was no significant variation in the frequency of independent mutations of the lacItrans gene and deletion mutation in the brain.
Tarsaei et al. (2022) (GE)	Hippocampus of rats exposed to 2450 MHz RFR for 7 or 30 days, at 4 mW/cm^2	Increased Bax (Bcl2-associated x) and reduced Bcl-2 (B-cell lymphoma 2) genes expression. (Genes are related to apoptosis.)
Tice et al. (2002)	Human blood leukocytes and lymphocytes exposed to voice modulated 837 MHz produced by an analog signal generator or by a time division multiple access (TDMA) cellular telephone, 837 MHz generated by a code division multiple access (CDMA) cellular telephone (not voice modulated), and voice modulated 1909.8 MHz generated by a global system of mobile communication (GSM)-type personal communication systems (PCS) cellular telephone for 3 or 24 h, SAR 1-10 W/kg	No significant effect on DNA single strand break (Comet assay). Exposure to each of the four RF signal technologies for 24 h at an average SAR of 5.0 or 10.0 W/kg resulted in a significant and reproducible increase in the frequency of micronucleated lymphocytes.
Tiwari et al. (2008)	Blood samples from male human subjects exposed to a CDMA cell phone for 1 h	In vitro exposure to RFR induces reversible DNA single strand breaks (Comet assay) in synergism with aphidicolin, a DNA repair inhibitor,
Tkalec et al. (2009)	Allium cepa L root meristematic cells from	Lagging chromosomes, vagrants, disturbed anaphase and chromosome stickiness were

	seeds exposed to 400 and 900 MHz RFR for 2 h, power density 10, 23, 41 and 120 V/m).	observed.
Tkalec et al. (2013) (LI)	Earthworm (Eisenia fetida) exposed to continuous-wave and AM-modulated 900-MHz RFR for 2 - 4 h; SAR 0.00013, 0.00035, 0.0011, and 0.00933 W/kg	Increased DNA single strand breaks (Comet assay) in earthworms and oxidative stress (lipid and protein oxidation)
Tohidi et al. (2021) (GE)	Hippocampus of mice exposed to RFR from a mobile phone jammer with 4 bands (900 MHz, 1800 MHz, CDMA and GSM) for 0.5, 1, 2, or 4 twice a day for 30 days, or 4 h/day for 30 days	expression of both *Bax* and *Bcl2* genes (related to apoptosis) were upregulated in mice exposed for one and two hours, and downregulated in animals with longer exposure.
*Tomruk et al. (2010)	Nonpregnant and pregnant New Zealand White rabbits exposed to GSM 1800 MHz RFR 15 min/day for a week	No oxidative damage in liver of exposed adult and offspring, increased lipid peroxidation.
Tran et al. (2023) (GE)	Lettuce plants (Lactuca sativa) exposed to 1800-1900 MHz (0.0008 mW/cm^2) and 2450 and 5000 MHz RFR (0.0002 mW/cm^2) from DECT base station and phones for 2-6 weeks	Down-regulation of two stress-related: violaxanthin de-epoxidase and zeaxanthin epoxidase.
Trivino Pardo et al (2012)	T-lymphoblastoid leukemia cells exposed to 900 MHz RFR for 2 or 48 h; SAR 9.0035 W/kg	Changes in gene expressions (e.g., an early activation of genes involved in DNA double- and single-strand breaks repair).
Trosic (2001)	Rats exposed to 2450 MHz RFR for 2, 8, 13 and 22 irradiation treatments of two hours each; power density 5-15 mW/cm^2 SAR 20 W/kg	Increased multinucleated alveolar macrophages- the elevation of the number of nuclei per cell was exposure time- and dose-dependent.
Trosic and Busljeta (2005)	Wistar rats exposed to continuous-wave 2450	The frequency of micronucleated bone

