

## **RFR Free Radical (Oxidative Damage) Studies**

**Of 288 total studies:                      E = 263 (91%)                      NE = 25 (9%)**

(E = reported effect    NE = reported no significant effect)

**(E) Abu Khadra KM, Khalil AM, Abu Samak M, Aljaberi A. Evaluation of selected biochemical parameters in the saliva of young males using mobile phones. *Electromagn Biol Med.* 2014 Feb 5. [Epub ahead of print]**

The biochemical status in the saliva of 12 males before/after using mobile phone has been evaluated. Radio frequency signals of 1800 MHz (continuous wave transmission, 217 Hz modulate and Global System for Mobile Communications [GSM - non-DTX]) with 1.09 w/kg specific absorption rate (SAR) value were used for 15 and 30 min. Cell phone radiation induced a significant increase of superoxide dismutase (SOD); there was a statistically significant effect of talking time on the levels of SOD,  $F(2, 33) = 8.084$ ,  $p < 0.05$ ,  $\omega = 0.53$ . The trend analysis suggests a significant quadratic trend,  $F(1, 33) = 4.891$ ,  $p < 0.05$ ; indicating that after 15 min of talking the levels of SOD increased, but as talking time increased the SOD activity started to drop. In contrast to this, there was no statistically significant effect of talking time on the level of salivary albumin, cytochrome c, catalase or uric acid. Results suggest that exposure to electromagnetic radiation may exert an oxidative stress on human cells as evidenced by the increase in the concentration of the superoxide radical anion released in the saliva of cell phone users.

**(E) Achudume A, Onibere B, Aina F, Tchokossa P. Induction of oxidative stress in male rats subchronically exposed to electromagnetic fields at non-thermal intensities. *J Electromagnetic Analysis and Applications* 2(8), 482-487, 2010. (LI)**

To investigate the oxidative stress-inducing potential of non-thermal electromagnetic fields in rats. Male Wister rats were exposed to electrical field intensity of  $2.3 \pm 0.82 \mu\text{V/m}$ . Exposure was in three forms: continuous waves, or modulated at 900 MHz or modulated GSM-nonDTX. The radio frequency radiation (RFR) was 1800 MHz, specific absorption radiation (SAR) (0.95-3.9 W/kg) for 40 and/or 60 days continuously. Control animals were located > 300 m from base station, while sham control animals were located in a similar environmental conditions, but in the vicinity of a non-functional base station. The rats were assessed for thiobarbituric and reactive species (TBARS), reduced glutathione (GSH) content, catalase activity, glutathione reductase (GR) and glucose residue after 40 and 60 days of exposure. At

40 days, electromagnetic radiation failed to induce any significant alterations. However, at 60 days of exposure various attributes evaluated decreased. The respective decreases in both nicotinamide adenine dinucleotide phosphate (NADPH) and Ascorbate-linked lipid peroxidation (LPO) with concomitant diminution in enzymatic antioxidative defense systems resulted in decreased glucose residue. The present studies showed some biochemical changes that may be associated with a prolong exposure to electromagnetic fields and its relationship to the activity of antioxidant system in rat Regular assessment and early detection of antioxidative defense system among people working around the base stations are recommended.

**(E) 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(4) 1318-1325, 2009.**

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) Ahmed NA, Radwan NM, Aboul Ezz HS, Salama NA. The antioxidant effect of Green Tea Mega EGCG against electromagnetic radiation-induced oxidative stress in the hippocampus and striatum of rats. Electromagn Biol Med. 2017;36(1):63-73, 2017.**

Electromagnetic radiation (EMR) of cellular phones may affect biological systems by increasing free radicals and changing the antioxidant defense systems of tissues, eventually leading to oxidative stress. Green tea has recently attracted significant attention due to its health benefits in a variety of disorders, ranging from cancer to weight loss. Thus, the aim of the present study was to investigate the effect of EMR (frequency 900 MHz modulated at 217 Hz, power density 0.02 mW/cm<sup>2</sup>, SAR 1.245 W/kg) on different oxidative stress parameters in the hippocampus and striatum of adult rats. This study also extends to evaluate the therapeutic effect of green tea mega EGCG on the previous parameters in

animals exposed to EMR after and during EMR exposure. The experimental animals were divided into four groups: EMR-exposed animals, animals treated with green tea mega EGCG after 2 months of EMR exposure, animals treated with green tea mega EGCG during EMR exposure and control animals. EMR exposure resulted in oxidative stress in the hippocampus and striatum as evident from the disturbances in oxidant and antioxidant parameters. Co-administration of green tea mega EGCG at the beginning of EMR exposure for 2 and 3 months had more beneficial effect against EMR-induced oxidative stress than oral administration of green tea mega EGCG after 2 months of exposure. This recommends the use of green tea before any stressor to attenuate the state of oxidative stress and stimulate the antioxidant mechanism of the brain.

**(E) Akbal, A., Kiran, Y., Sahin, A., Turgut-Balik, D., Balik HH. Effects of electromagnetic waves emitted by mobile phones on germination, root growth, and root tip cell mitotic division of lens culinaris medik. Pol. J. Environ. Stud. 21:23–29, 2012.**

In this study, the effects of electromagnetic waves emitted from mobile phones operating at 1800 MHz were investigated on germination, root growth and mitotic division of root tips of *Lens culinaris Medik*. Seeds were split into three groups. The first group was exposed to a mobile phone electromagnetic field for 48 hours at the state of dormancy, and the second group was exposed to the same electromagnetic field at the state of division. The third group, the control group, was not exposed to an electromagnetic field beyond the natural background. The results obtained in the study indicate that electromagnetic waves emitted from mobile phones affect seeds in the state of dormancy more than the state of germination. Germination rate was not affected under the specified exposure conditions, but root growth decreased due to a possible effect of oxidative stress in the state of dormant seeds. There was also a noticeable increment in the c-mitosis rates, especially in the state of dormant seeds. The reason for this increment could be problems in spindle function.

**(E) Akakin D, Tok OE, Anil D, Akakin A, Sirvanci S, Sener G, Ercan F. Electromagnetic Waves from Mobile Phones may Affect Rat Brain During Development. Turk Neurosurg 2021;31(3):412-421.**

**Aim:** To investigate the effects of electromagnetic waves (EMWs) from mobile phones (MPs) on rat brains of rats by morphological and biochemical analysis. **Material and methods:** EMW was applied for two hours/day until birth in stand-by fetal and EMW fetal groups and postnatal 60 <sup>th</sup> day in stand-by and EMW groups. The control group was not exposed to MP. On postnatal 60 <sup>th</sup> day, brain malondialdehyde (MDA) and glutathione (GSH) levels were measured, and western blot analysis was performed to determine glial fibrillary acidic protein (GFAP) content. Hematoxylin and eosin staining and GFAP immunohistochemistry were applied. Trigeminal nerves were examined using the transmission electron microscope. **Results:** In comparison to controls, rats exposed to MP in stand-by or talk modes had significantly increased neuronal damage in the cortex and hippocampus. Increased MDA levels in the EMW group and decreased GSH levels in the stand-by, EMW fetal and EMW groups were found compared with controls. Increased GFAP content in the EMW group and increased GFAP staining in the

EMW fetal and EMW groups compared to controls were observed. EMW group had a significantly decreased number of myelinated axons than control animals. **Conclusion:** The results of this study suggests that 1800 MHz EMWs (SAR=1.79 W/kg) exposure in the prenatal and early postnatal life may lead to trigeminal nerve damage in addition to oxidative stress-induced neuronal degeneration and astroglial activation in the rat brain. Effects seem to be mode related, being more detrimental in groups exposed to MP during talk mode

**(E) Akbari A, Jelodar G, Nazifi S. Vitamin C protects rat cerebellum and encephalon from oxidative stress following exposure to radiofrequency wave generated by a BTS antenna model. Toxicol Mech Methods. 24(5):347-352, 2014.**

Radio frequency wave (RFW) generated by base transceiver station has been reported to produce deleterious effects on the central nervous system function, possibly through oxidative stress. This study was conducted to evaluate the effect of RFW-induced oxidative stress in the cerebellum and encephalon and the prophylactic effect of vitamin C on these tissues by measuring the antioxidant enzymes activity, including: glutathione peroxidase, superoxide dismutase, catalase, and malondialdehyde (MDA). Thirty-two adult male Sprague-Dawley rats were randomly divided into four equal groups. The control group; the control-vitamin C group received L-ascorbic acid (200 mg/kg of body weight/day by gavage) for 45 days. The RFW group was exposed to RFW and the RFW+ vitamin C group was exposed to RFW and received vitamin C. At the end of the experiment, all groups were killed and encephalon and cerebellum of all rats were removed and stored at -70 °C for measurement of antioxidant enzymes activity and MDA. The results indicate that exposure to RFW in the test group decreased antioxidant enzymes activity and increased MDA compared with the control groups ( $p < 0.05$ ). The protective role of vitamin C in the treated group improved antioxidant enzymes activity and reduced MDA compared with the test group ( $p < 0.05$ ). It can be concluded that RFW causes oxidative stress in the brain and vitamin C improves the antioxidant enzymes activity and decreases MDA.

**(E) Akoev IG, Pashovkina MS, Dolgacheva LP, Semenova TP, Kalmykov VL. [Enzymatic activity of some tissues and blood serum from animals and humans exposed to microwaves and hypothesis on the possible role of free radical processes in the nonlinear effects and modification of emotional behavior of animals]. [Article in Russian] Radiats Biol Radioecol. 42(3):322-330, 2002. (LI)**

The dependence of activities of actomyosin ATPase, alkaline phosphatase, aspartataminotransferase, monoaminoxidase and that of affective rat behavior on frequency of modulation of microwaves (0.8-10 microW/cm<sup>2</sup>) was explored at short-time actions. Series of nonlinear phenomena, inexplicable from positions of the energy approaches are revealed, The working hypothesis explaining opportunity of high performance of weak and super-weak microwaves and other revealed phenomena by resonance interaction of such electromagnetic radiofrequency radiation with paramagnetic molecules of biological tissues was proposed. This resonance interaction activate free radicals and initiate auto-supporting and auto-intensifying of chain chemical reactions. The spontaneous autocatalytic oxidation of catecholamines enlarges a common

pool of free radicals, capable to participate in such enhanced generating. The protective role of monoaminoxidase is postulated. Monoaminoxidase is basically located on an outer surface of mitochondrias and it is deaminating monoamines. The deaminating prevents penetration of catecholamines inside of mitochondrias and their quinoid oxidation there with formation of free-radical semi-quinons, capable to destroy system of ATP synthesis. These inferences are obliquely confirmed by the experimentally revealed correlation between activity of monoaminoxidase and integrative activity of the rat brain.

**(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, 2019.**

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, 2019.**

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) Almášiová V, Holovská K, Andrašková S, Cigánková V, Ševčíková Z, Raček A, Andrejčáková Z, Beňová K, Tóth S, Tvrdá E, Molnár J, Račková E. Potential influence of prenatal 2.45 GHz radiofrequency electromagnetic field exposure on Wistar albino rat testis. *Histol Histopathol* 2021 Mar 25;18331.**

An ever-increasing use of wireless devices over the last decades has forced scientists to clarify their impact on living systems. Since prenatal development is highly sensitive to numerous noxious agents, including radiation, we focused on the assessment of potential adverse effects of microwave radiation (MR) on testicular development. Pregnant Wistar albino rats (3 months old, weighing  $282 \pm 8$  g) were exposed to pulsed MR at a frequency of 2.45 GHz, mean power density of  $2.8 \text{ mW/cm}^2$ , and a specific absorption rate of  $1.82 \text{ W/kg}$  for 2 hours/day throughout pregnancy. Male offspring were no longer exposed to MR following birth. Samples of biological material were collected after reaching adulthood (75 days). In utero MR exposure caused degenerative changes in the testicular parenchyma of adult rats. The shape of the seminiferous tubules was irregular, germ cells were degenerated and often desquamated. The diameters of the seminiferous tubules and the height of the germinal epithelium were significantly decreased (both at  $**p < 0.01$ ), while the interstitial space was significantly increased ( $**p < 0.01$ ) when compared to the controls. In the group of rats prenatally exposed to MR, the somatic and germ cells were rich in vacuoles and their organelles were often altered. Necrotizing cells were more frequent and empty spaces between Sertoli cells and germ cells were observed. The Leydig cells contained more lipid droplets. An increased Fluoro Jade - C and superoxide dismutase 2 positivity was detected in the rats exposed to MR. Our results confirmed adverse effects of MR on testicular development.

**(cancer) Anghileri LJ, Mayayo E, Domingo JL. Iron-radiofrequency synergism in lymphomagenesis. *Immunopharmacol Immunotoxicol*. 28(1):175-183, 2006.**

The parenteral **iron** administration effects on the acceleration of lymphomagenesis by radiofrequency exposure were investigated using an animal model that develops spontaneous lymphomas with ageing. Complementary studies of the in vivo uptake of  $^{59}\text{Fe}$ -labeled ferric gluconate and ferric-ATP complex showed differences of absorption and excretion between both **iron** compounds. In vitro assays of their effects on calcium cellular uptake using a cell model and tissues homogenates showed a molecular structure-dependence. The current results

(mortality, clinical and histopathological examinations) demonstrated a synergism between radiofrequency and ferric gluconate, and the increased risk of radiofrequency exposure when it is simultaneous to parenteral **iron** administration.

**(E) Arbabi-Kalati F, Salimi S, Vaziry-Rabiee A, Noraei M. Effect of mobile phone usage time on total antioxidant capacity of saliva and salivary immunoglobulin a. Iran J Public Health. 43(4):480-484, 2014.**

**BACKGROUND:** Nowadays mobile phone is very popular, causing concern about the effect it has on people's health. Parotid salivary glands are in close contact to cell phone while talking with the phone and the possibility of being affected by them. Limited studies have evaluated the effect of cell phone use on the secretions of these glands; so this study was designed to investigate the effects of duration of mobile phone use on the total antioxidant capacity of saliva. **METHODS:** Unstimulated saliva from 105 volunteers without oral lesions collected. The volunteers based on daily usage of mobile phones were divided into three groups then total antioxidant capacity of saliva was measured by Ferric Reducing Ability of Plasma (FRAP) method. Data were analyzed by SPSS software version 19. ANOVA was used to compare 3 groups and post-hoc Tukey test to compare between two groups. **RESULTS:** Average total antioxidant capacities of saliva in 3 groups were 657.91  $\mu\text{mol/lit}$ , 726.77  $\mu\text{m/lit}$  and 560.17  $\mu\text{mol/lit}$ , respectively. The two groups had statistically significant different ( $P = 0.039$ ). **CONCLUSION:** Over an hour talking with a cell phone decreases total antioxidant capacity of saliva in comparison with talking less than twenty minutes.

**(E) Asl JF, Goudarzi M, Shoghi H. The radio-protective effect of rosmarinic acid against mobile phone and Wi-Fi radiation-induced oxidative stress in the brains of rats. Pharmacol Rep. 72:857-866, 2020.**

**BACKGROUND:** Rosmarinus officinalis L. is an aromatic perennial herb from which rosmarinic acid (RA) can be extracted. This research was conducted to assess the effectiveness of RA against radio frequency (RF) radiation-induced oxidative stress due to 915 MHz (mobile phone) and 2450 MHz (Wi-Fi) frequencies in rats. **METHODS:** The animals were separated into six groups, including group 1 receiving normal saline (NS), group 2 (NS/Wi-Fi) and group 4 (NS/mobile), which received NS plus 60 min/day of exposure to the electromagnetic radiation (EMR) for 1 month, group 3 (RA/Wi-Fi) and group 5 (RA/mobile) received RA (20 mg/kg/day, po) plus 60 min/day of EMR, and group 6 (RA) received only RA. **RESULTS:** There was a significant elevation of protein carbonylation (PC), nitric oxide (NO) and malondialdehyde (MDA) and significant reduction in glutathione (GSH), glutathione peroxidase (GPx), total antioxidant capacity (TAC), superoxide dismutase (SOD) and catalase (CAT) in the RF radiation-exposed rats' brain compared to the control group. RA reduced the levels of NO, PC and MDA and it also elevated the TAC, GPx, SOD, CAT and GSH levels in the rats' brains in the RA/Wi-Fi and RA/mobile groups compared to the NS/Wi-Fi and NS/mobile groups, respectively. **CONCLUSION:** It can be concluded that RA can be considered a useful candidate for protecting brain tissues against RF radiation-induced oxidative stress at 915 and

2450 MHz frequencies through ameliorative effects on the antioxidant enzyme activities and oxidative stress indices.

**(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(2): 223-229, 2013. (LI)**

**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) \*Avcı B, Akar A, Bilgici B, Tunçel ÖK. Oxidative stress induced by 1.8 GHz radio frequency electromagnetic radiation and effects of garlic extract in rats. Int J Radiat Biol. 88(11):799-805, 2012.**

**PURPOSE:** We aimed to study the oxidative damage induced by radiofrequency electromagnetic radiation (RF-EMR) emitted by mobile telephones and the protective effect of garlic extract used as an anti-oxidant against this damage. **MATERIALS AND METHODS:** A total of 66 albino Wistar rats were divided into three groups. The first group of rats was given 1.8 GHz, 0.4 W/kg specific absorption rate (SAR) for 1 h a day for three weeks. The second group was given 500 mg/kg garlic extract in addition to RF-EMR. The third group of rats was used as the control group. At the end of the study, blood and brain tissue samples were collected from the rats. **RESULTS:** After the RF-EMR exposed, the advanced oxidation protein product (AOPP) levels of brain tissue increased compared with the control group ( $p < 0.001$ ). Garlic administration accompanying the RF-EMR, on the other hand, significantly reduced AOPP levels in brain tissue ( $p < 0.001$ ). The serum nitric oxide (NO) levels significantly increased both in the first and second group ( $p < 0.001$ ). However, in the group for which garlic administration accompanied that of RF-EMR, there was no difference in serum NO levels compared with the RF-EMR exposed group ( $p > 0.05$ ). There was no significant difference among the groups with respect to malondialdehyde (MDA) levels in brain tissue and blood samples ( $p > 0.05$ ). Similarly, no difference was detected among the groups regarding serum paroxonase (PON) levels ( $p > 0.05$ ). We did not detect any PON

levels in the brain tissue. **CONCLUSIONS:** The exposure of RF-EMR similar to 1.8 GHz Global system for mobile communication (GSM) leads to protein oxidation in brain tissue and an increase in serum NO. We observed that garlic administration reduced protein oxidation in brain tissue and that it did not have any effects on serum NO levels.

**(E) Aweda MA, Gbenebitse S, Meidinyo RO. Effects of 2.45 GHz microwave exposures on the peroxidation status in Wistar rats. Niger Postgrad Med J. 10(4):243-246, 2003.**

One of the consequences of exposures to microwave (MW) radiations is the enhanced production of free O<sub>2</sub>, free radicals, peroxides and superoxides. The effects on the lipid peroxidation status (LPS) of whole body irradiation of 120 Wistar rats with 2.45 GHz MW at a power density of 6mWcm<sup>-2</sup> have been studied using the MW generator model ER6660E from Toshiba UK Ltd. The LPS in the rats was monitored for a period of 8 weeks post irradiation using thiobarbituric acid (TRA) method. The MW exposures caused an increase in the LPS from the mean control value of 4.18 x 10<sup>-6</sup>g l<sup>-1</sup> to a maximum of 6.50 x 10<sup>-6</sup> g l<sup>-1</sup> within the first 24 hrs, and then gradually reduced to control value after about a week. 1mg kg<sup>-1</sup> of ascorbic acid administered before irradiation caused a decrease in the LPS from the control value to a minimum of 2.86 x 10<sup>-6</sup>g l<sup>-1</sup> within the first week. The value then gradually rose to a maximum of 3.96 x 10<sup>-6</sup>g l<sup>-1</sup> within the monitoring period. 1 mg kg<sup>-1</sup> of α-tocopherol also administered before irradiation also caused a decrease in the LPS from the control value to a minimum of 2.10 x 10<sup>-6</sup> g l<sup>-1</sup> within the first week. The value then gradually rose to a maximum of 3.94 x 10<sup>-6</sup> g l<sup>-1</sup> within the monitoring period. The results obtained from this study demonstrate that MW exposures cause significant increase in the LPS and there are protective effects of the anti-oxidants ascorbic acid and alpha-tocopherol.

**(E) Ayata A, Mollaoglu H, Yilmaz HR, Akturk O, Ozguner F, Altuntas I. Oxidative stress-mediated skin damage in an experimental mobile phone model can be prevented by melatonin. J Dermatol. 31(11):878-883, 2004.**

Most mobile phones emit 900 MHz of radiation that is mainly absorbed by the external organs. The effects of 900 MHz of radiation on fibrosis, lipid peroxidation, and anti-oxidant enzymes and the ameliorating effects of melatonin (Mel) were evaluated in rat skin. Thirty Wistar-Albino rats were used in the study. The experimental groups were the control group, the irradiated group (IR), and the irradiated+Mel treated group (IR+Mel). A dose of 900 MHz, 2 W radiation was applied to the IR group every day for 10 days (30 min/day). The IR+Mel group received 10 mg/kg/day melatonin in tap water for 10 days before the irradiation. At the end of the 10th day, a skin specimen was excised from the thoracoabdominal area. The levels of malondialdehyde (MDA) and hydroxyproline and the activities of superoxide dismutase (SOD), glutathione peroxidase (GSH-Px), and catalase (CAT) were studied in the skin samples. MDA and hydroxyproline levels and activities of CAT and GSH-Px were increased significantly in the IR group compared to the control group (p<0.05) and decreased significantly in the IR+Mel group (p<0.05). SOD activity was decreased significantly in the IR group and this decrease was not prevented by the Mel treatment. These results suggest that rats irradiated with 900 MHz suffer from increased fibrosis and lipid peroxidation (LPO). Mel treatment can reduce the fibrosis and LPO caused by radiation.

**(E) \*Aydin B, Akar A. Effects of a 900-MHz electromagnetic field on oxidative stress parameters in rat lymphoid organs, polymorphonuclear leukocytes and plasma. Arch Med Res. 42(4):261-267, 2011.**

BACKGROUND AND AIMS: The present study investigated the effects of a 900-MHz electromagnetic field (EMF) for 2 h/day for 45 days on lymphoid organs (spleen, thymus, bone marrow), polymorphonuclear leukocytes (PMNs) and plasma of rats, focusing on changes in the enzymatic and nonenzymatic antioxidant system. We determined whether there is any difference between immature and mature rats in terms of oxidative damage caused by EMF and tested recovery groups to determine whether EMF-induced damage is reversible in immature and mature rats. METHODS: Twenty four immature and 24 mature rats were divided randomly and equally into six groups as follows: two control groups, immature (2 weeks old) and mature (10 weeks old); two groups were exposed to 900 MHz ( $28.2 \pm 2.1$  V/m) EMF for 2 h/day for 45 days. Two recovery groups were kept for 15 days after EMF exposure. RESULTS: Substantial, deleterious biochemical changes were observed in oxidative stress metabolism after EMF exposure. Antioxidant enzyme activity, glutathione levels in lymphoid organs and the antioxidant capacity of the plasma decreased, but lipid peroxidation and nitric oxide levels in PMNs and plasma and also myeloperoxidase activity in PMNs increased. Oxidative damage was tissue specific and improvements seen after the recovery period were limited, especially in immature rats. CONCLUSIONS: In the present study, much higher levels of irreversible oxidative damage were observed in the major lymphoid organs of immature rats than in mature rats.

**(E) \*Aynali G, Nazırođlu M, Celik O, Dođan M, Yarıktas M, Yasan H. Modulation of wireless (2.45 GHz)-induced oxidative toxicity in laryngotracheal mucosa of rat by melatonin. Eur Arch Otorhinolaryngol. 270(5):1695-1700, 2013. (LI)**

It is well known that oxidative stress induces larynx cancer, although antioxidants induce modulator role on etiology of the cancer. It is well known that electromagnetic radiation (EMR) induces oxidative stress in different cell systems. The aim of this study was to investigate the possible protective role of melatonin on oxidative stress induced by Wi-Fi (2.45 GHz) EMR in laryngotracheal mucosa of rat. For this purpose, 32 male rats were equally categorized into four groups, namely controls, sham controls, EMR-exposed rats, EMR-exposed rats treated with melatonin at a dose of 10 mg/kg/day. Except for the controls and sham controls, the animals were exposed to 2.45 GHz radiation during 60 min/day for 28 days. The lipid peroxidation levels were significantly ( $p < 0.05$ ) higher in the radiation-exposed groups than in the control and sham control groups. The lipid peroxidation level in the irradiated animals treated with melatonin was significantly ( $p < 0.01$ ) lower than in those that were only exposed to Wi-Fi radiation. The activity of glutathione peroxidase was lower in the irradiated-only group relative to control and sham control groups but its activity was significantly ( $p < 0.05$ ) increased in the groups treated with melatonin. The reduced glutathione levels in the mucosa of rat did not change in the four groups. There is an apparent protective effect of melatonin on the Wi-Fi-induced oxidative stress in the laryngotracheal mucosa of rats by inhibition of free radical formation and support of the glutathione peroxidase antioxidant system.

**(E) Azimzadeh M, Jelodar G. The protective effect of vitamin supplementation (E and E + C) on passive avoidance learning and memory during exposure to 900 MHz RFW emitted from BTS. Toxicol Ind Health. 2020 Feb; 36(2):93-98.**

Deleterious effects of exposure to electromagnetic radiation on public health have been widely studied. This study was conducted to evaluate the protective effect of vitamin supplementation (E or E + C) on passive avoidance learning (PAL) and memory in rats subjected to 900 MHz radiofrequency waves (RFW). Thirty adult male Sprague-Dawley rats ( $190 \pm 20$  g) were randomly divided into six groups as: control I (vehicle), control II (vitamin E 250 mg/kg), control III (vitamin E 100 mg/kg + l-ascorbic acid 200 mg/kg), and three exposed groups to RFW as: sham-exposed, treatment I (vitamin E), and treatment II (vitamin E + C). The duration of exposure was 30 continuous days (4 h/day). The PAL was evaluated on the last day by the shuttle box. Learning and memory of animals demonstrated as the duration of remaining within the light area, which is called the light time (LT). The sham-exposed group showed a significant decrease in LT on the learning, consolidation, and retention days compared to other groups ( $p < 0.05$ ). Pretreatment with vitamins (E and E + C) could protect PAL against adverse effects of RFW, and the administration of vitamin E + C improved PAL performance in control III compared to control I and treatment II groups ( $p < 0.05$ ). Administration of vitamin E + C to exposed group (treatment II) caused a significant increase in LT on the learning ( $p = 0.013$ ), consolidation, and retention ( $p = 0.009$ ) sessions compared to the treatment group I (vitamin E). Long-term exposure to 900 MHz RFW impaired PAL and memory, and pretreatment of vitamin (E or E + C) prevented these effects, which may be a new potential mechanism against side effects of RFW.

**(E) Azimzadeh M, Jelodar G. Trace elements homeostasis in brain exposed to 900 MHz RFW emitted from a BTS-antenna model and the protective role of vitamin E. J Anim Physiol Anim Nutr (Berl). 2020 Apr 12. doi: 10.1111/jpn.13360. [Epub ahead of print]**

Advances in telecommunication and their broad usage in the community have become a great concern from the health aspect. The object of the present study was to examine the effects of exposure to 900 MHz RFW on brain Iron (Fe), Copper (Cu), Zinc (Zn) and Manganese (Mn) concentration, and the protective role of pre-treatment of vitamin E on mentioned elements homeostasis. Twenty adult male Sprague-Dawley rats ( $200 \pm 20$  g) randomly were divided into four groups. Control group (without any exposure, received distilled water), treatment control group (orally received 250 mg/kg BW/d vitamin E), treatment group (received 250 mg/kg BW/d vitamin E and exposed to 900 MHz RFW) and sham-exposed group (exposed to 900 MHz RFW). Animals (with freely moving in the cage) were exposed to RFW for 30 consecutive days (4 hr/day). The levels of the above mentioned elements in the brain tissue were determined on the last day using atomic absorption spectrophotometry. Exposure to 900 MHz RFW induced a significant increase in the Fe, Cu, Mn levels and Cu/Zn ratio accompanied by a significant decrease in Zn level in the sham-exposed group compare to control group. Vitamin E pre-treatment improved the level of Fe, Cu, Mn and Cu/Zn ratio, except in the Zn concentration. Exposure to 900 MHz RFW caused disrupted trace elements homeostasis in the brain tissue and administration of vitamin E as an antioxidant and neuroprotective agent improved the situation.

**(E) Bahreyni Toossi MH, Sadeghnia HR, Mohammad Mahdizadeh Feyzabadi M, Hosseini M, Hedayati M, Mosallanejad R, Beheshti F, Alizadeh Rahvar Z. Exposure to mobile phone (900-1800 MHz) during pregnancy: tissue oxidative stress after childbirth. J Matern Fetal Neonatal Med. 31(10):1298-1303, 2018.**

**BACKGROUND:** The present study has investigated the effects of **mobile phone** (900-1800 MHz)-induced electromagnetic radiation on redox status in the heart, liver, kidney, cerebellum, and hippocampus of dams and the offspring mice. **MATERIALS AND METHODS:** Pregnant Balb/C were divided into two groups including the control and the experimental group. The experimental group was exposed to **mobile phone** (900-1800 MHz), during pregnancy (2 h/d for 20 d). The dams and the offspring of both groups were sacrificed and tissues of interest were harvested immediately after delivery. Malondialdehyde (MDA) concentration, total thiol groups (TTG) content, superoxide dismutase (SOD), and catalase (CAT) activities were determined in the tissues. **RESULTS:** In the experimental groups, MDA levels were significantly increased, while TTG, SOD, and CAT were significantly decreased in the total tissues of dams and their offspring. **CONCLUSION:** Exposure to **mobile phone** (900-1800 MHz) during pregnancy induced oxidative stress in tissues of dams and their offspring.

**(E) Balci M, Devrim E, Durak I. Effects of mobile phones on oxidant/antioxidant balance in cornea and lens of rats. Curr Eye Res. 32(1):21-25, 2007.**

**Purpose:** To investigate the effects of mobile-phone-emitted radiation on the oxidant/antioxidant balance in corneal and lens tissues and to observe any protective effects of vitamin C in this setting. **Methods:** Forty female albino Wistar rats were assigned to one of four groups containing 10 rats each. One group received a standardized daily dose of mobile phone radiation for 4 weeks. The second group received this same treatment along with a daily oral dose of vitamin C (250 mg/kg). The third group received this dose of vitamin C alone, while the fourth group received standard laboratory care and served as a control. In corneal and lens tissues, malondialdehyde (MDA) levels and activities of superoxide dismutase (SOD), glutathione peroxidase (GSH-Px), and catalase (CAT) were measured with spectrophotometric methods. **Results:** In corneal tissue, MDA level and CAT activity significantly increased in the mobile phone group compared with the mobile phone plus vitamin C group and the control group ( $p < 0.05$ ), whereas SOD activity was significantly decreased ( $p < 0.05$ ). In the lens tissues, only the MDA level significantly increased in the mobile phone group relative to mobile phone plus vitamin C group and the control groups ( $p < 0.05$ ). In lens tissue, significant differences were not found between the groups in terms of SOD, GSH-Px, or CAT ( $p > 0.05$ ). **Conclusions:** The results of this study suggest that mobile telephone radiation leads to oxidative stress in corneal and lens tissues and that antioxidants such as vitamin C can help to prevent these effects.

**(E) Balci M, Namuslu M, Devrim E, Durak I. Effects of computer monitor-emitted radiation on oxidant/antioxidant balance in cornea and lens from rats. Mol Vis. 15:2521-2525, 2009.**

**PURPOSE:** This study aims to investigate the possible effects of computer monitor-emitted radiation on the oxidant/antioxidant balance in corneal and lens tissues and to observe any protective effects of vitamin C (vit C). **METHODS:** Four groups (PC monitor, PC monitor plus vitamin C, vitamin C, and control) each consisting of ten Wistar rats were studied. The study lasted for three weeks. Vitamin C was administered in oral doses of 250 mg/kg/day. The computer and computer plus vitamin C groups were exposed to computer monitors while the other groups were not. Malondialdehyde (MDA) levels and superoxide dismutase (SOD), glutathione peroxidase (GSH-Px), and catalase (CAT) activities were measured in corneal and lens tissues of the rats. **RESULTS:** In corneal tissue, MDA levels and CAT activity were found to increase in the computer group compared with the control group. In the computer plus vitamin C group, MDA level, SOD, and GSH-Px activities were higher and CAT activity lower than those in the computer and control groups. Regarding lens tissue, in the computer group, MDA levels and GSH-Px activity were found to increase, as compared to the control and computer plus vitamin C groups, and SOD activity was higher than that of the control group. In the computer plus vitamin C group, SOD activity was found to be higher and CAT activity to be lower than those in the control group. **CONCLUSION:** The results of this study suggest that computer-monitor radiation leads to oxidative stress in the corneal and lens tissues, and that vitamin C may prevent oxidative effects in the lens.

**(E) Bansal D, Chhapparwal Y, Pai KM, Kumar M, Vineetha R, Chhapparwal S, Kamath S, Kamath A. Effect of Duration of Mobile Phone Use on the Salivary Flow and Total Antioxidant Capacity of Saliva and Salivary Immunoglobulin A Level: A Cross-sectional Study. J Int Soc Prev Community Dent. 2022 Apr 8;12(2):260-265.**

**Introduction:** The objective of this study was to observe the effects of the duration of handheld mobile phone usage on the salivary flow, salivary immunoglobulin A (IgA) level, and salivary markers for oxidative stress. **Materials and methods:** Eighty-one students were categorized into three groups based on their duration of mobile phone usage after age- and gender-matching. Students were grouped as: mobile phone usage <20 min/day (Group A), mobile phone usage 20-60 min/day (Group B), and mobile phone usage >60 min/day (Group C). Saliva was collected to evaluate salivary flow rate, salivary IgA level, and salivary markers for oxidative stress. **Results:** The salivary flow rate showed no statistically significant difference between the three groups ( $P = 0.180$ ). There was no statistically significant difference in the salivary IgA between the three groups ( $P = 0.237$ ). There was a statistically significant difference in the malondialdehyde (MDA) level between the three groups ( $P = 0.042$ ). On pair comparison between the groups, group B and group C had a statistically significant difference ( $P = 0.019$ ) in the MDA level. There was no statistically significant difference in the salivary thiol level between the three groups ( $P = 0.237$ ). **Conclusion:** The duration of handheld mobile phone usage did not show any significant effects on the salivary flow rate, salivary IgA, and thiol levels. There was an increase in the salivary MDA concentration in subjects using handheld mobile phones for a longer duration, indicating higher oxidative stress in salivary glands exposed to mobile phone radiofrequency electromagnetic waves for a longer duration.

**(E) Barteri M, De Carolis R, Marinelli F, Tomassetti G, Montemiglio LC. Effects of microwaves (900 MHz) on peroxidase systems: a comparison between lactoperoxidase and horseradish peroxidase. Electromagn Biol Med. 2015 Jan 12:1-7. [Epub ahead of print]**

This work shows the effects of exposure to an electromagnetic field at 900 MHz on the catalytic activity of the enzymes lactoperoxidase (LPO) and horseradish peroxidase (HRP). Experimental evidence that irradiation causes conformational changes of the active sites and influences the formation and stability of the intermediate free radicals is documented by measurements of enzyme kinetics, circular dichroism spectroscopy (CD) and cyclic voltammetry.

**(E) Bedir R, Tumkaya L, Mercantepe T, Yilmaz A. Pathological Findings Observed in the Kidneys of Postnatal Male Rats Exposed to the 2100 MHz Electromagnetic Field. Arch Med Res 2018 Oct;49(7):432-440.**

**Background:** The widespread use by young people of modern communication devices such as mobile phones means that they are particularly exposed to electromagnetic fields (EMF) and other problems. However, few studies have researched the effects of long-term exposure to EMF in the kidney. We therefore investigated oxidative stress and apoptosis in long-term exposure to 2100 megahertz (MHz) in a rat model. **Materials and methods:** Twenty-four Sprague Dawley rats were divided into a control group (n = 8, no EMF exposure), a group exposed to 2100 MHz for 6 h for 30 d (n = 8), and a group exposed to 2100 MHz for 12 h for 30 d (n = 8). Immunohistochemical analysis was performed, using caspase-3 to evaluate apoptosis. Immediately after treatment, reduced glutathione (GSH), malondialdehyde (MDA) in kidney tissue and serum levels of various biochemical compounds were measured to detect oxidative stress. **Results:** Deterioration was observed in the brush border in renal tubules of the EMF groups. The results of the immunohistochemical analysis revealed a greater number of positively stained renal tubular epithelial cells in the EMF groups as compared with that in the control group. In the EMF groups, renal MDA levels increased, and renal GSH levels decreased compared with those in the control group, as shown by a biochemical examination (p = 0.00 and p = 0.00, respectively). **Conclusion:** The findings showed that exposure to 2100 MHz for 6 and 12 h induced oxidative stress-mediated acute renal injury, depending on the length of exposure and dosage.

**(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.**

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), totaloxidant 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) \*Bilgici B, Akar A, Avci B, Tuncel OK. Effect of 900 MHz radiofrequency radiation on oxidative stress in rat brain and serum. Electromagn Biol Med. 32:20-29, 2013.**

The increasing use of mobile telephones raises the question of possible adverse effects of the electromagnetic fields (EMF) that these phones produce. In this study, we examined the oxidative stress in the brain tissue and serum of rats that resulted from exposure to a 900-MHz EMF at a whole body average specific absorption rate (SAR) of 1.08 W/kg for 1 h/day for 3 weeks. We also examined the antioxidant effect of garlic powder (500 mg/kg/day) given orally to EMF-exposed rats. We found that malondialdehyde (MDA) ( $p < 0.001$ ) and advanced oxidation protein product (AOPP) ( $p < 0.05$ ) increased in rat brain tissue exposed to the EMF and that garlic reduced these effects ( $p < 0.05$ ). There was no significant difference in the nitric oxide (NO) levels in the brain. Paraoxonase (PON) was not detected in the brain. There was a significant increase in the levels of NO ( $p < 0.001$ ) detected in the serum after EMF exposure, and garlic intake did not affect this increase in NO. Our results suggest that there is a significant increase in brain lipid and protein oxidation after electromagnetic radiation (EMR) exposure and that garlic has a protective effect against this oxidative stress.