	MHz RFR 2 h/day 7 days /week for a total of 4, 16, 30, and 60 h. power density 5-10 mW/cm^2 SAR 1-2 W/kg	marrow erythrocytes was significantly increased after 15 irradiation treatments. No effect after 2, 8, and 30 exposure treatments.
Trosic and Busljeta (2006)	Rats exposed to 2450 MHz RFR 2 h/day, 15 days; SAR 1.24 W/kg	Bone marrow cell micronucleus frequency increased on experimental day 15, and micronucleated polychromatic erythrocytes in peripheral blood increased on day 8.
Trosic et al. (2002)	Male Wistar rats exposed for 2 h/day, 7 days a week for up to 30 days to continuous-wave 2450 MHz RFR; power density 5-10 mW/cm^2 SAR 1-2 W/kg	Increased micronuclei in peripheral blood polychromatic erythrocytes on the 2nd, 8th, and 15th day of exposure. It is likely that an adaptive mechanism, both in erythrocytopoiesis and genotoxicity occurred.
Trosic et al. (2004)	Male Wistar rats exposed for 2 h/day, 7 days/week for 4, 16, 30, and 60 h to continuous-wave 2450 MHz RFR; power density 5-10 mW/cm^2 SAR 1.25 W/kg	The frequency of micronucleated polychromatic erythrocytes in bone marrow was significantly increased on experimental day 15, but not on 2, 8, and 30 days.
Trosic et al. (2011)	Male Wistar rats exposed to GSM 915 MHz RFR for 1 h /day 7 days/week for 2 weeks; SAR 0.6 W/kg	Increased DNA single strand breaks (Comet assay) in brain, renal, and liver cells.
Tsybulin et al. (2013)	Japanese Quail embryos exposed in ovo to GSM 900 MHz signal from a cell phone intermittently (48 sec ON/12 sec OFF) during initial 38 h of brooding or for 158 h (120 h before brooding plus initial 38 h of brooding): SAR 0.000003 W/kg	The lower duration of exposure led to a significant ($p < 0.001$) decrease in DNA single strand breaks (Comet assay) in cells of 38-h embryos, while the higher duration of exposure resulted in a significant increase in DNA damage.
Usikalu et al. (2013)	Blood of rats exposed to 2450 MHz RFR for 10 min at 0.48-4.3 W/kg	Increased DNA strand breaks (Comet assay) at 2.39 W/kg. Hyperchromasia was observed in the ovary.
Üstündağ et al. (2020) (GE)	Zebrafish embryos exposed to 3000 MHz	Decreased tp53 and increased Casp3a gene expression

	RFR	
Vafaei et al. (2020) (LI) (GE)	Placenta of pregnant mice exposed to 2.4 GHz RFR from a D-link WiFi router; 2 or 4 h/day at 30 or 60 cm from router from 5 days after mating to 1 day before expected delivery, 0.09 W/kg at 30 cm	Increased gene expression of SOD, and GDKN1A, and GADD45a (DNA repair enzymes)
*Valbonesi et al. (2008) (GE)	Human trophoblast cell line HTR-8/SVneo exposed to pulsed 1817 MHz RFR or 1 h; SAR 2 W/kg	No significant change in either HSP70 or HSC70 protein or gene expression, or DNA single strand breaks (Comet assay).
Valbonesi et al. (2014) (GE)	Rat PC12 cells exposed to continuous-wave 1.8 GHz RFR or GSM-217 Hz and GSM-Talk signals for 4, 6, or24 h, SAR 2 W/kg	After PC12 cells exposure to the GSM-217 Hz signal for 16 or 24 h, HSP70 mRNA transcription significantly increased, whereas no effect was observed in cells exposed to the CW or GSM-Talk signals.
*Valbonesi et al. (2016) (GE)	Rat PC12 cells exposed to 1.8 GHz 217-GSM signal for 24 h. SAR 2 W/kg	AChE transcriptional or translational pathways not affected, whereas AChE enzymatic activity increased.
Vanishree et al. (2018)	Buccal cells from low and high cellular phone users	There was a significant increase in micronucleus counts in subjects who use the phone longer. There was highly significant difference in the mean micronucleus count of participants using (code division multiple access) CDMA than (global system for mobiles) GSM cellular phones.
Veerachari and Vasan. (2012)	Human semen exposed to radiation from a 900 MHz GSM mobile phone for 60 min at 1-40 $\mu W/cm^2$	Increased DNA fragmentation in sperm.
Verma and Dutta (1993) (LI) (GE)	pNGE7 cells exposed to 16-Hz modulated 915 MHz RFR at 0.00005 W/kg	Altered expression of neuron specific enolase gene.
*Verschaeve et al. (2006)	Female rats exposed to RFR for 2 h per day, 5 days per week for 2	No significant genotoxic activity of MX in blood and liver cells measured by micronucleus and DNA single strand breaks