**(E) Bin-Meferij MM, El-Kott AF. The radioprotective effects of Moringa oleifera against mobile phone electromagnetic radiation-induced infertility in rats. Int J Clin Exp Med. 2015 Aug 15;8(8):12487-97. eCollection 2015.**

The present study has investigated the effects of mobile phone electromagnetic radiation (EMR) on fertility in rats. The purpose of this study was to explore the capability of polyphenolic-rich Moringa oleifera leaf extract in protecting rat testis against EMR-induced impairments based on evaluation of sperm count, viability, motility, sperm cell morphology, anti-oxidants (SOD & CAT), oxidative stress marker, testis tissue histopathology and PCNA immunohistochemistry. The sample consisted of sixty male Wistar rats which were divided into four equal groups. The first group (the control) received only standard diet while the second group was supplemented daily and for eight weeks with 200 mg/kg aqueous extract of Moringa leaves. The third group was exposed to 900 MHz fields for one hour a day and for (7) days a week. As for the fourth group, it was exposed to mobile phone radiation and received the Moringa extract. The results showed that the EMR treated group exhibited a significantly decrease sperm parameters. Furthermore, concurrent exposure to EMR and treated with MOE significantly enhanced the sperm parameters. However, histological results in EMR group showed irregular seminiferous tubules, few spermatogonia, giant multinucleated cells, degenerated spermatozoa and the number of Leydig cells was

significantly reduced. PCNA labeling indices were significant in EMR group versus the control group. Also, EMR affects spermatogenesis and causes to apoptosis due to the heat and other stress-related EMR in testis tissue. This study concludes that chronic exposure to EMR marked testicular injury which can be prevented by *Moringa oleifera* leaf extract.

**(E) \*Bodera P, Stankiewicz W, Zawada K, Antkowiak B, Paluch M, Kieliszek J, Kalicki B, Bartosiński A, Wawer I. Changes in antioxidant capacity of blood due to mutual action of electromagnetic field (1800 MHz) and opioid drug (tramadol) in animal model of persistent inflammatory state. Pharmacol Rep. 65(2):421-428, 2013.**

Background: The biological effects and health implications of electromagnetic field (EMF) associated with cellular mobile telephones and related wireless systems and devices have become a focus of international scientific interest and world-wide public concern. It has also been proved that EMF influences the production of reactive oxygen species (ROS) in different tissues. Methods: Experiments were performed in healthy rats and in rats with persistent inflammatory state induced by Complete Freund's Adjuvant (CFA) injection, which was given 24 h before EMF exposure and drug application. Rats were injected with CFA or the same volume of paraffin oil into the plantar surface of the left hind paw. Animals were exposed to the far-field range of an antenna at 1800 MHz with the additional modulation which was identical to that generated by mobile phone GSM 1800. Rats were given 15 min exposure, or were sham-exposed with no voltage applied to the field generator in control groups. Immediately before EMF exposure, rats were injected intraperitoneally with tramadol in the 20 mg/kg dose or vehicle in the 1 ml/kg volume. Results: Our study revealed that single EMF exposure in 1800 MHz frequency significantly reduced antioxidant capacity both in healthy animals and those with paw inflammation. A certain synergic mode of action between applied electromagnetic fields and administered tramadol in rats treated with CFA was observed. Conclusions: The aim of the study was to examine the possible, parallel/combined effects of electromagnetic radiation, artificially induced inflammation and a centrally-acting synthetic opioid analgesic drug, tramadol, (used in the treatment of severe pain) on the antioxidant capacity of blood of rats. The antioxidant capacity of blood of healthy rats was higher than that of rats which received only tramadol and were exposed to electromagnetic fields.

**(E) Bodera P, Stankiewicz W, Antkowiak B, Paluch M, Kieliszek J, Sobiech J, Niemcewicz M. Influence of electromagnetic field (1800 MHz) on lipid peroxidation in brain, blood, liver and kidney in rats. Int J Occup Med Environ Health. 28(4):751-759, 2015.**

OBJECTIVES: The aim of this study is the evaluation of the influence of repeated (5 times for 15 min) exposure to electromagnetic field (EMF) of 1800 MHz frequency on tissue lipid peroxidation (LPO) both in normal and inflammatory state, combined with analgesic treatment. MATERIAL AND METHODS: The concentration of malondialdehyde (MDA) as the end-product of the lipid peroxidation (LPO) was estimated in blood, liver, kidneys, and brain of Wistar rats, both healthy and those with complete Freund's adjuvant (CFA)-induced persistent paw inflammation. RESULTS: The slightly elevated levels of the MDA in blood, kidney, and brain were observed among healthy rats in electromagnetic field (EMF)-exposed groups, treated with tramadol (TRAM/EMF and exposed to the EMF). The malondialdehyde

remained at the same level in the liver in all investigated groups: the control group (CON), the exposed group (EMF), treated with tramadol (TRAM) as well as exposed to and treated with tramadol (TRAM/EMF). In the group of animals treated with the complete Freund's adjuvant (CFA) we also observed slightly increased values of the MDA in the case of the control group (CON) and the exposed groups (EMF and TRAM/EMF). The MDA values concerning kidneys remained at the same levels in the control, exposed, and not-exposed group treated with tramadol. Results for healthy rats and animals with inflammation did not differ significantly. **CONCLUSIONS:** The electromagnetic field exposure (EMF), applied in the repeated manner together with opioid drug tramadol (TRAM), slightly enhanced lipid peroxidation level in brain, blood, and kidneys.

**(E) Borzoueisileh S, Shabestani Monfared A, Ghorbani H, Mortazavi SMJ, Zabihi E, Pouramir M, Doustimotlagh AH, Shafiee M, Niksirat F.**

**Assessment of function, histopathological changes, and oxidative stress in liver tissue due to ionizing and non-ionizing radiations. Caspian J Intern Med 2020 11(3):315-323.**

**Background:** Compared to past decades, humans are exposed to rapidly increasing levels of radiofrequency electromagnetic radiations (RF-EMF). Despite numerous studies, the biological effects of human exposure to different levels of RF-EMF are not fully understood yet. This study aimed to evaluate the bioeffects of exposure to "900/1800 MHz" and "2.4 GHz" RF-EMFs, and x-rays alone as well as their potential interactions, i.e. inducing simple additive, adaptive, or synergistic effects. **Methods:** 120 Wistar rats were randomly divided into ten groups of 12 each. The rats were exposed to RF-EMF, 10 cGy, and 8 Gy x-rays, a combination of these exposures, or only sham-exposed. The levels of liver enzymes were determined in serum samples by an auto-analyzer. Moreover, the histopathological changes, and the levels of malondialdehyde (MDA), nitric oxide, ferric reducing antioxidant power, total thiols, and protein carbonyl (PCO) were measured. **Results:** Among the markers of liver function, gamma-glutamyltransferase was not associated with irradiation but, aspartate transaminase, alanine transaminase, and alkaline phosphatase showed some levels of association. MDA and PCO levels after 8 Gy irradiation increased, but pre-exposure to RF-EMF could modulate their changes. At the cellular level, the frequency of lobular inflammation was associated with the type of intervention. **Conclusion:** The exposure to both ionizing and non-ionizing radiations could alter some liver function tests. A short term pre-exposure to RF-EMF before exposure to an 8 Gy challenging dose of x-rays caused the alterations in oxidative stress markers and liver function tests, which indicate that oxidative stress is possibly involved in the adaptive response.

**(E) Bouji M, Lecomte A, Gamez C, Blazy K, Villégier AS. Impact of Cerebral Radiofrequency Exposures on Oxidative Stress and Corticosterone in a Rat Model of Alzheimer's Disease. J Alzheimers Dis. 73:467-476, 2020.**

**BACKGROUND:** Alzheimer's disease (AD) is the most common type of neurodegenerative disease leading to dementia. Several studies suggested that mobile phone radiofrequency electromagnetic field (RF-EMF) exposures modified AD memory deficits in rodent models.

**OBJECTIVE:** Here we aimed to test the hypothesis that RF-EMF exposure may modify memory through corticosterone and oxidative stress in the Samaritan rat model of AD.

**METHODS:** Long-Evans male rats received intracerebroventricular infusion with ferrous sulphate, amyloid-beta 1-42 peptide, and buthionine-sulfoximine (AD rats) or with vehicle (control rats). To mimic cell phone use, RF-EMF were exposed to the head for 1 month (5 days/week, in restraint). To look for hazard thresholds, high brain averaged specific absorption rates (BASAR) were tested: 1.5 W/Kg (15 min), 6 W/Kg (15 min), and 6 W/Kg (45 min). The sham group was in restraint for 45 min. Endpoints were spatial memory in the radial maze, plasmatic corticosterone, heme oxygenase-1 (HO1), and amyloid plaques.

**RESULTS:** Results indicated similar corticosterone levels but impaired memory performances and increased cerebral staining of thioflavine and of HO1 in the sham AD rats compared to the controls. A correlative increase of cortical HO1 staining was the only effect of RF-EMF in control rats. In AD rats, RF-EMF exposures induced a correlative increase of hippocampal HO1 staining and reduced corticosterone.

**DISCUSSION:** According to our data, neither AD nor control rats showed modified memory after RF-EMF exposures. Unlike control rats, AD rats showed higher hippocampal oxidative stress and reduced corticosterone with the higher BASAR. This data suggests more fragility related to neurodegenerative disease toward RF-EMF exposures.

**(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.**

*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<sup>-1</sup> for a period of two hours (corresponding to specific absorption rates ranging from 0.13 to 9.33 mW kg<sup>-1</sup>). 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<sup>-1</sup>. 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<sup>-1</sup> field levels. Western blots showing an increased quantity of HSP70 and MTCO1 proteins confirmed this stress response. HSP70 and LYS genes were up-regulated after 24 h of recovery following a two hour-exposure, meaning that the effect of EMF exposure lasted for hours. Exposure to mobile phone (900-1800 MHz) during pregnancy: tissue oxidative stress after childbirth.

**(NE) Brescia F, Sarti M, Massa R, Calabrese ML, Sannino A, Scarfi MR. Reactive oxygen species formation is not enhanced by exposure to UMTS 1950 MHz radiation and co-exposure to ferrous ions in Jurkat cells. Bioelectromagnetics. 30:525-535, 2009.**

This study was designed to assess if radiofrequency (RF) radiation induces oxidative stress in cultured mammalian cells when given alone or in combination with ferrous ions (FeSO(4)). For this purpose the production of reactive oxygen species (ROS) was measured by flow cytometry in human lymphoblastoid cells exposed to 1950 MHz signal used by the third generation wireless technology of the Universal Mobile Telecommunication System (UMTS) at Specific Absorption Rate of 0.5 and 2.0 W/kg. Short (5-60 min) or long (24 h) duration exposures were carried out in a waveguide system under strictly controlled conditions of both dosimetry and environment. Cell viability was also measured after 24 h RF exposure using the Resazurin and Neutral Red assays. Several co-exposure protocols were applied to test if RF radiation is able to alter ROS formation induced by FeSO(4) (RF given before or concurrently to FeSO(4)). The results obtained indicate that non-thermal RF exposures do not increase spontaneous ROS formation in any of the experimental conditions investigated. Consistent with the lack of ROS production, no change in cell viability was observed in Jurkat cells exposed to RF radiation for 24 h. Similar results were obtained when co-exposures were considered: combined exposures to RF radiation and FeSO(4) did not increase ROS formation induced by the chemical treatment alone. In contrast, in cultures treated with FeSO(4) as positive control, a dose-dependent increase in ROS formation was recorded, validating the sensitivity of the method employed.

**(E) Burlaka A, Tsybulin O, Sidorik E, Lukin S, Polishuk V, Tshmistrenko 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, LI, IFR)**

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 ( $0.25 \mu\text{W}/\text{cm}^2$ ) during 158-360 h discontinuously (48 c - ON, 12 c - OFF) before and in the initial stages of development. The levels of superoxide ( $\text{O}_2^-$ ), nitrogen oxide ( $\text{NO}\cdot$ ), 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) Burlaka A, Selyuk M, Gafurov M, Lukin S, Potaskalova V, Sidorik E. Changes in mitochondrial functioning with electromagnetic radiation of ultra high frequency as revealed by electron paramagnetic resonance methods. Int J Radiat Biol. 90:357-362, 2014.**

Purpose: To study the effects of electromagnetic radiation (EMR) of ultra high frequency (UHF) in the doses equivalent to the maximal permitted energy load for the staffs of the radar stations on the biochemical processes that occur in the cell organelles. Materials and Methods: Liver, cardiac and aorta tissues from the male rats exposed to non-thermal UHF EMR in pulsed and continuous modes were studied during 28 days after the irradiation by the electron paramagnetic resonance (EPR) methods including a spin trapping of superoxide radicals. Results: The qualitative and quantitative disturbances in electron transport chain (ETC) of mitochondria are registered. A formation of the iron-nitrosyl complexes of nitric oxide (NO) radicals with the iron-sulphide (FeS) proteins, the decreased activity of FeS-protein N2 of NADH-ubiquinone oxidoreductase complex and flavo ubisemiquinone growth combined with the increased rates of superoxide production are obtained. Conclusions: (1) Abnormalities in the mitochondrial ETC of liver and aorta cells are more pronounced for animals radiated in a pulsed mode. (2) The alterations in the functioning of the mitochondrial ETC cause increase of superoxide radicals generation rate in all samples, formation of cellular hypoxia, and intensification of the oxide-initiated metabolic changes. (3) Electron paramagnetic resonance methods could be used to track the qualitative and quantitative changes in the mitochondrial ETC caused by the UHF EMR.

**(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(1):52-55, 2010.**

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 20min to either 900MHz continuous waves or 900MHz waves modulated in amplitude at 50Hz 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 20min. 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) Cao H, Qin F, Liu X, Wang J, Cao Y, Tong J, Zhao H. Circadian Rhythmicity of Antioxidant Markers in Rats Exposed to 1.8 GHz Radiofrequency Fields. Int J Environ Res Public Health. 12(2):2071-2087, 2015.**

**BACKGROUND:** The potential health risks of exposure to Radiofrequency Fields (RF) emitted by mobile phones are currently of considerable public interest, such as the adverse effects on the circadian rhythmicities of biological systems. To determine whether circadian rhythms of the plasma antioxidants (Mel, GSH-Px and SOD) are affected by RF, we performed a study on male Sprague Dawley rats exposed to the 1.8 GHz RF. **METHODS:** All animals were divided into seven groups. The animals in six groups were exposed to 1.8 GHz RF (201.7  $\mu\text{W}/\text{cm}^2$  power density, 0.05653 W/kg specific absorption rate) at a specific period of the day (3, 7, 11, 15, 19 and 23 h GMT, respectively), for 2 h/day for 32 consecutive days. The rats in the seventh group were used as sham-exposed controls. At the end of last RF exposure, blood samples were collected from each rat every 4 h (total period of 24 h) and also at similar times from sham-exposed animals. The concentrations of three antioxidants (Mel, GSH-Px and SOD) were determined. The data in RF-exposed rats were compared with those in sham-exposed animals. **RESULTS:** circadian rhythms in the synthesis of Mel and antioxidant enzymes, GSH-Px and SOD, were shifted in RF-exposed rats compared to sham-exposed animals: the Mel, GSH-Px and SOD levels were significantly decreased when RF exposure was given at 23 and 3 h GMT. **CONCLUSION:** The overall results indicate that there may be adverse effects of RF exposure on antioxidant function, in terms of both the daily antioxidative levels, as well as the circadian rhythmicity.

**(cancer) (E) Cao Y, Zhang W, Lu MX, Xu Q, Meng QQ, Nie JH, Tong J. 900-MHz microwave radiation enhances gamma-ray adverse effects on SHG44 cells. J Toxicol Environ Health A. 72(11):727-732, 2009.**

Mobile phones are widely used globally. However, the biological effects due to exposure to electromagnetic fields (EMF) produced by mobile phones are largely unknown. Environmental and occupational exposure of humans to gamma-rays is a biologically relevant phenomenon. Consequently studies were undertaken to examine the interactions between gamma-rays and EMF on human health. In this study, exposure to 900-MHz EMF expanded gamma-ray damage to SHG44 cells. Preexposure EMF enhanced the decrease in cell proliferation induced by gamma-ray irradiation and the rate of apoptosis. The combination of EMF and gamma-ray exposure resulted in a synergistic effect by triggering stress response, which increased reactive oxygen species, but the expression of hsp70 at both mRNA and protein levels remained unaltered. Data indicate that the adverse effects of gamma-rays on cellular functions are strengthened by EMF.

**(cancer) (E) Castello PR, Hill I, Sivo F, Portelli L, Barnes F, Usselman R, Martino CF. Inhibition of cellular proliferation and enhancement of hydrogen peroxide production in fibrosarcoma cell line by weak radio frequency magnetic fields. Bioelectromagnetics. 35(8):598-602, 2014.**

This study presents experimental data for the effects of weak radio frequency (RF) magnetic fields on hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>) production and cellular growth rates of fibrosarcoma HT1080 cells in vitro. Cells were exposed either to 45  $\mu$ T static magnetic fields (SMFs)-oriented vertical to the plane of growth or to SMFs combined with weak 5 and 10 MHz RF magnetic fields of 10  $\mu$ TRMS intensity perpendicular to the static field. Cell numbers were reduced up to 30% on Day 2 for the cells exposed to the combination of SMF and a 10 MHz RF magnetic field compared with the SMF control cells. In addition, cells exposed to 10 MHz RF magnetic fields for 8 h increased H<sub>2</sub>O<sub>2</sub> production by 55%. The results demonstrate an overall magnetic field-induced biological effect that shows elevated H<sub>2</sub>O<sub>2</sub> levels with accompanying decrease in cellular growth rates.

**(E) Cetin H, Nazırođlu M, Celik O, Yüksel M, Pastacı N, Ozkaya MO. Liver antioxidant stores protect the brain from electromagnetic radiation (900 and 1800 MHz)-induced oxidative stress in rats during pregnancy and the development of offspring. J Matern Fetal Neonatal Med. 2014 Mar 3. [Epub ahead of print]**

Objectives: The present study determined the effects of mobile phone (900 and 1800 MHz)-induced electromagnetic radiation (EMR) exposure on oxidative stress in the brain and liver as well as the element levels in growing rats from pregnancy to 6 weeks of age. Methods: Thirty-two rats and their offspring were equally divided into 3 different groups: the control, 900 MHz, and 1800 MHz groups. The 900 MHz and 1800 MHz groups were exposed to EMR for 60 min/day during pregnancy and neonatal development. At the 4th, 5th, and 6th weeks of the experiment, brain samples were obtained. Results: Brain and liver glutathione peroxidase (GSH-Px) activities, as well as liver vitamin A and  $\beta$ -carotene concentrations decreased in the EMR groups, although brain iron, vitamin A, and  $\beta$ -carotene concentrations increased in the EMR groups. In the 6th week, selenium concentrations in the brain decreased in the EMR groups. There were no statistically significant differences in glutathione, vitamin E, chromium, copper, magnesium, manganese, and zinc concentrations between the 3 groups. Conclusion: EMR-induced oxidative stress in the brain and liver was reduced during the development of offspring. Mobile phone-induced EMR could be considered as a cause of oxidative brain and liver injury in growing rats.

**(E) \*Ceyhan AM, Akkaya VB, Güleçol ŞC, Ceyhan BM, Özgüner F, Chen W. Protective effects of  $\beta$ -glucan against oxidative injury induced by 2.45-GHz electromagnetic radiation in the skin tissue of rats. Arch Dermatol Res. 304(7):521-527, 2012.**

In recent times, there is widespread use of 2.45-GHz irradiation-emitting devices in industrial, medical, military and domestic application. The aim of the present study was to investigate the effect of 2.45-GHz electromagnetic radiation (EMR) on the oxidant and antioxidant status of skin and to examine the possible protective effects of  $\beta$ -glucans against the oxidative injury. Thirty-two male Wistar albino rats were randomly divided into four equal groups: control; sham exposed; EMR; and EMR +  $\beta$ -glucan. A 2.45-GHz EMR emitted device from the experimental exposure was applied to the EMR group and EMR +  $\beta$ -glucan group for 60 min daily, respectively, for 4 weeks.  $\beta$ -glucan was administered via gavage at a dose of 50 mg/kg/day before each exposure to radiation in

the treatment group. The activities of antioxidant enzymes, superoxide dismutase (SOD), glutathione peroxidase (GSH-Px) and catalase (CAT), as well as the concentration of malondialdehyde (MDA) were measured in tissue homogenates of the skin. Exposure to 2.45-GHz EMR caused a significant increase in MDA levels and CAT activity, while the activities of SOD and GSH-Px decreased in skin tissues. Systemic  $\beta$ -glucan significantly reversed the elevation of MDA levels and the reduction of SOD activities.  $\beta$ -glucan treatment also slightly enhanced the activity of CAT and prevented the depletion of GSH-Px activity caused by EMR, but not statistically significantly. The present study demonstrated the role of oxidative mechanisms in EMR-induced skin tissue damages and that  $\beta$ -glucan could ameliorate oxidative skin injury via its antioxidant properties.