	years; SAR 0.3 or 0.9 W/kg. the mutagen and carcinogen 3-chloro-4-(dichloromethyl)-5-hydroxy-2(5H)-furanone (MX) was given in the drinking water. at a concentration of 19 mug/ml.	(Comet assay). However, MX induced DNA damage in rat brain. Co-exposures to MX and RF radiation did not significantly increase the response of blood, liver and brain cells. (No data on RFR alone.)
Vian et al. (2006) (GE)	Tomato plants exposed to a 900-MHz RFR for 10 min at 0.0066 mW/cm^2	Induction of mRNA encoding the stress-related bZIP transcription factor (3.5 folds at 5-15 min post-exposure).
Vijayalaxmi et al. (1997a)	C3H/HeJ mice exposed to for 20 h/day, 7 days to continuous-wave 2450 MHz RFR MHz for 20 h/day. 7 days/week, over 18 months: SAR 1.0 W/kg	Significant increases in micronucleus formation in peripheral blood and bone marrow cells were observed.
*Vijayalaxmi et al. (1997b)	Blood lymphocytes from 2 human subjects exposed to 2450 MHz RFR for 90 min or intermittently at a total of 90 min (30 min on/30 min off) at 12.46 W/kg	RFR exposure had no effect on mitotic indices; incidence of cells showing chromosome damage; exchange aberrations; acentric fragments; binucleate lymphocytes, and micronuclei.
*Vijayalaxmi et al. (1999)	CF-1 male mice exposed to ultra-wideband electromagnetic radiation (UWBR) for 15 min; SAR 0.037 W/kg	No significant effects on micronucleus frequency and polychromatic erythrocytes in peripheral blood and bone marrow cells at 16 and 24 h post-exposure.
*Vijayalaxmi et al. (2000)	3 human peripheral blood samples exposed to pulsed 2450-MHz RFR for 2 h; SAR 2.135 W/kg	No significant effect on DNA single strand breaks (Comet assay) was observed in lymphocytes immediately and at 4 h post-exposure.
*Vijayalaxmi et al. (2001a)	4 human peripheral blood samples exposed to 835.62 MHz (FDMA) RFR for 24 h, SAR 4.4 or 5.0 W/kg	Lymphocytes were stimulated with a mitogen, phytohemagglutinin. No significant effects at 48 and 72 h post=exposure in mitotic indices, incidence of exchange aberrations, excess fragments, binucleate cells, and micronucleus frequency.

*Vijayalaxmi et al. (2001b)	Male Sprague-Dawley rats exposed to continuous-wave 2450 MHz RFR for 24 h; SAR 12 W/kg	Peripheral blood and bone marrow smears showed no effects on frequency of micronuclei in polychromatic erythrocytes at 24 h post-exposure. .
*Vijayalaxmi et al. (2001c)	4 human peripheral blood samples exposed to continuous-wave 847.74 MHz (CDMA) RFR for 24 h; SAR 4.9 or 5.5 W/kg	No significant effects on mitotic indices, frequencies of exchange aberrations, excess fragments, binucleate cells, and micronuclei in lymphocytes at 48 and 72 h post-exposure.
*Vijayalaxmi et al. (2003)	Timed-pregnant Fischer 344 rats (from nineteenth day of gestation) and their nursing offspring (until weaning) exposed to a far-field 1.6 GHz Iridium wireless communication signal for 2 h/day, 7 days/week for 2 years; SAR 0.036 to 0.077 W/kg	No significant effects on micronuclei in polychromatic erythrocytes in bone marrow.
*Vijayalaxmi et al. (2004)	Polychromatic erythrocytes of peripheral blood and bone marrow cells collected 24 h after treatment from mice exposed at the nasal region with 42.2 MHz RFR 30 min/day for 3 days, peak SAR 622 W/kg	No effect on micronucleus formation in both types of cells. RFR exposure did not affect micronucleus formation induced by cyclophosphamide.
*Vijayalaxmi et al. (2006)	Human peripheral blood samples exposed to 2.45 GHz or 8.2 GHz pulsed-wave RFR for 2 h; SAR 2.13 W/kg (245 MHz) or 20.71 W/kg (8.2 GHz),	No significant effects on chromosomal aberrations and micronuclei in lymphocytes.
Vilic et al. (2017)	Honeybee (Apis mellifera) larvae exposed to 900 MHz at field levels of 10, 23, 41 and 120 V m^{-1} for 2 h. At a field level of 23 V m^{-1}	DNA single strand breaks (Comet assay) increased significantly in honeybee larvae exposed to modulated (80% AM 1 kHz sinus) field at 23 V m^{-1}. Oxidative changes also observed. Modulated RF-EMF produced more negative effects than the corresponding

	the effect of 80% AM 1 kHz sinusoidal and 217 Hz modulation was investigated as well.	unmodulated field.
*Waldmann et al. (2013)	Human peripheral blood samples exposed to GSM 1800 MHz RFR for 28 h; SAR 0.2, 2, and 10 W/kg	No significant effects in lymphocytes on chromosome aberration, micronucleus frequency, sister chromatid exchange and DNA single strand breaks (Comet assay).
Wang X et al. (2015)	Neuro-2a (mouse neuroblastoma) cells exposed to GSM 900 MHz RFR for 24 h; SAR 0.5, 1 or 2 W/kg	Increased DNA oxidative damage (Comet assay) and reactive oxygen species. OGG1(a base excision DNA repair enzyme) may be involved.
Wang Y et al. (2022) (LI) (GE)	Drosophila melanogaster exposed to 3.5 GHz RFR for 2-3 days at 0.0026 W/kg, 0.026 W/kg, or 0.26 W/kg	In third-instar larvae, expression levels of the heat shock protein genes hsp22, hsp26 and hsp70 and humoral immune system genes AttC, TotC and TotA were all significantly increased
*Whitehead et al. (2005) (GE)	Murine embryonic fibroblasts exposed to 835.62 MHz FMCW, 847.74 MHz CDMA, or 836.55 MHz TDMA RFR at 5 or 10 W/kg	No effect on Fos proto-oncogene expression.
*Whitehead et al. (2006a) (GE)	Murine embryonic fibroblasts exposed to 835.62 MHz FMCW, 847.74 MHz CDMA RFR for 24 h at 5 W/kg	No statistically significant effect on gene expression by detection of level of mRNA
*Whitehead et al. (2006b) (GE)	Murine embryonic fibroblasts exposed to 835.62 MHz FMCW, 847.74 MHz CDMA RFR for 24 h at 5 W/kg	No statistically significant effect on gene expression (Gapd, Fos, Jun. Rasa3, hsp)
Wu H et al. (2012) (GE)	At Sertoli cells exposed to s-band microwaves for 4 min at 100 mW/cm^2	Upregulation of Bax/Bcl-2 and caspase-3 genes
Wu W et al. (2008)	Human lens epithelial cells exposed to 1800 MHz mobile phone radiation for 24 h; SAR 4 W/kg	Increased DNA single strand breaks (Comet assay) and reactive oxygen species.
Xu et al. (2010) (GE)	Sprague-Dawley rat primary cultured cortical neurons exposed to	Increased in the levels of 8-hydroxyguanine, a common biomarker of DNA oxidative damage, in the mitochondria of neurons,

	intermittent (5 min ON/10 min OFF) 217-Hz pulsed 1800 MHz RFR for 24 h; SAR 2 W/kg	levels of mitochondrial RNA (mtRNA) transcripts showed a reduction.
Xu et al. (2013)	Six different types of cells intermittently (5 min ON/10 min OFF) exposed to pulsed GSM 1800 MHz RFR for 1 or 24 h: SAR 3.0 W/kg	RFR induced DNA damage (γH2AX foci and alkaline and neutral Comet assay) in a cell type-dependent manner.
Yadav and Shama (2008)	Buccal-mucosa cells from 85 regular cell phone users (exposed) and 24 non-users (controls)	A positive correlation between 0-1, 1-2, 2-3 and 3-4 years of exposure and the frequency of micronucleated cells and total micronuclei.
Yakymenko et al. (2018)	Quail embryos exposed to GSM 1800 GHz signal from a smart phone (48 s ON/12 s OFF) for5 days before and 14 days during incubation, power density 0.00032 mW/cm^2	Increased DNA single sand breaks (Comet assay), oxidative DNA damage, reactive oxygen species, and mortality.
Yan et al. (2007) (GE)	Perm from rats exposed to RFR from a PCS CDMA (1900 MHz) mobile phone; two 3 h periods per day for 18 weeks	Increased mRNA od cell surface adhesion proteins CAD-1 and ICAM-1
Yan et al. (2008) (GE)	Adult Sprague-Dawley rats exposed to a cell phones 1.9 GHz (PCE CDMA) for 6 h per day for 126 days (18 weeks).	Significant mRNA up-regulation of injury-related proteins in the brain of rats exposed to cell phone radiation.
Yang et al. (2012) (GE)	Hippocampus of rats exposed to 2450 MHz RFR for 20 min at 6 W/kg	23 upregulated and 18 downregulated genes were identified including HSP27 and HSP70 genes
Yao et al. (2004)	Rabbit lens epithelial cells exposed to continuous-wave 2450-MHz RFR for 8 h, power densities 0.10, 0.25, 0.50, 1.00, and 2.00 mW/cm^2	The RFR higher than 0.50 mW/cm^2 can inhibit lens epithelial cell proliferation, and increase the expression of P27Kip1.