**(E) Chandel S, Kaur S, Singh HP, Batish DR, Kohli RK. Exposure to 2100 MHz electromagnetic field radiations induces reactive oxygen species generation in *Allium cepa* roots. *Journal of Microscopy and Ultrastructure*, 5(4):225-229, 2017.**

During the last few decades there has been an enormous increase in the usage of cell phones as these are one of the most convenient gadgets and provide excellent mode of communication without evoking any hindrance to movement. However, these are significantly adding to the electromagnetic field radiations (EMF-r) in the environment and thus, are required to be analysed for their impacts on living beings. The present study investigated the role of cell phone EMF-r in inciting oxidative damage in onion (*Allium cepa*) roots at a frequency of 2100 MHz. Onion roots were exposed to continuous wave homogenous EMF-r for 1, 2 and 4 h for single day and generation of reactive oxygen species (ROS) in terms of malondialdehyde (MDA), hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>) and superoxide anion (O<sub>2</sub><sup>-</sup>) content and changes in the activities of antioxidant enzymes- superoxide dismutases (SOD) and catalases (CAT) were measured. The results showed that EMF-r exposure enhanced the content of MDA, H<sub>2</sub>O<sub>2</sub> and O<sub>2</sub><sup>-</sup>. Also, there was an upregulation in the activity of antioxidant enzymes- SOD and CAT- in onion roots. The study concluded that 2100 MHz cell phone EMF-r incite oxidative damage in onion roots by altering the oxidative metabolism.

**(E) Chauhan P, Verma HN, Sisodia R, Kesari KK. Microwave radiation (2.45 GHz)-induced oxidative stress: Whole-body exposure effect on histopathology of Wistar rats. *Electromagn Biol Med*. 36(1):20-30, 2017.**

Man-made microwave and radiofrequency (RF) radiation technologies have been steadily increasing with the growing demand of electronic appliances such as microwave oven and cell phones. These appliances affect biological systems by increasing free radicals, thus leading to oxidative damage. The aim of this study was to explore the effect of 2.45 GHz microwave radiation on histology and the level of lipid peroxide (LPO) in Wistar rats. Sixty-day-old male Wistar rats with  $180 \pm 10$  g body weight were used for this study. Animals were divided into two groups: sham exposed (control) and microwave exposed. These animals were exposed for 2 h a day for 35 d to 2.45 GHz microwave radiation (power density, 0.2 mW/cm<sup>2</sup>). The whole-body specific absorption rate (SAR) was estimated to be 0.14 W/kg. After completion of the exposure period, rats were sacrificed, and brain, liver, kidney, testis and spleen were stored/preserved for determination of LPO and histological parameters. Significantly high level of LPO was observed in

the liver ( $p < 0.001$ ), brain ( $p < 0.004$ ) and spleen ( $p < 0.006$ ) in samples from rats exposed to microwave radiation. Also histological changes were observed in the brain, liver, testis, kidney and spleen after whole-body microwave exposure, compared to the control group. Based on the results obtained in this study, we conclude that exposure to microwave radiation 2 h a day for 35 d can potentially cause histopathology and oxidative changes in Wistar rats. These results indicate possible implications of such exposure on human health.

**(E) \*Chen YB, Li J, Liu JY, Zeng LH, Wan Y, Li YR, Ren D, Guo GZ. Effect of Electromagnetic Pulses (EMP) on associative learning in mice and a preliminary study of mechanism. Int J Radiat Biol. 87(12):1147-1154, 2011.**

**PURPOSE:** To investigate the effects of electromagnetic pulses (EMP) on associative learning in mice and test a preliminary mechanism for these effects. **MATERIALS AND METHODS:** A tapered parallel plate gigahertz transverse electromagnetic (GTEM) cell with a flared rectangular coaxial transmission line was used to expose male BALB/c mice to EMP (peak-intensity 400 kV/m, rise-time 10 ns, pulse-width 350 ns, 0.5 Hz and total 200 pulses). Concurrent sham-exposed mice were used as a control. Associative learning, oxidative stress in the brain, serum chemistry and the protective action of tocopherol monoglucoside (TMG) in mice were measured, respectively. **RESULTS:** (1) Twelve hour and 1 day post EMP exposure associative learning was reduced significantly compared with sham control ( $p < 0.05$ ) but recovered at 2 d post EMP exposure. (2) Compared with the sham control, lipid peroxidation of brain tissue and chemiluminescence (CL) intensity increased significantly ( $p < 0.05$ ), while the activity of the antioxidant enzymes Superoxide Dismutase [SOD], Glutathione [GSH], Glutathione Peroxidase [GSH-Px], Catalase [CAT] decreased significantly ( $p < 0.05$ ) at 3 h, 6 h, 12 h and 1 d post EMP exposure. All these parameters recovered at 2 d post EMP exposure. (3) No significant differences between the sham control group and EMP exposed group were observed in serum cholesterol and triglycerides. (4) Pretreatment of mice with TMG showed protective effects to EMP exposure. **CONCLUSIONS:** EMP exposure significantly decreased associative learning in mice and TMG acted as an effective protective agent from EMP exposure. This mechanism could involve an increase of oxidative stress in brain by EMP exposure.

**(E) 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. 2020 Jun 8;10(1):9238.**

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.

**(cancer) (E) Çiğ B, Nazıroğlu M. Investigation of the effects of distance from sources on apoptosis, oxidative stress and cytosolic calcium accumulation via TRPV1 channels induced by mobile phones and Wi-Fi in breast cancer cells. Biochim Biophys Acta. 2015 Feb 19. pii: S0005-2736(15)00053-X. doi: 10.1016/j.bbame.2015.02.013. [Epub ahead of print]**

TRPV1 is a  $\text{Ca}^{2+}$  permeable channel and gated by noxious heat, oxidative stress and capsaicin (CAP). Some reports have indicated that non-ionized electromagnetic radiation (EMR)-induces heat and oxidative stress effects. We aimed to investigate the effects of distance from sources on calcium signaling, cytosolic ROS production, cell viability, apoptosis, plus caspase-3 and -9 values induced by mobile phones and Wi-Fi in breast cancer cells MCF-7 human breast cancer cell lines were divided into A, B, C and D groups as control, 900, 1800 and 2450MHz groups, respectively. Cells in Group A were used as control and were kept in cell culture conditions without EMR exposure. Groups B, C and D were exposed to the EMR frequencies at different distances (0cm, 1cm, 5cm, 10cm, 20cm and 25cm) for 1h before CAP stimulation. The cytosolic ROS production,  $\text{Ca}^{2+}$  concentrations, apoptosis, caspase-3 and caspase-9 values were higher in groups B, C and D than in A group at 0cm, 1cm and 5cm distances although cell viability (MTT) values were increased by the distances. There was no statistically significant difference in the values between control, 20 and 25cm. Wi-Fi and mobile phone EMR placed within 10cm of the cells induced excessive oxidative responses and apoptosis via TRPV1-induced cytosolic  $\text{Ca}^{2+}$  accumulation in the cancer cells. Using cell phones and Wi-Fi sources which are farther away than 10cm may provide useful protection against oxidative stress, apoptosis and overload of intracellular  $\text{Ca}^{2+}$ . This article is part of a Special Issue entitled: Membrane channels and transporters in cancers.

**(E) Comelekoglu U, Aktas S, Demirbag B, Karagul MI, Yalin S, Yildirim M, Akar A, Korunur Engiz B, Sogut F, Ozbay E. Effect of low-level 1800 MHz radiofrequency radiation on the rat sciatic nerve and the protective role of paricalcitol. Bioelectromagnetics. 39(8):631-643, 2018.**

The nervous system is an important target of radiofrequency (RF) radiation exposure since it is the excitable component that is potentially able to interact with electromagnetic fields. The present study was designed to investigate the effects of 1,800 MHz RF radiation and the protective role of paricalcitol on the rat sciatic nerve. Rats were divided into four groups as control, paricalcitol, RF, and RF + paricalcitol. In RF groups, the rats were exposed to 1,800 MHz RF for 1 h per day for 4 weeks. Control and paricalcitol rats were kept under the same conditions without RF

application. In paricalcitol groups, the rats were given 0.2 µg/kg/day paricalcitol, three times per week for 4 weeks. Amplitude and latency of nerve compound action potentials, catalase activities, malondialdehyde (MDA) levels, and ultrastructural changes of sciatic nerve were evaluated. In the RF group, a significant reduction in amplitude, prolongation in latency, an increase in the MDA level, and an increase in catalase activity and degeneration in the myelinated nerve fibers were observed. The electrophysiological and histological findings were consistent with neuropathy, and the neuropathic changes were partially ameliorated with paricalcitol administration.

**(E) Dasdag S, Akdag MZ, Ulukaya E, Uzunlar AK, Ocak AR. Effect of mobile phone exposure on apoptotic glial cells and status of oxidative stress in rat brain. *Electromagn Biol Med.* 28(4):342-354, 2009.**

The aim of this study was to investigate the effects of mobile phone exposure on glial cells in brain. The study carried out on 31 Wistar Albino adult male rats. The rat heads in a carousel exposed to 900 MHz microwave. For the study group (n:14), rats exposed to the radiation 2 h per day (7 days in a week) for 10 months. For the sham group (n:7), rats were placed into the carousel and the same procedure was applied except that the generator was turned off. For the cage control (n:10), nothing applied to rats in this group. In this study, rats were euthanized after 10 months of exposure periods and brains were removed. Brain tissues were immunohistochemically stained for the active (cleaved) caspase-3, which is a well-known apoptosis marker, and p53. The expression of the proteins was evaluated by a semi-quantitative scoring system. However, total antioxidative capacity (TAC), catalase, total oxidant status (TOS), and oxidative stress index were measured in rat brain. Final score for apoptosis in the exposed group was significantly lower than the sham ( $p < 0.001$ ) and the cage control groups ( $p < 0.01$ ). p53 was not significantly changed by the exposure ( $p > 0.05$ ). The total antioxidant capacity and catalase in the experimental group was found higher than that in the sham group ( $p < 0.001$ ,  $p < 0.05$ ). In terms of the TOS and oxidative stress index, there was no statistically significant difference between exposure and sham groups ( $p > 0.05$ ). In conclusion, the final score for apoptosis, total antioxidant capacity and catalase in rat brain might be altered by 900 MHz radiation produced by a generator to represent exposure of global systems for mobile communication (GSM) cellular phones.

**(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(7): e6446, 2009. doi:10.1371/journal.pone.0006446**

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.

**(E) De Luca C, Chung Sheun Thai J, Raskovic D, Cesareo E, Caccamo D, Trukhanov A, Korkina L. Metabolic and genetic screening of electromagnetic hypersensitive subjects as a feasible tool for diagnostics and intervention. Mediators Inflamm. 2014;2014:924184. doi: 10.1155/2014/924184. Epub 2014 Apr 9.**

Growing numbers of "electromagnetic hypersensitive" (EHS) people worldwide self-report severely disabling, multiorgan, non-specific symptoms when exposed to low-dose electromagnetic radiations, often associated with symptoms of multiple chemical sensitivity (MCS) and/or other environmental "sensitivity-related illnesses" (SRI). This cluster of chronic inflammatory disorders still lacks validated pathogenetic mechanism, diagnostic biomarkers, and management guidelines. We hypothesized that SRI, not being merely psychogenic, may share organic determinants of impaired detoxification of common physic-chemical stressors. Based on our previous MCS studies, we tested a panel of 12 metabolic blood redox-related parameters and of selected drug-metabolizing-enzyme gene polymorphisms, on 153 EHS, 147 MCS, and 132 control Italians, confirming MCS altered (P < 0.05-0.0001) glutathione-(GSH), GSH-peroxidase/S-transferase, and catalase erythrocyte activities. We first described comparable-though milder-metabolic pro-oxidant/proinflammatory alterations in EHS with distinctively increased plasma coenzyme-Q10 oxidation ratio. Severe depletion of erythrocyte membrane polyunsaturated fatty acids with increased  $\omega 6/\omega 3$  ratio was confirmed in MCS, but not in EHS. We also identified significantly (P = 0.003) altered distribution-versus-control of the CYP2C19\*1/\*2 SNP variants in EHS, and a 9.7-fold increased risk (OR: 95% C.I. = 1.3-74.5) of developing EHS for the haplotype (null)GSTT1 + (null)GSTM1 variants. Altogether, results on MCS and EHS strengthen our proposal to adopt this blood metabolic/genetic biomarkers' panel as suitable diagnostic tool for SRI.

**(NE) \*Demirel S, Doganay S, Turkoz Y, Dogan Z, Turan B, Firat PG. Effects of third generation mobile phone-emitted electromagnetic radiation on oxidative stress parameters in eye tissue and blood of rats. *Cutan Ocul Toxicol.* 31(2):89-94, 2012.**

Purpose: To investigate the effects of electromagnetic radiation (EMR) emitted by a third generation (3G) mobile phone on the antioxidant and oxidative stress parameters in eye tissue and blood of rats. Methods: Eighteen Wistar albino rats were randomly assigned into two groups: Group I (n = 9) received a standardized a daily dose of 3G mobile phone EMR for 20 days, and Group II served as the control group (n = 9), receiving no exposure to EMR. Glutathione peroxidase (GSH-Px) and catalase (CAT) levels were measured in eye tissues; in addition, malondialdehyde (MDA) and reduced GSH levels were measured in blood. Results: There was no significant difference between groups in GSH-Px (p = 0.99) and CAT (p = 0.18) activity in eye tissue. There was no significant difference between groups in MDA (p = 0.69) and GSH levels (p = 0.83) in blood. Conclusions: The results of this study suggest that under a short period of exposure, 3G mobile phone radiation does not lead to harmful effects on eye tissue and blood in rats.

**(E) \*Deshmukh PS, Banerjee BD, Abegaonkar MP, Megha K, Ahmed RS, Tripathi AK, Mediratta PK. Effect of low level microwave radiation exposure on cognitive function and oxidative stress in rats. *Indian J Biochem Biophys.* 50(2):114-119, 2013. (LI)**

Use of wireless communicating devices is increasing at an exponential rate in present time and is raising serious concerns about possible adverse effects of microwave (MW) radiation emitted from these devices on human health. The present study aimed to evaluate the effects of 900 MHz MW radiation exposure on cognitive function and oxidative stress in blood of Fischer rats. Animals were divided into two groups (6 animals/group): Group I (MW-exposed) and Group II (Sham-exposed). Animals were subjected to MW exposure (Frequency 900 MHz; specific absorption rate  $8.4738 \times 10^{-5}$  W/kg) in Gigahertz transverse electromagnetic cell (GTEM) for 30 days (2 h/day, 5 days/week). Subsequently, cognitive function and oxidative stress parameters were examined for each group. Results showed significant impairment in cognitive function and increase in oxidative stress, as evidenced by the increase in levels of MDA (a marker of lipid peroxidation) and protein carbonyl (a marker of protein oxidation) and unaltered GSH content in blood. Thus, the study demonstrated that low level MW radiation had significant effect on cognitive function and was also capable of leading to oxidative stress.

**(NE) de Souza FT, Silva JF, Ferreira EF, Siqueira EC, Duarte AP, Gomez MV, Gomez RS, Gomes CC. Cell phone use and parotid salivary gland alterations: no molecular evidence. *Cancer Epidemiol Biomarkers Prev.* 2014 Apr 21. [Epub ahead of print]**

Background The association between cell phone use and the development of parotid tumors is controversial. Because there is unequivocal evidence that the microenvironment is important for tumor formation, we investigated in the parotid glands whether cell phone use alters the expression of gene products related to cellular stress. Methods We used the saliva produced by the parotid glands of 62 individuals to assess molecular alterations compatible with cellular

stress, comparing the saliva from the gland exposed to cell phone radiation (ipsilateral) to the saliva from the opposite, unexposed parotid gland (contralateral) of each individual. We compared salivary flow, total protein concentration, p53, p21, reactive oxygen species (ROS), and salivary levels of glutathione (GSH), heat shock proteins 27 and 70 and IgA between the ipsilateral and contralateral parotids. Results No difference was found for any of these parameters, even when grouping individuals by period of cell phone use in years or by monthly average calls in minutes. Conclusions and Impact We provide molecular evidence that the exposure of parotid glands to cell phone use does not alter parotid salivary flow, protein concentration or levels of proteins of genes that are directly or indirectly affected by heat-induced cellular stress.

**(E) Devrim E, Ergüder IB, Kılıçoğlu B, Yaykaşlı E, Cetin R, Durak I. Effects of Electromagnetic Radiation Use on Oxidant/Antioxidant Status and DNA Turn-over Enzyme Activities in Erythrocytes and Heart, Kidney, Liver, and Ovary Tissues From Rats: Possible Protective Role of Vitamin C. Toxicol Mech Methods. 18(9):679-683, 2008.**

In this study, the aim was to investigate possible effects of Electromagnetic Radiation (EMR) use on oxidant and antioxidant status in erythrocytes and kidney, heart, liver, and ovary tissues from rats, and possible protective role of vitamin C. For this aim, 40 Wistar albino female rats were used throughout the study. The treatment group was exposed to EMR in a frequency of 900 MHz, the EMR plus vitamin C group was exposed to the same EMR frequency and given vitamin C (250 mg/kg/day) orally for 4 weeks. There were 10 animals in each group including control and vitamin C groups. At the end of the study period, blood samples were obtained from the animals to get erythrocyte sediments. Then the animals were sacrificed and heart, kidney, liver, and ovary tissues were removed. Malondialdehyde (MDA) levels and superoxide dismutase (SOD), catalase (CAT), glutathione peroxidase (GSH-Px), xanthine oxidase (XO), and adenosine deaminase (ADA) enzyme activities were measured in the tissues and erythrocytes. It was observed that MDA level, XO, and GSH-Px activities significantly increased in the EMR group as compared with those of the control group in the erythrocytes. In the kidney tissues, it was found that MDA level and CAT activity significantly increased, whereas XO and ADA activities decreased in the cellular phone group as compared with those of the control group. However, in the heart tissues it was observed that MDA level, ADA, and XO activities significantly decreased in the cellular phone group as compared with those of the control group. The results suggest that EMR at the frequency generated by a cell phone causes oxidative stress and peroxidation in the erythrocytes and kidney tissues from rats. In the erythrocytes, vitamin C seems to make partial protection against the oxidant stress.