Yao et al. (2008)	Human lens epithelial cells intermittently (5 min ON/10 min OFF) exposed to GSM 1.8 GHz RFR for 2 h; SAR 1, 2, 3, and 4 W/kg	Increased DNA single strand breaks (Comet assay), no change in double strand breaks (γH2AX foci) and increased reactive oxygen species.
Ye et al. (2016)	Chicken embryos exposed to GSM 900 MHz RFR from a cell phones 3 h/day from day 2 to day 21 of incubation	Increased DNA single strand breaks (Comet assay) from blood cells and mortality.
*Yildirim et al. (2010)	People who lived around cell phone base stations and healthy controls	There was no significant difference in micronucleus frequency and chromosomal aberrations in blood lymphocytes between the two study groups.
Yu et al. (2023) (GE)	Rats exposed scrotally to 2605 MHz RFR, 6/hr per day for 50, 100, or 150 days, 1.05 W/kg	mRNA level of testicular androgen receptors increased in 50 day-exposed and decreased in 150 day-exposed group.
Zalata et al. (2015) (GE)	Human semen samples exposed to 850-MHz RFR from a cell phone for 1 h; SAR 1.46 W/kg at 10 cm	Significant increase in sperm DNA fragmentation percent, clusterin gene expression and clusterin protein (associated with clearance of cellular debris and apoptosis) levels in the exposed semen samples.
*Zeni et al. (2003)	Human peripheral blood exposed to continuous wave 925 MHz RFR or GSM 925 MHz (6 min ON/ 3 h OFF for 44h (SAR 1.6 W/kg); or GSM signal 1 h/day for 3 days (SAR 0.2 W/kg).	No statistically significant differences were detected in micronucleus frequency in lymphocytes.
*Zeni et al. (2005)	Human peripheral blood lymphocytes exposed to GSM 900 MHz signal for 2 h; SAR 0.3 and 1 W/kg	No significant effects on DNA single strand breaks (Comet assay), chromosome aberration, or sister chromatid exchange.
*Zeni et al. (2007)	Human blood exposed to 120 (0.4 W/kg) and 130 GHz (0.24, 1.4, or 2 W/kg) RFR for 20 min	No effect on Chromosomal damage (micronucleus) and DNA damage (Comet assay).
*Zeni et al. (2008)	Human peripheral blood exposed intermittently (6 min ON/2 h OFF) to	No significant effects on DNA single strand breaks (Comet assay) and micronucleus frequency in leukocytes.