**(E) Djordjevic B, Sokolovic D, Kocic G, Veljkovic A, Despotovic M, Basic J, Jevtovic-Stoimenov T, Sokolovic DM. The effect of melatonin on the liver of rats exposed to microwave radiation. Bratisl Lek Listy. 116(2):96-100, 2015.**

OBJECTIVES: We aimed to clarify if melatonin treatment (2 mg/kg i.p.) may favorably impact the liver tissue in rats exposed to microwave radiation. The experiment was performed on 84 six-weeks-old Wistar male rats exposed for 4h a day, for 20, 40 and 60 days,

respectively, to microwaves (900 MHz, 100-300 microT, 54-160 V/m). Rats were divided in to four groups: I (control) - rats treated with saline, II (Mel) - rats treated with melatonin, III (MWs) - microwave exposed rats, IV (MWs + Mel) - MWs exposed rats treated with melatonin. We evaluated oxidative stress parameters (malondialdehyde and carbonyl group content), catalase, xanthine oxidase, deoxyribonuclease I and II activity. **BACKGROUND:** Oxidative stress is the key mechanism of the microwave induced tissue injury. Melatonin, a lipophilic indoleamine primarily synthesized and released from the pineal gland is a powerful antioxidant. **RESULTS:** Exposure to microwaves caused an increase in malondialdehyde after 40 ( $p < 0.01$ ), protein carbonyl content after 20 ( $p < 0.05$ ), catalase ( $p < 0.05$ ) and xantine oxidase activity ( $p < 0.05$ ) after 40 days. Increase in deoxyribonuclease I activity was observed after 60 days ( $p < 0.05$ ), while deoxyribonuclease II activity was unaffected. Melatonin treatment led to malondialdehyde decrease after 40 days ( $p < 0.05$ ), but surprisingly had no effect on other analyzed parameters. **CONCLUSION:** Melatonin exerts certain antioxidant effects in the liver of rats exposed to microwaves, by diminishing the intensity of lipid peroxidation(Fig. 6, Ref. 32).

**(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.**

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  $\gamma$ -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.

**(E) 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. 2019 Nov 7;9(1):16182.**

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) Elamin AAE, Deniz OG, Kaplan S. The effects of Gum Arabic, curcumin (*Curcuma longa*) and *Garcinia kola* on the rat hippocampus after electromagnetic field exposure: A stereological and histological study. J Chem Neuroanat 120:102060, 2021.**

The present study was designed to focus on the potential effects of the electromagnetic field (EMF) emitted by mobile phones on hippocampal pyramidal neurons and to investigate the role of curcumin (Cur), *Garcinia kola* (GK) and Gum Arabic (GA) in reducing these adverse effects. Fifty-four 12-week-old male Wistar albino rats were used. These were randomly divided into nine groups of six rats each. The control, Cur, GK and GA groups were not exposed to EMF, while the sham group was kept in the EMF exposure system without being exposed to EMF. The EMF+Cur, EMF+GK, EMF+GA and EMF groups were exposed to 900 MHz EMF for one hour a day for 28 days. The number of the pyramidal neurons in the cornu ammonis (CA) of the hippocampus was estimated using the optical fractionator technique. Histopathological changes were evaluated under light and electron microscopes. The activities of the superoxide dismutase (SOD) and catalase (CAT) enzymes were also evaluated from serum samples. Significant levels of CAT and SOD activities were observed in the EMF group compared to the control group ( $p = 0.000$ ;  $p = 0.001$ ) respectively. Microscopic observations showed that dark-coloured nuclei with unclear neuron boundaries were frequently observed in the EMF group. Stereological data analysis revealed a significant decrease in the CA's total number of pyramidal neurons in the EMF group compared to the control and sham groups ( $p = 0.000$ ;  $p = 0.000$ ) respectively. Cur and GK were observed to provide significant protection in the EMF+Cur and EMF+GK groups compared to the EMF group ( $p = 0.000$ ;  $p = 0.000$ ) respectively. No significant difference was observed between the EMF+GA group and EMF group ( $p = 0.989$ ). Exposure to 900 MHz EMF causes severe alterations in the number and structure of hippocampal pyramidal neurons. Cur and GK exhibit a protective effect against these deleterious effects, but GA showed no protective effect.

**(E) Elhag MA, Nabil GM, Attia AM. Effects of electromagnetic field produced by mobile phones on the oxidant and antioxidant status of rats. Pak J Biol Sci. 10(23):4271-4274, 2007.**

This study was designed to investigate the effect of EMR produced by GSM Mobile Phones (MP) on the oxidant and antioxidant status in rats. Rats were divided into three groups: (1) controls, (2) rats exposed to a fractionated dose of EMR (15 min day<sup>-1</sup>) for four days (EMR-F) and (3) rats exposed to an acute dose of EMR (EMR-A). A net drop in the plasma concentration of vitamin C (-47 and -59.8%) was observed in EMR-F and EMR-A groups, respectively, when compared to controls. While, a significant decrease in the levels of lipophilic antioxidant vitamins: vitamin E (-33 and -65.8%), vitamin A (-44.4 and -46.8%) was observed in EMR-F and EMR-A groups, respectively, when compared to controls. A net drop in plasma level of reduced glutathione (GSH) (-19.8 and -35.3%) was observed in EMR-F and EMR-A groups, respectively. EMR exposure of rats produced a significant decrease in catalase (CAT) and superoxide dismutase (SOD) activities, with the values of these activities for EMR-A group is significantly lower than those of EMR-F. These results indicate that the effects of acute doses of EMR produced by mobile phones on the rat's antioxidant status is significantly higher than those of fractionated doses of the same type of radiation. On the basis of present results, it can be concluded that exposure to acute doses of EMR produced by mobile phones is more hazardous than that produced by fractionated doses of the same type of radiation.

**(E) Erdem Koç G, Kaplan S, Altun G, Gümüş H, Gülsüm Deniz Ö, Aydın I, Emin Onger M, Altunkaynak Z. Neuroprotective effects of melatonin and omega-3 on hippocampal cells prenatally exposed to 900 MHz electromagnetic fields. Int J Radiat Biol. 2016 Jul 21:1-6. [Epub ahead of print]**

**PURPOSE:** Adverse effects on human health caused by electromagnetic fields (EMF) associated with the use of mobile phones, particularly among young people, are increasing all the time. The potential deleterious effects of EMF exposure resulting from mobile phones being used in close proximity to the brain require particular evaluation. However, only a limited number of studies have investigated the effects of prenatal exposure to EMF in the development of the pyramidal cells using melatonin (MEL) and omega-3 ( $\omega$ -3).

**MATERIALS AND METHODS:** We established seven groups of pregnant rats consisting of three animals each; control (CONT), SHAM, EMF, EMF + MEL, MEL, EMF +  $\omega$ -3 and  $\omega$ -3 alone. The rats in the EMF, EMF + MEL, EMF +  $\omega$ -3 groups were exposed to 900 MHz EMF for 60 min/day in an exposure tube during the gestation period. The CONT, MEL and  $\omega$ -3 group rats were not placed inside the exposure tube or exposed to EMF during the study period. After delivery, only spontaneously delivered male rat pups were selected for the establishment of further groups. Each group of offspring consisted of six animals. The optical fractionator technique was used to determine total pyramidal neuron numbers in the rat hippocampal region. **RESULTS:** The total number of pyramidal cells in the cornu ammonis (CA) in the EMF group was significantly lower than in the CONT, SHAM, EMF + MEL, and EMF +  $\omega$ -3 groups. No significant difference was observed between the EMF, MEL and  $\omega$ -3 groups. No difference was also observed between any groups in terms of rats' body or brain

weights. CONCLUSION: MEL and  $\omega$ -3 can protect the cell against neuronal damage in the hippocampus induced by 900 MHz EMF. However, further studies are now needed to evaluate the chronic effects of 900 MHz EMF on the brain in the prenatal period.

**(E) Ertilav K, Uslusoy F, Ataizi S, Nazıroğlu M. Long term exposure to cell phone frequencies (900 and 1800 MHz) induces apoptosis, mitochondrial oxidative stress and TRPV1 channel activation in the hippocampus and dorsal root ganglion of rats. *Metab Brain Dis.* 33(3):753-763, 2018. Jan 13.**

Mobile phone providers use electromagnetic radiation (EMR) with frequencies ranging from 900 to 1800 MHz. The increasing use of mobile phones has been accompanied by several potentially pathological consequences, such as neurological diseases related to hippocampal (HIPPO) and dorsal root ganglion neuron (DRGN). The TRPV1 channel is activated different stimuli, including CapN, high temperature and oxidative stress. We investigated the contribution TRPV1 to mitochondrial oxidative stress and apoptosis in HIPPO and DRGN following long term exposure to 900 and 1800 MHz in a rat model. Twenty-four adult rats were equally divided into the following groups: (1) control, (2) 900 MHz, and (3) 1800 MHz exposure. Each experimental group was exposed to EMR for 60 min/ 5 days of the week during the one year. The 900 and 1800 MHz EMR exposure induced increases in TRPV1 currents, intracellular free calcium influx ( $Ca^{2+}$ ), reactive oxygen species (ROS) production, mitochondrial membrane depolarization (JC-1), apoptosis, and caspase 3 and 9 activities in the HIPPO and DRGN. These deleterious processes were further increased in the 1800 MHz experimental group compared to the 900 MHz exposure group. In conclusion, mitochondrial oxidative stress, programmed cell death and  $Ca^{2+}$  entry pathway through TRPV1 activation in the HIPPO and DRGN of rats were increased in the rat model following exposure to 900 and 1800 MHz cell frequencies. Our results suggest that exposure to 900 and 1800 MHz EMR may induce a dose-associated, TRPV1-mediated stress response.

**(E) \*Eser O, Songur A, Aktas C, Karavelioglu E, Caglar V, Aylak F, Ozguner F, Kanter M. The effect of electromagnetic radiation on the rat brain: an experimental study. *Turk Neurosurg.* 23(6):707-715, 2013.**

AIM: The aim of this study is to determine the structural changes of electromagnetic waves in the frontal cortex, brain stem and cerebellum. MATERIAL and METHODS: 24 Wistar Albino adult male rats were randomly divided into four groups: group I consisted of control rats, and groups II-IV comprised electromagnetically irradiated (EMR) with 900, 1800 and 2450 MHz. The heads of the rats were exposed to 900, 1800 and 2450 MHz microwaves irradiation for 1h per day for 2 months. RESULTS: While the histopathological changes in the frontal cortex and brain stem were normal in the control group, there were severe degenerative changes, shrunken cytoplasm and extensively dark pyknotic nuclei in the EMR groups. Biochemical analysis demonstrated that the Total Antioxidative Capacity level was significantly decreased in the EMR groups and also Total Oxidative Capacity and Oxidative Stress Index levels were significantly increased in the frontal cortex, brain stem and cerebellum. IL-1 $\beta$  level was significantly increased in the EMR groups in the brain stem.

**CONCLUSION:** EMR causes to structural changes in the frontal cortex, brain stem and cerebellum and impair the oxidative stress and inflammatory cytokine system. This deterioration can cause to disease including loss of these areas function and cancer development.

**(E) Eslami, E., Fatahian, S. & Shahanipour, K. Assessment of the Cellphone Radiations Effects on Anti-oxidative and Renal Parameters in Rat. *Toxicol. Environ. Health Sci.* 11, 278–282, 2019.**

**Objective:** In the present study an attempt is to assess the cellphones waves effects on the stress oxida-tive activities as well as renal parameters in rat. **Methods:** 30 mature male Wister rats were divided into 3 groups of 10. Group 1 (control) was kept in normal laboratory conditions. Groups 2 and 3 were exposed with two and four cellphones in opposite sides of the cage, respectively. Blood samples were collected directly from the rat's heart 15 and 30 days post exposure and the activity of anti-oxidative factors including Glutathione Peroxidase, Superoxide Dis-mutase, Catalase, Glutathione Peroxidase, Paraoxo-nase and Malondialdehyde as well as renal parameters (Urea, Uric acid and Creatinine) were measured. **Results:** Activity of catalase, superoxide dismutase and paraoxonase enzymes in all treated groups, 15 and 30 days post cellphone exposure were significantly increased in comparison with control. The activity of glutathione peroxidase enzyme in all treated groups, 15 and 30 days post cellphone exposure was decreased in comparison with control. Malondialde-hyde and creatinine activity did not show any significant changes. Uric acid and urea were significantly increased in the group just exposed with four cellphones, 30 days post exposure. **Conclusion:** According to the results, many changes were seen in biochemical parameters due to the cellphone exposure during a month. Therefore it can be concluded that radio frequency devices could be destructive and have many negative effects on the tissues and enzyme function.

**(E) Esmekaya MA, Ozer C, Seyhan N. 900 MHz pulse-modulated radiofrequency radiation induces oxidative stress on heart, lung, testis and liver tissues. *Gen Physiol Biophys.* 30(1):84-89, 2011.**

Oxidative stress may affect many cellular and physiological processes including gene expression, cell growth, and cell death. In the recent study, we aimed to investigate whether 900 MHz pulse-modulated radiofrequency (RF) fields induce oxidative damage on lung, heart and liver tissues. We assessed oxidative damage by investigating lipid peroxidation (malondialdehyde, MDA), nitric oxide (NOx) and glutathione (GSH) levels which are the indicators of tissue toxicity. A total of 30 male Wistar albino rats were used in this study. Rats were divided randomly into three groups; control group (n = 10), sham group (device off, n = 10) and 900 MHz pulsed-modulated RF radiation group (n = 10). The RF rats were exposed to 900 MHz pulsed modulated RF radiation at a specific absorption rate (SAR) level of 1.20 W/kg 20 min/day for three weeks. MDA and NOx levels were increased significantly in liver, lung, testis and heart tissues of the exposed group compared to sham and control groups ( $p < 0.05$ ). Conversely GSH levels were significantly lower in exposed rat tissues ( $p < 0.05$ ). No significantly difference was observed between sham and control groups. Results of our study showed that pulse-modulated RF radiation causes oxidative injury in liver, lung, testis and

heart tissues mediated by lipid peroxidation, increased level of NOx and suppression of antioxidant defense mechanism.

**(E) Esmekaya MA, Tuysuz MZ, Tomruk A, Canseven AG, Yücel E, Aktuna Z, Keskil S, Seyhan N. Effects of cell phone radiation on lipid peroxidation, glutathione and nitric oxide levels in mouse brain during epileptic seizure. J Chem Neuroanat. 75(pt.B):111-115, 2016.**

The objective of the this study was to evaluate the effects of cellular phone radiation on oxidative stress parameters and oxide levels in mouse brain during pentylenetetrazole (PTZ) induced epileptic seizure. Eight weeks old mice were used in the study. Animals were distributed in the following groups: Group I: Control group treated with PTZ, Group II: 15min cellular phone radiation+PTZ treatment+30min cellular phone radiation, Group III: 30min cellular phone radiation+PTZ treatment+30min cellular phone radiation. The RF radiation was produced by a 900MHz cellular phone. Lipid peroxidation, which is the indicator of oxidative stress was quantified by measuring the formation of thiobarbituric acid reactive substances (TBARS). The glutathione (GSH) levels were determined by the Ellman method. Tissue total nitric oxide (NOx) levels were obtained using the Griess assay. Lipid peroxidation and NOx levels of brain tissue increased significantly in group II and III compared to group I. On the contrary, GSH levels were significantly lower in group II and III than group I. However, no statistically significant alterations in any of the endpoints were noted between group II and Group III. Overall, the experimental findings demonstrated that cellular phone radiation may increase the oxidative damage and NOx level during epileptic activity in mouse brain.

**(E) Falone S, Sannino A, Romeo S, Zeni O, Santini SJ, Rispoli R, Amicarelli F, Scarfi MR. Protective effect of 1950 MHz electromagnetic field in human neuroblastoma cells challenged with menadione. Sci Rep. 8(1):13234, 2018.**

This study aims to assess whether a 1950 MHz radiofrequency (RF) electromagnetic field could protect human neuroblastoma SH-SY5Y cells against a subsequent treatment with menadione, a chemical agent inducing DNA damage via reactive oxygen species formation. Cells were pre-exposed for 20 h to specific absorption rate of either 0.3 or 1.25 W/kg, and 3 h after the end of the exposure, they were treated with 10  $\mu$ M menadione (MD) for 1 h. No differences were observed between sham- and RF-exposed samples. A statistically significant reduction in menadione-induced DNA damage was detected in cells pre-exposed to either 0.3 or 1.25 W/kg ( $P < 0.05$ ). Moreover, our analyses of gene expression revealed that the pre-exposure to RF almost inhibited the dramatic loss of glutathione peroxidase-based antioxidant scavenging efficiency that was induced by MD, and in parallel strongly enhanced the gene expression of catalase-based antioxidant protection. In addition, RF abolished the MD-dependent down-regulation of oxoguanine DNA glycosylase, which is a critical DNA repairing enzyme. Overall, our findings suggested that RF pre-exposure reduced menadione-dependent DNA oxidative damage, most probably by enhancing antioxidant scavenging efficiency and restoring DNA repair capability. Our results provided some insights into the molecular mechanisms underlying the RF-induced adaptive response in human neuroblastoma cells challenged with menadione.

**(NE) Fasseas MK, Fragopoulou AF, Manta AK, Skouroliakou A, Vekrellis K, Margaritis LH, Syntichaki P. Response of *Caenorhabditis elegans* to wireless devices radiation exposure. *Int J Radiat Biol.* 91(3):286-293, 2015.**

Purpose: The aim of this study was to examine the impact of electromagnetic radiation produced by GSM (Global System for Mobile communications) mobile phones, Wi-Fi (Wireless-Fidelity) routers and wireless DECT (Digital Enhanced Cordless Telecommunications) phones, on the nematode *C. elegans*. Materials and methods: We exposed synchronized populations, of different developmental stages, to these wireless devices at E-field levels below ICNIRP's (International Commission on Non-Ionizing Radiation Protection) guidelines for various lengths of time. WT (wild-type) and aging- or stress-sensitive mutant worms were examined for changes in growth, fertility, lifespan, chemotaxis, short-term memory, increased ROS (Reactive Oxygen Species) production and apoptosis by using fluorescent marker genes or qRT-PCR (quantitative Reverse Transcription-Polymerase Chain Reaction). Results: No statistically significant differences were found between the exposed and the sham/control animals in any of the experiments concerning lifespan, fertility, growth, memory, ROS, apoptosis or gene expression. Conclusions: The worm appears to be robust to this form of (pulsed) radiation, at least under the exposure conditions used.

**(E) Fatma M. Ghoneim, Eetmad A. Arafat. Histological and histochemical study of the protective role of rosemary extract against harmful effect of cell phone electromagnetic radiation on the parotid glands. *Acta Histochemica*, Available online 4 May 2016.**

Electromagnetic fields (EMFs) are a class of non-ionizing radiation (NIR) that is emitted from mobile phone. It may have hazardous effects on parotid glands. So, we aimed to investigate the histological and histochemical changes of the parotid glands of rats exposed to mobile phone and study the possible protective role of rosemary against its harmful effect. Forty adult male albino rats were used in this study. They were classified into 4 equal groups. Group I (control), group II (control receiving rosemary), group III (mobile phone exposed group) and group IV (mobile exposed, rosemary treated group). Parotid glands were dissected out for histological and histochemical study. Moreover, measurement of oxidative stress markers; malondialdehyde (MDA) and total antioxidant capacity (TAC) was done. The results of this study revealed that rosemary has protective effect through improving the histological and histochemical picture of the parotid gland in addition of its antioxidant effect. It could be concluded from the current study, that exposure of parotid gland of rat models to electromagnetic radiation of mobile phone resulted in structural changes at the level of light and electron microscopic examination which could be explained by oxidative stress effect of mobile phone. Rosemary could play a protective role against this harmful effect through its antioxidant activity.

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**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(1)43-50, 2006.**

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.