	1945 MHz RFR for 24 – 68 h; SAR 2.2 W/kg	
Zeni et al. (2012a) (LI)	Human blood lymphocytes exposed to 1950 MHz UMTS RFR for 20 h at 1.25, 0.6, 0.3, or 0.15 W/kg, and treated with mitomycin C	RFR at 0.3 W/kg reduced micronucleus formation induced by mitomycin C.
*Zeni et al. (2012b)	Rat pheochromocytoma (PC12) neuron-like cells exposed to 1950 MHz UMTS RFR for24 h at 10 W/kg	No effect on DNA integrity.
Zeni et al. (2021) (LI)	Human SH-SY5Y neuroblastoma cells exposed to 1950 MHz UMTS RFR for 20 h at 0.3 W/kg	RFR direct exposure or the culture medium of exposed cells decreased menadione-induced DNA damage (Comet assay).
*Zhadobov et al. (2007)	Human U-251 MG glial cells exposed to 60 GHz RFR for 1-33 h at 5.4 or 0.54 mW/cm^2	The RFR did not modify stress-sensitive gene expression of chaperone proteins.
Zhang DY et al. (2006)	Chinese hamster lung cells exposed intermittently (5 min on/10 min off) to GSM 1800 MHz RFR for 1 or 24 h; SAR 3 W/kg	Cells exposed for 24 h showed increased DNA double strand breaks (γH2AX foci).
Zhang MB et al. (2002)	Human whole blood exposed to 2450 MHz RFR for 2 h; Power density 5 mW/cm^2	2450-MHz RFR cannot induce DNA and chromosome damage, but can increase DNA single strand breaks (Comet assay) induced by mitomycin C.
Zhang SZ et al. (2008) (GE)	Primary culture of rat neurons exposed to a 1800 MHz RFR for 24 h; SAR 2 W/kg.	Changes (up- and down-regulation) of many genes transcription (involving cytoskeleton, signal transduction pathway, metabolism, etc.) were observed.
Zhao L et al. (2022) (GE)	Rats exposed to 1500 MHz, 4300 MHz, or combination of the two RFRs at 10 mw/cm^2	Observed immune suppressive responses via regulating immune regulation and cellular metabolism-associated genes (noted in lymphocytes and spleen).
Zhao R. et al. (2007) (GE)	Rat neurons exposed to pulsed 217-Hz modulated 1800 MHz RFR for 24 h; SAR 2	up- and down-regulation of genes transcriptions were observed.

	W/kg	
Zhao TY. et al. (2007) (GE)	Primary cultured neurons and astrocytes exposed to a GSM 1900 MHz cell phone for 2 h	Up-regulation of caspase-2, caspase-6 and Asc (apoptosis associated speck-like protein containing a card) gene expression in neurons and astrocytes. Additionally, astrocytes showed up-regulation of the Bax gene. Neurons appeared to be more sensitive to this effect than astrocytes.
*Zhijian et al. (2009)	Leukocytes from four young healthy donors exposed intermittent (5 min ON/10 min OFF) to 1800 MHz RFR for 24 h; SAR 2 W/kg; Cell also exposed x-ray	No significant effect on DNA single strand breaks (Comet assay) and no synergistic effect with x-ray.
*Zhijian et al. (2010)	Human B-cell lymphoblastoid cells exposed to 1800 GHz RFR for 2 h; SAR 2 W/kg	RFR did not directly induce DNA single strand breaks (Comet assay)
*Ziemann et al. (2009)	Peripheral blood erythrocytes of B6C3F1 mice exposed to GSM 902 MHz or DCS 1747 MHz RFR 2 h/day, 5 days /week for 2 years; SAR 0.4, 1.3 and 4 W/kg	No significant effect on micronucleus frequency.
Zong et al. (2015) (LI)	Blood leukocytes exposed to 900 MHz RFR 4 h/day for 7 days at 0.05 W/kg and injected with bleomycin	RFR exposure reduced DNA single strand breaks (Comet assay) induced by bleomycin.
Zothansiama et al. (2017)	Blood samples from people lived closed to cell phone base station	The exposed group, residing within a perimeter of 80 m of mobile base stations, showed significantly higher frequency of micronuclei in lymphocytes when compared to the control group, residing 300 m away from the mobile base stations.
Zotti-Martelli et al. (2000)	Human peripheral blood lymphocytes exposed to 2.45 and 7.7 GHz RFR for 15, 30, or 60 min; power density 10, 20, or 30	Increased micronucleus frequency at a power density of 30 mW/cm^2 and after an exposure of 30 or 60 min.

	mW/cm^2	
Zotti-Martelli et al. (2005)	Human whole blood samples exposed to continuous-wave 1800 MHz RFR for for 60, 120, or 180 min; power density 5, 10, or 20 mW/cm^2	A statistically significant increase of micronucleus was observed in lymphocytes dependent on exposure time and applied power density.
*Zuo et al. (2015)	Sprague-Dawley rat spiral ganglion neurons exposed intermittently (5 min ON/10 min OFF) to GSM 1800 MHz RFR for 24 h; SAR 2 and 4 W/kg	The RFR could not directly induce DNA single strand breaks (Comet assay) in normal spiral ganglion neurons, but it could cause the changes of cellular ultrastructure at SAR 4.0 W/kg when cells are in fragile or micro-damaged condition.