**(E) Friedman J, Kraus S, Hauptman Y, Schiff Y, Seger R. Mechanism of a short-term ERK activation by electromagnetic fields at mobile phone frequency. Biochem J. 405:559-568, 2007.**

The exposure to non-thermal microwave electromagnetic field generated by mobile phones affects the expression of many proteins. This effect on transcription and protein stability can be mediated by the mitogen-activated protein kinase (MAPK) cascades, which serve as central signaling pathways, and govern essentially all stimulated cellular processes. Indeed, a long-term exposure of cells to mobile phone irradiation results in the activation of p38MAPKs as well as the ERK/MAPKs. Here we studied the immediate effect of irradiation on the MAPK cascades, and found that ERKs, but not stress related MAPKs are rapidly activated in response to various frequencies and intensities. Using signaling inhibitors we delineated the mechanism that is involved in this activation. We found that the first step is mediated in the plasma membrane by NADH oxidase, which rapidly generates reactive oxygen species (ROS). These ROS then directly stimulate matrix metalloproteinases and allow them to cleave and release heparin binding-EGF. This secreted factor, activates EGF receptor, which in turn further activates the ERK cascade. Thus, this study demonstrates for the first time a detailed molecular mechanism by which electromagnetic irradiation by mobile phones induces the activation of the ERK cascade and thereby induces transcription and other cellular processes.

**(NE) \*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.**

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. 91:891-897, 2015.**

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) Gajski G, Garaj-Vrhovac V. Radioprotective effects of honeybee venom (*Apis mellifera*) against 915-MHz microwave radiation-induced DNA damage in wistar rat lymphocytes: in vitro study. *Int J Toxicol.* 28(2):88-98, 2009.**

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) \*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.**

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.

**(E) \*Ghanbari M<sup>1</sup>, Mortazavi SB<sup>1</sup>, Khavanin A<sup>1</sup>, Khazaei M<sup>2</sup>. The Effects of Cell Phone Waves (900 MHz-GSM Band) on Sperm Parameters and Total Antioxidant Capacity in Rats. *Int J Fertil Steril.* 7(1):21-28, 2013.**

**BACKGROUND:** There is tremendous concern regarding the possible adverse effects of cell phone microwaves. Contradictory results, however, have been reported for the effects of these waves on the body. In the present study, the effect of cell phone microwaves on sperm parameters and total antioxidant capacity was investigated with regard to the duration of exposure and the frequency of these waves. **MATERIALS AND METHODS:** This experimental study was performed on 28 adult male Wistar rats (200-250 g). The animals were randomly assigned to four groups (n=7): i. control; ii. two-week exposure to cell phone-simulated waves; iii. three-week exposure to cell phone-simulated waves; and iv. two-week exposure to cell phone antenna waves. In all groups, sperm analysis was performed based on standard methods and we determined the mean sperm total antioxidant capacity according to the ferric reducing ability of plasma (FRAP) method. Data were analyzed by one-way ANOVA followed by Tukey's test using SPSS version 16 software. **RESULTS:** The results indicated that sperm viability, motility, and total antioxidant capacity in all exposure groups decreased significantly compared to the control group ( $p < 0.05$ ). Increasing the duration of exposure from 2 to 3 weeks caused a statistically significant decrease in sperm viability and motility ( $p < 0.05$ ). **CONCLUSION:** Exposure to cell phone waves can decrease sperm viability and motility in rats. These waves can also decrease sperm total antioxidant capacity in rats and result in oxidative stress.

**(E) Ghazizadeh V, Nazırođlu M. Electromagnetic radiation (Wi-Fi) and epilepsy induce calcium entry and apoptosis through activation of TRPV1 channel in hippocampus and dorsal root ganglion of rats. Metab Brain Dis. 2014 May 3. [Epub ahead of print]**

Incidence rates of epilepsy and use of Wi-Fi worldwide have been increasing. TRPV1 is a  $\text{Ca}^{2+}$  permeable and non-selective channel, gated by noxious heat, oxidative stress and capsaicin (CAP). The hyperthermia and oxidant effects of Wi-Fi may induce apoptosis and  $\text{Ca}^{2+}$  entry through activation of TRPV1 channel in epilepsy. Therefore, we tested the effects of Wi-Fi (2.45 GHz) exposure on  $\text{Ca}^{2+}$  influx, oxidative stress and apoptosis through TRPV1 channel in the murine dorsal root ganglion (DRG) and hippocampus of pentylenetetrazol (PTZ)-induced epileptic rats. Rats in the present study were divided into two groups as controls and PTZ. The PTZ groups were divided into two subgroups namely PTZ + Wi-Fi and PTZ + Wi-Fi + capsazepine (CPZ). The hippocampal and DRG neurons were freshly isolated from the rats. The DRG and hippocampus in PTZ + Wi-Fi and PTZ + Wi-Fi + CPZ groups were exposed to Wi-Fi for 1 hour before CAP stimulation. The cytosolic free  $\text{Ca}^{2+}$ , reactive oxygen species production, apoptosis, mitochondrial membrane depolarization, caspase-3 and -9 values in hippocampus were higher in the PTZ group than in the control although cell viability values decreased. The Wi-Fi exposure induced additional effects on the cytosolic  $\text{Ca}^{2+}$  increase. However, pretreatment of the neurons with CPZ, results in a protection against epilepsy-induced  $\text{Ca}^{2+}$  influx, apoptosis and oxidative damages. In results of whole cell patch-clamp experiments, treatment of DRG with  $\text{Ca}^{2+}$  channel antagonists [thapsigargin, verapamil + diltiazem, 2-APB, MK-801] indicated that Wi-Fi exposure induced  $\text{Ca}^{2+}$  influx via the TRPV1 channels. In conclusion, epilepsy and Wi-Fi in our experimental model is involved in  $\text{Ca}^{2+}$  influx and oxidative stress-induced hippocampal and DRG death through activation of TRPV1 channels, and negative modulation of this channel activity by CPZ pretreatment may account for the neuroprotective activity against oxidative stress.

**(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.**

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) Grigor'ev IuG, Mikhaïlov VF, Ivanov AA, Mal'tsev VN, Ulanova AM, Stavrakova NM, Nikolaeva IA, Grigor'ev OA. [Autoimmune processes after long-term low-level exposure to electromagnetic fields (the results of an experiment). Part 4. Manifestation of oxidative intracellular stress-reaction after long-term non-thermal EMF exposure of rats] Radiats Biol Radioecol. 50(1):22-27, 2010. [Article in Russian]**

This paper presents the results of the study of the effects of long-term low-level exposure of rats to microwaves. Rats were exposed in far field to 2450 MHz continuous wave fields providing an incident power density at the cages of 500 microW/cm<sup>2</sup> for 7 hours daily for a total of 30 days resulting in a whole-body SAR of 0.16 +/- 0.04 W/kg. Three groups ("EMF-exposure", "sham-exposure" and cage-control) were formed, each consisting of 16 rats. Circulating antibodies (IgA, IgG and IgM) directed against 16 chemical substances were evaluated in coded serum from each group of rats by enzyme multiplied analysis (ELISA test). **An increased amount of compounds resulting from interaction of amino acids with nitric oxide (NO) or its derivatives (NO<sub>2</sub>-Tyrosine, NO-Arginine, NO-Cysteine + NO-Bovine Serum Albumin, NJ-Methionine + NO-Asparagine + No-Histidine, NO-BTrypnohan + NJ-Tyrosin), fatty acids with small chains, hydroxylated fatty acids, palmitic/myristic/oleic acid, AZE (product of oxidation of fatty**

acids) was found in blood serum from EMF-exposed rats. As a rule, antibodies to conjugated antigens were seen for IgM, rarely seen for IgG and were completely absent for IgA. The levels of antibodies were higher on day 7 after the exposure compared to those on day 14 after the exposure.

**(E) Gulati S, Yadav A, Kumar N, Priya K, Aggarwal NK, Gupta R. Phenotypic and genotypic characterization of antioxidant enzyme system in human population exposed to radiation from mobile towers. Mol Cell Biochem. 440:1-9, 2018.**

In the present era, cellular phones have changed the life style of human beings completely and have become an essential part of their lives. The number of cell phones and cell towers are increasing in spite of their disadvantages. These cell towers transmit radiation continuously without any interruption, so people living within 100s of meters from the tower receive 10,000 to 10,000,000 times stronger signal than required for mobile communication. In the present study, we have examined superoxide dismutase (SOD) enzyme activity, catalase (CAT) enzyme activity, lipid peroxidation assay, and effect of functional polymorphism of SOD and CAT antioxidant genes against mobile tower-induced oxidative stress in human population. From our results, we have found a significantly lower mean value of manganese superoxide dismutase (MnSOD) enzyme activity, catalase (CAT) enzyme activity, and a high value of lipid peroxidation assay in exposed as compared to control subjects. Polymorphisms in antioxidant MnSOD and CAT genes significantly contributed to its phenotype. In the current study, a significant association of genetic polymorphism of antioxidant genes with genetic damage has been observed in human population exposed to radiations emitted from mobile towers.

**(NE) Gulati, S, Kosik, P, Durdik, M, Skorvaga, M., Jakl, L., Markova, E., Belyaev, I. (2020). Effects of different mobile phone UMTS signals on DNA, apoptosis and oxidative stress in human lymphocytes. Environ Pollut. 267:115632. doi: 10.1016/j.envpol.2020.115632. (VT, AE, IFR)**

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(1):59-66, 2010.**

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.**

**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) 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. 90:892-896, 2014.**

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 ( $3.68 \pm 0.36$  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) Guney M, Ozguner F, Oral B, Karahan N, Mungan T. 900 MHz radiofrequency-induced histopathologic changes and oxidative stress in rat endometrium: protection by vitamins E and C. Toxicol Ind Health. 23(7):411-420, 2007.**

There are numerous reports on the effects of electromagnetic radiation (EMR) in various cellular systems. Mechanisms of adverse effects of EMR indicate that reactive oxygen species (ROS) may play a role in the biological effects of this radiation. The aims of this study were to examine 900 MHz mobile phone-induced oxidative stress that promotes production of ROS and to investigate the role of vitamins E and C, which have antioxidant properties, on endometrial tissue against possible 900 MHz mobile phone-induced endometrial impairment in rats. The animals were randomly grouped (eight each) as follows: 1) Control group (without stress and EMR, Group I), 2) sham-operated rats stayed without exposure to EMR (exposure device off, Group II), 3) rats exposed to 900 MHz EMR (EMR group, Group III) and 4) a 900 MHz EMR exposed + vitamin-treated group (EMR + Vit group, Group IV). A 900 MHz EMR was applied to EMR and EMR + Vit group 30 min/day, for 30 days using an experimental exposure device. Endometrial

levels of nitric oxide (NO, an oxidant product) and malondialdehyde (MDA, an index of lipid peroxidation), increased in EMR exposed rats while the combined vitamins E and C caused a significant reduction in the levels of NO and MDA. Likewise, endometrial superoxide dismutase (SOD), catalase (CAT) and glutathione peroxidase (GSH-Px) activities decreased in EMR exposed animals while vitamins E and C caused a significant increase in the activities of these antioxidant enzymes. In the EMR group histopathologic changes in endometrium, diffuse and severe apoptosis was present in the endometrial surface epithelial and glandular cells and the stromal cells. Diffuse eosinophilic leucocyte and lymphocyte infiltration were observed in the endometrial stroma whereas the combination of vitamins E and C caused a significant decrease in these effects of EMR. It is concluded that oxidative endometrial damage plays an important role in the 900 MHz mobile phone-induced endometrial impairment and the modulation of oxidative stress with vitamins E and C reduces the 900 MHz mobile phone-induced endometrial damage both at biochemical and histological levels.

**(E) \*Hamzany Y, Feinmesser R, Shpitzer T, Mizrahi A, Hilly O, Hod R, Bahar G, Otradnov I, Gavish M, Nagler RM. Is human saliva an indicator of the adverse health effects of using mobile phones? Antioxid Redox Signal. 18(6):622-627, 2013.**

Increasing use of mobile phones creates growing concerns regarding harmful effects of radiofrequency nonionizing electromagnetic radiation on human tissues located close to the ear, where phones are commonly held for long periods of time. We studied 20 subjects in the mobile-phone group who had a mean duration of mobile phone use of 12.5 years (range 8-15) and a mean time use of 29.6 h per month (range 8-100). Deaf individuals served as controls. We compared salivary outcomes (secretion, oxidative damage indices, flow rate, and composition) between mobile phone users and nonusers. We report a significant increase in all salivary oxidative stress indices studied in mobile phone users. Salivary flow, total protein, albumin, and amylase activity were decreased in mobile phone users. These observations lead to the hypothesis that the use of mobile phones may cause oxidative stress and modify salivary function.

**(E) \*Hancı H, Odacı E, Kaya H, Aliyazıcıoğlu Y, Turan I, Demir S, Colakoğlu S. The effect of prenatal exposure to 900-MHz electromagnetic field on the 21-old-day rat testicle. Reprod Toxicol. 42:203-209, 2013.**

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.

**(E) Hancı H, Türedi S, Topal Z, Mercantepe T, Bozkurt I, Kaya H, Ersöz Ş, Ünal B, Odacı E. Can prenatal exposure to a 900 MHz electromagnetic field affect the morphology of the spleen and thymus, and alter biomarkers of oxidative damage in 21-day-old male rats? Biotech Histochem. 2015 May 19:1-9. [Epub ahead of print]**

We investigated the effects of a 900 Megahertz (MHz) electromagnetic field (EMF), applied during the prenatal period, on the spleen and thymus of 21-day-old male rat pups. Pregnant Sprague-Dawley rats were divided into control and EMF groups. We applied 900 MHz EMF for 1 h/day to the EMF group of pregnant rats. Newborn male rat pups were removed from their mothers and sacrificed on postnatal day 21. Spleen and thymus tissues were excised and examined. Compared to the control group, thymus tissue malondialdehyde levels were significantly higher in the group exposed to EMF, while glutathione levels were significantly decreased. Increased malondialdehyde and glutathione levels were observed in splenic tissue of rats exposed to EMF, while a significant decrease occurred in superoxide dismutase values compared to controls. Transmission electron microscopy showed pathological changes in cell morphology in the thymic and splenic tissues of newborn rats exposed to EMF. Exposure to 900 MHz EMF during the prenatal period can cause pathological and biochemical changes that may compromise the development of the male rat thymus and spleen.

**(E) Hässig M, Jud F, Naegeli H, Kupper J, Spiess BM. Prevalence of nuclear cataract in Swiss veal calves and its possible association with mobile telephone antenna base stations. Schweiz Arch Tierheilkd. 151(10):471-478, 2009. (LI)**

The purpose of this study was to evaluate the prevalence of nuclear cataract in veal calves and to elucidate a possible impact by mobile phone base stations (MPBS). For this experiment a cohort study was conducted. A follow-up of the geographical location of each dam and its calf from conception through the fetal period up to slaughter was performed. The first trimester of gestation (organogenesis) was particularly emphasized. The activities of selected protective antioxidants (superoxide dismutase, catalase, glutathione peroxidase [GPx]) were assessed in aqueous humor of the eye to evaluate the redox status. Of 253 calves, 79 (32 %) had various degrees of nuclear cataract, but only 9 (3.6 %) calves had severe nuclear cataract. Results demonstrate a relation between the location of veal calves with nuclear cataracts in the first trimester of gestation and the strength of antennas. The number of antennas within 100 to 199 meters was associated with oxidative stress and there was an association between oxidative stress and the distance to the nearest MPBS. Oxidative stress was increased in eyes with cataract (OR per kilometer: 0.80, confidence interval 95 % 0.62,0.93). It has not been shown that the antennas actually affected stress. Hosmer-Lemeshow statistics showed an accuracy of 100 % in negative cases with low radiation, and only 11.11 % accuracy in positive cases with high radiation. This reflects, that there are a lot of other possibilities for nuclear cataract beside MPBS. Further studies on the influence of electromagnetic fields during embryonic development animal or person at risk are indicated.

**(E) Hässig M, Wullschleger M, Naegeli HP, Kupper J, Spiess B, Kuster N, Capstick M, Murbach M. Influence of non ionizing radiation of base stations on the activity of redox**

**proteins in bovines. BMC Vet Res. 2014 Jun 19;10(1):136. [Epub ahead of print]**

**BACKGROUND:** The influence of electromagnetic fields on the health of humans and animals is still an intensively discussed and scientifically investigated issue (Prakt Tierarz 11:15-20, 2003; Umwelt Medizin Gesellschaft 17:326-332, 2004; J Toxicol Environment Health, Part B 12:572-597, 2009). We are surrounded by numerous electromagnetic fields of variable strength, coming from electronic equipment and its power cords, from high-voltage power lines and from antennas for radio, television and mobile communication. Particularly the latter cause's controversy, as everyone likes to have good mobile reception at anytime and anywhere, whereas nobody wants to have such a base station antenna in their proximity. **RESULTS:** In this experiment, the non-ionizing radiation (NIR) has resulted in changes in the enzyme activities. Certain enzymes were disabled, others enabled by NIR. Furthermore, individual behavior patterns were observed. While certain cows reacted to NIR, others did not react at all, or even inversely. **CONCLUSION:** The present results coincide with the information from the literature, according to which NIR leads to changes in redox proteins, and that there are individuals who are sensitive to radiation and others that are not. However, the latter could not be distinctly attributed - there are cows that react clearly with one enzyme while they do not react with another enzyme at all, or even the inverse. The study approach of testing ten cows each ten times during three phases has proven to be appropriate. Future studies should however set the post-exposure phase later on.

**(E) Hatice Ş. Gürler, Birşen Bilgici, Ayşegül K. Akar, Leman Tomak & Abdülkerim Bedir. 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. International Journal of Radiation Biology. Posted online on August 4, 2014.**

*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 ( $3.68 \pm 0.36$  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) Hernández-Morales M, Shang T, Chen J, Han V, Liu C. Lipid Oxidation Induced by RF Waves and Mediated by Ferritin Iron Causes Activation of Ferritin-Tagged Ion Channels. Cell Rep. 2020 30(10):3250-3260.e7. doi: 10.1016/j.celrep.2020.02.070.**

One approach to magnetogenetics uses radiofrequency (RF) waves to activate transient receptor potential channels (TRPV1 and TRPV4) that are coupled to cellular ferritins. The mechanisms underlying this effect are unclear and controversial. Theoretical calculations suggest that the heat produced by RF fields is likely orders of magnitude weaker than needed for channel activation. Using the FeRIC (Ferritin iron Redistribution to Ion Channels) system, we have uncovered a mechanism of activation of ferritin-tagged channels via a biochemical pathway initiated by RF disturbance of ferritin and mediated by ferritin-associated iron. We show that, in cells expressing TRPVFeRIC channels, RF increases the levels of the labile iron pool in a ferritin-dependent manner. Free iron participates in chemical reactions, producing reactive oxygen species and oxidized lipids that ultimately activate the TRPVFeRIC channels. This biochemical pathway predicts a similar RF-induced activation of other lipid-sensitive TRP channels and may guide future magnetogenetic designs.

**(E) Hidisoglu E, Kantar Gok D, Er H, Akpinar D, Uysal F, Akkoyunlu G, Ozen S, Agar A, Yargicoglu P. 2100-MHz electromagnetic fields have different effects on visual evoked potentials and oxidant/antioxidant status depending on exposure duration. Brain Res. 1635:1-11, 2016.**

The purpose of the present study was to investigate the duration effects of 2100-MHz electromagnetic field (EMF) on visual evoked potentials (VEPs) and to assess lipid peroxidation (LPO), nitric oxide (NO) production and antioxidant status of EMF exposed rats. Rats were randomized to following groups: Sham rats (S1 and S10) and rats exposed to 2100-MHz EMF (E1 and E10) for 2h/day for 1 or 10 weeks, respectively. At the end of experimental periods, VEPs were recorded under anesthesia. Brain thiobarbituric acid reactive substances (TBARS) and 4-hydroxy-2-nonenal (4-HNE) levels were significantly decreased in the E1 whereas increased in the E10 compared with their control groups. While brain catalase (CAT), glutathione peroxidase (GSH-Px) activities and NO and glutathione (GSH) levels were significantly increased in the E1, reduction of superoxide dismutase (SOD) activity was detected in the same group compared with the S1. Conversely, decreased CAT, GSH-Px activities and NO levels were observed in the E10 compared with the S10. Latencies of all VEP components were shortened in the E1 compared with the S1, whereas latencies of all VEP components, except P1, were prolonged in the E10 compared with the S10. There was a positive correlation between all VEP latencies and brain TBARS and 4-HNE values. Consequently, it could be concluded that different effects of EMFs on VEPs depend on exposure duration. Additionally, our results indicated that short-term EMF could provide protective effects, while long-term EMF could have an adverse effect on VEPs and oxidant/antioxidant status.

**(NE) Hong MN, Kim BC, Ko YG, Lee YS, Hong SC, Kim T, Pack JK, Choi HD, Kim N, Lee JS. Effects of 837 and 1950 MHz radiofrequency radiation exposure alone or combined on oxidative stress in MCF10A cells. Bioelectromagnetics. 33(7):604-611, 2012.**

The aim of this study was to determine whether the exposure to either single or multiple radio-frequency (RF) radiation frequencies could induce oxidative stress in cell cultures. Exposures

of human MCF10A mammary epithelial cells to either a single frequency (837 MHz alone or 1950 MHz alone) or multiple frequencies (837 and 1950 MHz) were conducted at specific absorption rate (SAR) values of 4 W/kg for 2 h. During the exposure period, the temperature in the exposure chamber was maintained isothermally. Intracellular levels of reactive oxygen species (ROS), the antioxidant enzyme activity of superoxide dismutase (SOD), and the ratio of reduced/oxidized glutathione (GSH/GSSG) showed no statistically significant alterations as the result of either single or multiple RF radiation exposures. In contrast, ionizing radiation-exposed cells, used as a positive control, showed evident changes in all measured biological endpoints. These results indicate that single or multiple RF radiation exposure did not elicit oxidative stress in MCF10A cells under our exposure conditions.

**(NE) Hook, G. J., Spitz, D. R., Sim, J. E., Higashikubo, R., Baty, J. D., Moros, E. G. and Roti Roti, J. L. Evaluation of Parameters of Oxidative Stress after In Vitro Exposure to FMCW- and CDMA-Modulated Radiofrequency Radiation Fields. Radiat. Res. 162, 497–504, 2004.**

The goal of this study was to determine whether radiofrequency (RF) radiation is capable of inducing oxidative stress or affecting the response to oxidative stress in cultured mammalian cells. The two types of RF radiation investigated were frequency-modulated continuous-wave with a carrier frequency of 835.62 MHz (FMCW) and code division multiple access centered on 847.74 MHz (CDMA). To evaluate the effect of RF radiation on oxidative stress, J774.16 mouse macrophage cells were stimulated with  $\gamma$ -interferon (IFN) and bacterial lipopolysaccharide (LPS) prior to exposure. Cell cultures were exposed for 20–22 h to a specific absorption rate of 0.8 W/kg at a temperature of  $37.0 \pm 0.3^\circ\text{C}$ . Oxidative stress was evaluated by measuring oxidant levels, antioxidant levels, oxidative damage and nitric oxide production. Oxidation of thiols was measured by monitoring the accumulation of glutathione disulfide (GSSG). Cellular antioxidant defenses were evaluated by measuring superoxide dismutase activity (CuZnSOD and MnSOD) as well as catalase and glutathione peroxidase activity. The trypan blue dye exclusion assay was used to measure any changes in viability. The results of these studies indicated that FMCW- and CDMA-modulated RF radiation did not alter parameters indicative of oxidative stress in J774.16 cells. FMCW- and CDMA-modulated fields did not alter the level of intracellular oxidants, accumulation of GSSG or induction of antioxidant defenses in IFN/LPS-stimulated cells. Consistent with the lack of an effect on oxidative stress parameters, no change in toxicity was observed in J774.16 cells after either optimal (with or without inhibitors of nitric oxide synthase) or suboptimal stimulation.

**(E) 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.**

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'-dichlorofluorescein diacetate fluorescence probe was used to detect intracellular ROS levels, immunofluorescence was used to detect  $\gamma$ H2AX 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  $\gamma$ H2AX 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, King BV, De Iuliis GN, Aitken RJ. The effects of radiofrequency electromagnetic radiation on sperm function. *Reproduction*. 152(6):R263-R276, 2016.**

Mobile phone usage has become an integral part of our lives. However, the effects of the radiofrequency electromagnetic radiation (RF-EMR) emitted by these devices on biological systems and specifically the reproductive systems are currently under active debate. A fundamental hindrance to the current debate is that there is no clear mechanism of how such non-ionising radiation influences biological systems. Therefore, we explored the documented impacts of RF-EMR on the male reproductive system and considered any common observations that could provide insights on a potential mechanism. Among a total of 27 studies investigating the effects of RF-EMR on the male reproductive system, negative consequences of exposure were reported in 21. Within these 21 studies, 11 of the 15 that investigated sperm motility reported significant declines, 7 of 7 that measured the production of reactive oxygen species (ROS) documented elevated levels and 4 of 5 studies that probed for DNA damage highlighted increased damage due to RF-EMR exposure. Associated with this, RF-EMR treatment reduced the antioxidant levels in 6 of 6 studies that discussed this phenomenon, whereas consequences of RF-EMR were successfully ameliorated with the supplementation of antioxidants in all 3 studies that carried out these experiments. In light of this, we envisage a two-step mechanism whereby RF-EMR is able to induce mitochondrial dysfunction leading to elevated ROS production. A continued focus on research, which aims to shed light on the biological effects of RF-EMR will allow us to test and assess this proposed mechanism in a variety of cell types.

**(E) Houston BJ, Nixon B, King BV, Aitken RJ, De Iuliis GN. Probing the Origins of 1,800 MHz Radio Frequency Electromagnetic Radiation Induced Damage in Mouse Immortalized Germ Cells and Spermatozoa *in vitro*. *Front Public Health*. 6:270, 2018.**

As the use of mobile phone devices is now highly prevalent, many studies have sought to evaluate the effects of the radiofrequency-electromagnetic radiation (RF-EMR) on both human health and biology. While several such studies have shown RF-EMR is capable of inducing cellular stress, the physicobiological origin of this stress remains largely unresolved. To explore the effect of RF-EMR on the male reproductive system, we exposed cultured mouse spermatogonial GC1 and spermatocyte GC2 cell lines, as well as cauda epididymal spermatozoa to a waveguide generating continuous wave RF-EMR (1.8 GHz, 0.15 and 1.5 W/kg). This

study demonstrated that a 4 h exposure is capable of inducing the generation of mitochondrial reactive oxygen species (ROS) in populations of GC1 (7 vs. 18%;  $p < 0.001$ ) and GC2 cells (11.5 vs. 16 %;  $p < 0.01$ ), identifying Complex III of the electron transport chain (ETC) as the potential source of electrons producing ROS. Assessing the generation of ROS in the presence of an antioxidant, penicillamine, as well as measuring lipid peroxidation via 4-hydroxynonenal levels, indicated that the elevated incidence of ROS generation observed under our exposure conditions did not necessarily induce an overt cellular oxidative stress response. However, exposure to RF-EMR at 0.15 W/kg for 3 h did induce significant DNA fragmentation in spermatozoa (that was no longer significant after 4 h), assessed by the alkaline comet assay ( $p < 0.05$ ). Furthermore, this fragmentation was accompanied by an induction of oxidative DNA damage in the form of 8-hydroxy-2'-deoxyguanosine, which was significant ( $p < 0.05$ ) after spermatozoa were exposed to RF-EMR for 4 h. At this exposure time point, a decline in sperm motility ( $p < 0.05$ ) was also observed. This study contributes new evidence toward elucidating a mechanism to account for the effects of RF-EMR on biological systems, proposing Complex III of the mitochondrial ETC as the key target of this radiation.

**(E) Houston BJ, Nixon B, McEwan KE, Martin JH, King BV, Aitken RJ, De Iulius 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.**

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.

**(cancer) (E) Höytö A, Luukkonen J, Juutilainen J, Naarala J. Proliferation, oxidative stress and cell death in cells exposed to 872 MHz radiofrequency radiation and oxidants. Radiat. Res. 170(2):235-243, 2008. (WS)**

Human SH-SY5Y neuroblastoma and mouse L929 fibroblast cells were exposed to 872 MHz radiofrequency (RF) radiation using continuous waves (CW) or a modulated signal similar to that emitted by GSM mobile phones at a specific absorption rate (SAR) of 5 W/kg in isothermal conditions. To investigate possible combined effects with other agents, menadione was used to induce reactive oxygen species, and tert-butylhydroperoxide (t-BOOH) was used to induce lipid peroxidation. After 1 or 24 h of exposure, reduced cellular glutathione levels, lipid peroxidation, proliferation, caspase 3 activity, DNA fragmentation and viability were measured. Two statistically significant differences related to RF radiation were observed: Lipid peroxidation induced by t-BOOH was increased in SH-SY5Y (but not in L929) cells, and menadione-induced caspase 3 activity was increased in L929 (but not in SH-SY5Y) cells. Both differences were statistically significant only for the GSM-modulated signal. The other end points were not significantly affected in any of the experimental conditions, and no effects were observed from exposure to RF radiation alone. The positive findings may be due to chance, but they may also reflect effects that occur only in cells sensitized by chemical stress. Further studies are required to investigate the reproducibility and dose response of the possible effects.

**(E) Hu S, Peng R, Wang C, Wang S, Gao Y, Dong J, Zhou H, Su Z, Qiao S, Zhang S, Wang L, Wen X. Neuroprotective effects of dietary supplement Kang-fu-ling against high-power microwave through antioxidant action. Food Funct. 2014 Jul 24. [Epub ahead of print]**

Kang-fu-ling (KFL) is a polybotanical dietary supplement with antioxidant properties. This study aimed to evaluate the potential protective effects of KFL on cognitive deficit induced by high-power microwave (HPM) and the underlying mechanism for this neuroprotection. The electron spin resonance technique was employed to evaluate the free radical scavenging activity of KFL in vitro and KFL exhibited scavenging hydroxyl radical activity. KFL at doses of 0.75, 1.5 and 3 g kg<sup>-1</sup> and vehicle were administered orally once daily for 14 days to male Wistar rats after being exposed to 30 mW cm<sup>-2</sup> HPM for 15 minutes. KFL reversed HPM-induced memory loss and the histopathological changes in hippocampus of rats. In addition, KFL displayed a protective effect against HPM-induced oxidative stress and activated the nuclear factor-E2-related factor 2 (Nrf2) and its target genes in the hippocampus of rats. The Nrf2-antioxidant response element (ARE) signaling pathway may be involved in the neuroprotective effects of KFL against HPM-induced oxidative stress. In summary, the dietary supplement KFL is a promising natural complex, which ameliorates oxidative stress, with neuroprotective effects against HPM.

**(E) İkinci A, Mercantepe T, Unal D, Erol HS, Şahin A, Aslan A, Baş O, Erdem H, Sönmez OF, Kaya H, Odacı E. Morphological and antioxidant impairments in the spinal cord of male offspring rats following exposure to a continuous 900-MHz electromagnetic field during early and mid-adolescence. J Chem Neuroanat. 2015 Dec 17. pii: S0891-0618(15)00096-4. doi: 10.1016/j.jchemneu.2015.11.006. [Epub ahead of print]**

The effects on human health of devices emitting electromagnetic field (EMF) have become the subject of intense research among scientists due to the rapid increase in their use. Children

and adolescents are particularly attracted to the use of devices emitting EMF, such as mobile phones. The aim of this study was therefore to investigate changes in the spinal cords of male rat pups exposed to the effect of 900 megahertz (MHz) EMF. The study began with 24 Sprague Dawley male rats aged 3 weeks. Three groups containing equal numbers of rats were established - control group (CG), sham group (SG) and EMF group (EMFG). EMFG rats were placed inside an EMF cage every day between postnatal days (PD) 21 and 46 and exposed to the effect of 900MHz EMF for 1hour. SG rats were kept in the EMF cage for 1hour without being exposed to the effect of EMF. At the end of the study, the spinal cords in the upper thoracic region of all rats were removed. Tissues were collected for biochemistry, light microscopy (LM) and transmission electron microscopic (TEM) examination. Biochemistry results revealed significantly increased malondialdehyde and glutathione levels in EMFG compared to CG and SG, while SG and EMFG catalase and superoxide dismutase levels were significantly higher than those in CG. In EMFG, LM revealed atrophy in the spinal cord, vacuolization, myelin thickening and irregularities in the perikarya. TEM revealed marked loss of myelin sheath integrity and invagination into the axon and broad vacuoles in axoplasm. The study results show that biochemical alterations and pathological changes may occur in the spinal cords of male rats following exposure to 900MHz EMF for 1hour a day on PD 21-46.

**(E) Ilhan A, Gurel A, Armutcu F, Kamisli S, Iraz M, Akyol O, Ozen S. Ginkgo biloba prevents mobile phone-induced oxidative stress in rat brain. Clin Chim Acta. 340(1-2): 153-162, 2004.**

**BACKGROUND:** The widespread use of mobile phones (MP) in recent years has raised the research activities in many countries to determine the consequences of exposure to the low-intensity electromagnetic radiation (EMR) of mobile phones. Since several experimental studies suggest a role of reactive oxygen species (ROS) in EMR-induced oxidative damage in tissues, in this study, we investigated the effect of Ginkgo biloba (Gb) on MP-induced oxidative damage in brain tissue of rats. **METHODS:** Rats (EMR+) were exposed to 900 MHz EMR from MP for 7 days (1 h/day). In the EMR+Gb groups, rats were exposed to EMR and pretreated with Gb. Control and Gb-administrated groups were produced by turning off the mobile phone while the animals were in the same exposure conditions. Subsequently, oxidative stress markers and pathological changes in brain tissue were examined for each groups. **RESULTS:** Oxidative damage was evident by the: (i) increase in malondialdehyde (MDA) and nitric oxide (NO) levels in brain tissue, (ii) decrease in brain superoxide dismutase (SOD) and glutathione peroxidase (GSH-Px) activities and (iii) increase in brain xanthine oxidase (XO) and adenosine deaminase (ADA) activities. These alterations were prevented by Gb treatment. Furthermore, Gb prevented the MP-induced cellular injury in brain tissue histopathologically. **CONCLUSION:** Reactive oxygen species may play a role in the mechanism that has been proposed to explain the biological side effects of MP, and Gb prevents the MP-induced oxidative stress to preserve antioxidant enzymes activity in brain tissue.

**(E) Imge EB, Kiliçoğlu B, Devrim E, Cetin R, Durak I. Effects of mobile phone use on brain tissue from the rat and a possible protective role of vitamin C - a preliminary study. Int J Radiat Biol. 86(12):1044-1049, 2010.**

Purpose: To evaluate effects of mobile phone use on brain tissue and a possible protective role of vitamin C. Materials and methods: Forty female rats were divided into four groups randomly (Control, mobile phone, mobile phone plus vitamin C and, vitamin C alone). The mobile phone group was exposed to a mobile phone signal (900 MHz), the mobile phone plus vitamin C group was exposed to a mobile phone signal (900 MHz) and treated with vitamin C administered orally (per os). The vitamin C group was also treated with vitamin C per os for four weeks. Then, the animals were sacrificed and brain tissues were dissected to be used in the analyses of malondialdehyde (MDA), antioxidant potential (AOP), superoxide dismutase, catalase (CAT), glutathione peroxidase (GSH-Px), xanthine oxidase, adenosine deaminase (ADA) and 5'nucleotidase (5'-NT). Results: Mobile phone use caused an inhibition in 5'-NT and CAT activities as compared to the control group. GSH-Px activity and the MDA level were also found to be reduced in the mobile phone group but not significantly. Vitamin C caused a significant increase in the activity of GSH-Px and non-significant increase in the activities of 5'-NT, ADA and CAT enzymes. Conclusion: Our results suggest that vitamin C may play a protective role against detrimental effects of mobile phone radiation in brain tissue.

**(E) Irmak MK, Fadillioglu E, Gulec M, Erdogan H, Yagmurca M, Akyol O. Effects of electromagnetic radiation from a cellular telephone on the oxidant and antioxidant levels in rabbits. Cell Biochem Funct. 20(4):279-283, 2002.**

The number of reports on the effects induced by electromagnetic radiation (EMR) in various cellular systems is still increasing. Until now no satisfactory mechanism has been proposed to explain the biological effects of this radiation. Oxygen free radicals may play a role in mechanisms of adverse effects of EMR. This study was undertaken to investigate the influence of electromagnetic radiation of a digital GSM mobile telephone (900 MHz) on oxidant and antioxidant levels in rabbits. Adenosine deaminase, xanthine oxidase, catalase, myeloperoxidase, superoxide dismutase (SOD) and glutathione peroxidase activities as well as nitric oxide (NO) and malondialdehyde levels were measured in sera and brains of EMR-exposed and sham-exposed rabbits. Serum SOD activity increased, and serum NO levels decreased in EMR-exposed animals compared to the sham group. Other parameters were not changed in either group. This finding may indicate the possible role of increased oxidative stress in the pathophysiology of adverse effect of EMR. Decreased NO levels may also suggest a probable role of NO in the adverse effect.

**(E) Ismail LA, Joumaa WH, Moustafa ME. The impact of exposure of diabetic rats to 900 MHz electromagnetic radiation emitted from mobile phone antenna on hepatic oxidative stress. Electromagn Biol Med. 2019;38(4):287-296, 2019.**

The excessive exposure of patients with type 2 diabetes mellitus (T2DM) to electromagnetic radiation (EMR) from mobile phones or their base stations antenna may influence oxidative stress and development of diabetic complications. Here, we

investigated the effects of exposing type 2 diabetic rats to EMR of 900 MHz emitted from GSM mobile phone antenna for 24 hours/day over a period of 28 days on hyperglycemia and hepatic oxidative stress. Male Sprague-Dawley rats were divided into 4 groups (12 rats/group): control rats, normal rats exposed to EMR, T2DM rats generated by nicotinamide/streptozotocin administration, and T2DM rats exposed to EMR. Our results showed that the exposure of T2DM rats to EMR nonsignificantly reduced the hyperglycemia and hyperinsulinemia compared to unexposed T2DM rats. The exposure of T2DM rats to EMR for 28 days increased the hepatic levels of MDA and Nrf-2 as well as the activities of superoxide dismutase (SOD) and catalase but decreased phosphorylated Akt-2 (pAkt-2) as compared to unexposed T2DM rats. Therefore, the decrease in the hepatic pAkt-2 in T2DM rats after the exposure to EMR may result in elevated level of hepatic MDA, even though the level of Nrf-2 and the activities of SOD and catalase were increased.

**Jammoul M, Lawand N. Melatonin: a Potential Shield against Electromagnetic Waves. *Curr Neuropharmacol.* 2021 Jun 9. doi: 10.2174/1570159X19666210609163946. (Review)**

Melatonin, a vital hormone synthesized by the pineal gland, has been implicated in various physiological functions and in circadian rhythm regulation. Its role in the protection against the non-ionizing electromagnetic field (EMF), known to disrupt the body's oxidative/anti-oxidative balance, has been called into question due to inconsistent results observed across studies. This review provides the current state of knowledge on the interwoven relationship between melatonin, EMF, and oxidative stress. Based on synthesized evidence, we present a model that best describes the mechanisms underlying the protective effects of melatonin against RF/ELF-EMF induced oxidative stress. We show that the free radical scavenger activity of melatonin is enabled through reduction of the radical pair singlet-triplet conversion rate and the concentration of the triplet products. Moreover, this review aims to highlight the potential therapeutic benefits of melatonin against the detrimental effects of EMF, in general, and electromagnetic hypersensitivity (EHS), in particular.

**(E) \*\*Jelodar G, Akbari A, Nazifi S. The prophylactic effect of vitamin C on oxidative stress indexes in rat eyes following exposure to radiofrequency wave generated by a BTS antenna model. *Int J Radiat Biol.* 89(2):128-131, 2013.**

**Purpose:** This study was conducted to evaluate the effect of radiofrequency wave (RFW)-induced oxidative stress in the eye and the prophylactic effect of vitamin C on this organ by measuring the antioxidant enzymes activity including: glutathione peroxidase (GPx), superoxide dismutase (SOD) and catalase (CAT), and malondialdehyde (MDA). **Materials and methods:** Thirty-two adult male Sprague-Dawley rats were randomly divided into four experimental groups and treated daily for 45 days as follows: Control, vitamin C (L-ascorbic acid 200 mg/kg of body weight/day by gavage), test (exposed to 900 MHz RFW) and the treated group (received vitamin C in addition to exposure to RFW). At the end of the experiment all animals were sacrificed, their eyes were removed and were used for measurement of antioxidant enzymes and MDA activity. **Results:** The results indicate that exposure to RFW in the test group decreased antioxidant enzymes activity and increased MDA compared with the control groups ( $P < 0.05$ ). In the treated group vitamin C improved

antioxidant enzymes activity and reduced MDA compared to the test group ( $P < 0.05$ ).  
Conclusions: It can be concluded that RFW causes oxidative stress in the eyes and vitamin C improves the antioxidant enzymes activity and decreases MDA.

**(E) \*Jelodar G, Nazifi S, Akbari A. The prophylactic effect of vitamin C on induced oxidative stress in rat testis following exposure to 900 MHz radio frequency wave generated by a BTS antenna model. Electromagn Biol Med. 32(3):409-416, 2013.**

Radio frequency wave (RFW) generated by base transceiver station (BTS) has been reported to make deleterious effects on reproduction, possibly through oxidative stress. This study was conducted to evaluate the effect of RFW generated by BTS on oxidative stress in testis and the prophylactic effect of vitamin C by measuring the antioxidant enzymes activity, including glutathione peroxidase, superoxide dismutase (SOD) and catalase, and malondialdehyde (MDA). Thirty-two adult male Sprague-Dawley rats were randomly divided into four experimental groups and treated daily for 45 days as follows: sham, sham+vitamin C (l-ascorbic acid 200 mg/kg of body weight/day by gavage), RFW (exposed to 900 MHz RFW) 'sham' and 'RFW' animals were given the vehicle, i.e., distilled water and the RFW+vitamin C group (received vitamin C in addition to exposure to RFW). At the end of the experiment, all the rats were sacrificed and their testes were removed and used for measurement of antioxidant enzymes and MDA activity. The results indicate that exposure to RFW in the test group decreased antioxidant enzymes activity and increased MDA compared with the control groups ( $p < 0.05$ ). In the treated group, vitamin C improved antioxidant enzymes activity and reduced MDA compared with the test group ( $p < 0.05$ ). It can be concluded that RFW causes oxidative stress in testis and vitamin C improves the antioxidant enzymes activity and decreases MDA.

**(E) 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. 19:2103, 2018.**

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,  $\gamma$ H2AX, 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.

**(cancer) (E) Kahya MC, Nazıroğlu M, Ciğ B. Selenium Reduces Mobile Phone (900 MHz)-Induced Oxidative Stress, Mitochondrial Function, and Apoptosis in Breast Cancer Cells. Biol Trace Elem Res. 160: 285-293, 2014.**

Exposure to mobile phone-induced electromagnetic radiation (EMR) may affect biological systems by increasing free oxygen radicals, apoptosis, and mitochondrial depolarization levels although selenium may modulate the values in cancer. The present study was designed to investigate the effects of 900 MHz radiation on the antioxidant redox system, apoptosis, and mitochondrial depolarization levels in MDA-MB-231 breast cancer cell line. Cultures of the cancer cells were divided into four main groups as controls, selenium, EMR, and EMR + selenium. In EMR groups, the cells were exposed to 900 MHz EMR for 1 h (SAR value of the EMR was  $0.36 \pm 0.02$  W/kg). In selenium groups, the cells were also incubated with sodium selenite for 1 h before EMR exposure. Then, the following values were analyzed: (a) cell viability, (b) intracellular ROS production, (c) mitochondrial membrane depolarization, (d) cell apoptosis, and (e) caspase-3 and caspase-9 values. Selenium suppressed EMR-induced oxidative cell damage and cell viability (MTT) through a reduction of oxidative stress and restoring mitochondrial membrane potential. Additionally, selenium indicated anti-apoptotic effects, as demonstrated by plate reader analyses of apoptosis levels and caspase-3 and caspase-9 values. In conclusion, 900 MHz EMR appears to induce apoptosis effects through oxidative stress and mitochondrial depolarization although incubation of selenium seems to counteract the effects on apoptosis and oxidative stress.

**(E) Kalanjati VP, Purwantari KE, Prasetiowati L. Aluminium foil dampened the adverse effect of 2100 MHz mobile phone-induced radiation on the blood parameters and myocardium in rats. Environ Sci Pollut Res Int. 26(12):11686-11689, 2019.**

Mobile phones emit a radiofrequency radiation (RFR) that might have adverse health effects. We aimed to investigate the possible protective effects of aluminium foil (AF) as a physical shield against the RFR from mobile phones on the blood parameters and the myocardium in rats. The effects of whole body 2100 MHz with 0.84-1.86 W/kg of SAR, 4 h/day for 30 days Global System for Mobile Communications (GSM)-RFR exposure for 4 h/day for 30 days on blood parameters (i.e. haemoglobin, leucocytes, thrombocytes, erythrocyte sedimentation rate, white blood cell differential count, corticosterone, CKMB), and the histology of myocardium were investigated. Three-month-old male rats ( $n = 32$ ) were studied and randomised equally in the following four groups: K1 (non-AF non-RFR control), K2 (AF non-RFR control), P1 (non-AF RFR-exposed), P2 (AF RFR-exposed). Data were analysed with level of significance of  $p < 0.05$ . In P1, lower leucocytes and neutrophils counts with high corticosterone levels were found compared with the control groups, whilst a significantly higher CKMB was observed compared with P2 ( $p = 0.034$ ). Lower

cardiomyocyte counts congruent to the area fraction of the non-fibrotic myocardium were observed in P1 compared with the other groups ( $p < 0.01$ ). AF might decrease the inflammatory-oxidative stress on rodent's blood cells and myocardium induced by the exposures of radiofrequency radiation of the mobile phones

**(E) Kamali K, Taravati A, Sayyadi S, Gharib FZ, Maftoon H. Evidence of oxidative stress after continuous exposure to Wi-Fi radiation in rat model. Environ Sci Pollut Res Int. 25(35):35396-35403, 2018.**

Exposure to electromagnetic radiation (EMR) is rapidly increasing in everyday environment, consequently conferring potential health effects. Oxidative stress is emerging as a mechanism implicated in pathophysiology and progression of various diseases. To our knowledge, no report has been made on the status of antioxidant redox systems after continuous exposure to radiofrequency radiation emitted from a Wi-Fi access point in animal model so far. Therefore, we aimed to continuously subject rats in the experimental group to radiofrequency (RF) radiation emitted from a commercially available Wi-Fi device. Male Wister rats were exposed to 2.45 GHz RF radiation emitted from a Wi-Fi for 24 h/day for 10 consecutive weeks. In order to assess the change in antioxidant redox system of plasma after continuous exposure to a Wi-Fi device, the total antioxidant capacity of plasma, level of thiobarbituric acid reactive substances, concentration of reduced glutathione (GSH), and activity of different enzymatic antioxidants, e.g., superoxide dismutase [SOD], catalase [CAT], glutathione peroxidase [GSH-Px], and glutathione S-transferase [GST], were measured. In the Wi-Fi exposed group, a significant decrease was detected in total antioxidant capacity of plasma and the activities of several antioxidant enzymes, including CAT, GSH-Px, and SOD ( $P < 0.05$ ). Meanwhile, the GST activity was significantly increased in this group ( $P < 0.05$ ). However, no significant changes were found in GSH and TBARS levels following exposure to RF radiation. According to the results, oxidative defense system in rats exposed to Wi-Fi signal was significantly affected compared to the control group. Further studies are needed to better understand the possible biological mechanisms of EMR emitted from Wi-Fi device and relevant outcomes

**(NE) \*Kang KA, Lee HC, Lee JJ, Hong MN, Park MJ, Lee YS, Choi HD, Kim N, Ko YK, Lee JS. Effects of combined radiofrequency radiation exposure on levels of reactive oxygen species in neuronal cells. J Radiat Res. 2013 Oct 8. [Epub ahead of print]**

The objective of this study was to investigate the effects of the combined RF radiation (837 MHz CDMA plus 1950 MHz WCDMA) signal on levels of intracellular reactive oxygen species (ROS) in neuronal cells. Exposure of the combined RF signal was conducted at specific absorption rate values of 2 W/kg of CDMA plus 2 W/kg of WCDMA for 2 h. Co-exposure to combined RF radiation with either H<sub>2</sub>O<sub>2</sub> or menadione was also performed. The experimental exposure groups were incubator control, sham-exposed, combined RF radiation-exposed with or without either H<sub>2</sub>O<sub>2</sub> or menadione groups. The intracellular ROS level was measured by flow cytometry using the fluorescent probe dichlorofluorescein diacetate. Intracellular ROS levels were not consistently affected by combined RF radiation exposure

alone in a time-dependent manner in U87, PC12 or SH-SY5Y cells. In neuronal cells exposed to combined RF radiation with either H<sub>2</sub>O<sub>2</sub> or menadione, intracellular ROS levels showed no statically significant alteration compared with exposure to menadione or H<sub>2</sub>O<sub>2</sub> alone. These findings indicate that neither combined RF radiation alone nor combined RF radiation with menadione or H<sub>2</sub>O<sub>2</sub> influences the intracellular ROS level in neuronal cells such as U87, PC12 or SH-SY5Y.

**(E) Kazemi E, Mortazavi SM, Ali-Ghanbari A, Sharifzadeh S, Ranjbaran R, Mostafavi-Pour Z, Zal F, Haghani M. Effect of 900 MHz Electromagnetic Radiation on the Induction of ROS in Human Peripheral Blood Mononuclear Cells. *J Biomed Phys Eng.* 5(3):105-114, 2015.**

BACKGROUND: Despite numerous studies over a decade, it still remains controversial about the biological effects of RF EMF emitted by mobile phone telephony. OBJECTIVE: Here we investigated the effect of 900 MHz GSM on the induction of oxidative stress and the level of intracellular reactive oxygen species (ROS) in human mononuclear cells, monocytes and lymphocytes as defence system cells. METHOD: 6 ml Peripheral Blood samples were obtained from 13 healthy volunteers (21-30 year-old). Each sample was divided into 2 groups: one was exposed RF radiation emitted from a mobile phone simulator for 2 hour and the other used as control group which was not exposed to any fields. After that, mononuclear cells were isolated from peripheral blood by density gradient centrifugation in Ficoll-Paque. The intracellular ROS content in monocytes and lymphocytes was measured by the CM-H<sub>2</sub>DCFDA fluorescence probe using flowcytometry technique. RESULTS: Our results showed significant increase in ROS production after exposure in population rich in monocytes. This effect was not significant in population rich in lymphocytes in comparison with non exposed cells. CONCLUSION: The results obtained in this study clearly showed the oxidative stress induction capability of RF electromagnetic field in the portion of PBMCs mostly in monocytes, like the case of exposure to micro organisms, although the advantages or disadvantages of this effect should be evaluated.

**(E) Kerimoğlu G, Aslan A, Baş O, Çolakoğlu S, Odacı E. Adverse effects in lumbar spinal cord morphology and tissue biochemistry in Sprague Dawley male rats following exposure to a continuous 1-h a day 900-MHz electromagnetic field throughout adolescence. *J Chem Neuroanat.* 78:125-130, 2016.**

Cell phones, an indispensable element of daily life, are today used at almost addictive levels by adolescents. Adolescents are therefore becoming increasingly exposed to the effect of the electromagnetic field (EMF) emitted by cell phones. The purpose of this study was to investigate the effect of exposure to a 900-MHz EMF throughout adolescence on the lumbar spinal cord using histopathological, immunohistochemical and biochemical techniques. Twenty-four Sprague Dawley (28.3-43.9g) aged 21 days were included in the study. These were divided equally into three groups - control (CG), sham (SG) and electromagnetic (ELMAG). No procedure was performed on the CG rats until the end of the study. SG and ELMAG rats were kept inside an EMF cage (EMFC) for 1h a day every day at the same time between postnatal days 22 and 60. During this time, ELMAG rats were exposed to the effect of a 900-MHz EMF, while the SG rats were kept in the EMFC without being exposed to

EMF. At the end of the study, the lumbar regions of the spinal cords of all rats in all groups were extracted. Half of each extracted tissue was stored at -80°C for biochemical analysis, while the other half was used for histopathological and immunohistochemical analyses. In terms of histopathology, a lumbar spinal cord with normal morphology was observed in the other groups, while morphological irregularity in gray matter, increased vacuolization and infiltration of white matter into gray matter were pronounced in the ELMAG rats. The cytoplasm of some neurons in the gray matter was shrunken and stained dark, and vacuoles were observed in the cytoplasm. The apoptotic index of glia cells and neurons were significantly higher in ELMAG compared to the other groups. Biochemical analysis revealed a significantly increased MDA value in ELMAG compared to CG, while SOD and GSH levels decreased significantly. In conclusion, our study results suggest that continuous exposure to a 900-MHz EMF for 1h a day through all stages of adolescence can result in impairments at both morphological and biochemical levels in the lumbar region spinal cords of Sprague Dawley rats.

**(E) Kerimoğlu G, Hancı H, Baş O, Aslan A, Erol HS, Turgut A, Kaya H, Çankaya S, Sönmez OF, Odacı E. Pernicious effects of long-term, continuous 900-MHz electromagnetic field throughout adolescence on hippocampus morphology, biochemistry and pyramidal neuron numbers in 60-day-old Sprague Dawley male rats. J Chem Neuroanat. 77:169-175, 2016.**

The central nervous system (CNS) begins developing in the intrauterine period, a process that continues until adulthood. Contact with chemical substances, drugs or environmental agents such as electromagnetic field (EMF) during adolescence therefore has the potential to disturb the development of the morphological architecture of components of the CNS (such as the hippocampus). The hippocampus is essential to such diverse functions as memory acquisition and integration and spatial maneuvering. EMF can result in severe damage to both the morphology of the hippocampus and its principal functions during adolescence. Although children and adolescents undergo greater exposure to EMF than adults, the information currently available regarding the effects of exposure to EMF during this period is as yet insufficient. This study investigated the 60-day-old male rat hippocampus following exposure to 900 megahertz (MHz) EMF throughout the adolescent period using stereological, histopathological and biochemical analysis techniques. Eighteen male Sprague Dawley rats aged 21 days were assigned into control, sham and EMF groups on a random basis. No procedure was performed on the control group rats. The EMF group (EMFGr) was exposed to a 900-MHz EMF for 1h daily from beginning to end of adolescence. The sham group rats were held in the EMF cage but were not exposed to EMF. All rats were sacrificed at 60 days of age. Their brains were extracted and halved. The left hemispheres were set aside for biochemical analyses and the right hemispheres were subjected to stereological and histopathological evaluation. Histopathological examination revealed increased numbers of pyknotic neurons with black or dark blue cytoplasm on EMFGr slides stained with cresyl violet. Stereological analyses revealed fewer pyramidal neurons in EMFGr than in the other two groups. Biochemical analyses showed an increase in malondialdehyde and glutathione levels, but a decrease in catalase levels in EMFGr. Our results indicate that oxidative stress-related morphological damage and pyramidal neuron loss may be observed in the rat hippocampus following exposure to 900-MHz EMF throughout the adolescent period.

**(E) Kerimoğlu G, Mercantepe T, Erol HS, Turgut A, Kaya H, Çolakoğlu S, Odacı E. Effects of long-term exposure to 900 megahertz electromagnetic field on heart morphology and biochemistry of male adolescent rats. *Biotech Histochem.* 2016 Aug 11:1-10. [Epub ahead of print]**

The pathological effects of exposure to an electromagnetic field (EMF) during adolescence may be greater than those in adulthood. We investigated the effects of exposure to 900 MHz EMF during adolescence on male adult rats. Twenty-four 21-day-old male rats were divided into three equal groups: control (Cont-Gr), sham (Shm-Gr) and EMF-exposed (EMF-Gr). EMF-Gr rats were placed in an EMF exposure cage (Plexiglas cage) for 1 h/day between postnatal days 21 and 59 and exposed to 900 MHz EMF. Shm-Gr rats were placed inside the Plexiglas cage under the same conditions and for the same duration, but were not exposed to EMF. All animals were sacrificed on postnatal day 60 and the hearts were extracted for microscopic and biochemical analyses. Biochemical analysis showed increased levels of malondialdehyde and superoxide dismutase, and reduced glutathione and catalase levels in EMF-Gr compared to Cont-Gr animals. Hematoxylin and eosin stained sections from EMF-Gr animals exhibited structural changes and capillary congestion in the myocardium. The percentage of apoptotic myocardial cells in EMF-Gr was higher than in either Shm-Gr or Cont-Gr animals. Transmission electron microscopy of myocardial cells of EMF-Gr animals showed altered structure of Z bands, decreased myofilaments and pronounced vacuolization. We found that exposure of male rats to 900 MHz EMF for 1 h/day during adolescence caused oxidative stress, which caused structural alteration of male adolescent rat heart tissue.

**(E) Kerimoğlu G, Güney C, Ersöz Ş, Odacı E. A histopathological and biochemical evaluation of oxidative injury in the sciatic nerves of male rats exposed to a continuous 900-megahertz electromagnetic field throughout all periods of adolescence. *J Chem Neuroanat.* 91:1-7, 2018.**

The effects on human health of the electromagnetic field (EMF) emitted by mobile phones, used by approximately 7 billion people worldwide, have become an important subject for scientific research. Studies have suggested that the EMF emitted by mobile phones can cause oxidative stress in different tissues and age groups. Young people in adolescence, a time period when risky behaviors and dependences increase, use mobile phones more than adults. The EMF emitted by mobile phones, which are generally carried in the pocket or in bags when not in use, will very probably affect the sciatic nerve. No previous study has investigated the effect of mobile phone use in adolescence on peripheral nerve. This study was planned accordingly. Twenty-four male Sprague Dawley rats aged 21 days were divided equally into control (CGr), Sham (SGr) and EMF (EMFGr) groups. No procedure was performed on CGr rats. EMFGr were exposed to the effect of a 900-megahertz (MHz) EMF for 1 h at the same time every day between postnatal days 21-59 (the entire adolescent period) inside a cage in the EMF apparatus. SGr rats were placed inside the cage for 1 h every day without being exposed to EMF. All rats were sacrificed at the end of the study period, and 1 cm sections of sciatic nerve were extracted. Malondialdehyde (MDA), glutathione, catalase (CAT) superoxide dismutase (SOD) values were investigated biochemically in half of the right sciatic nerve tissues. The other halves of the nerve tissues were subjected to routine histopathological tissue

procedures, sectioned and stained with hematoxylin and eosin (H&E) and Masson's trichrome. Histopathological evaluation of slides stained with Masson's trichrome and H&E revealed a normal appearance in Schwann cells and axons in all groups. However, there was marked thickening in the epineurium of sciatic nerves from EMFGr rats. MDA, SOD and CAT levels were higher in EMFGr than in CGr and SGr at biochemical analyses. Apoptotic index (AI) analysis revealed a significant increase in the number of TUNEL (+) cells when EMFGr was compared with CGr and SGr. **In conclusion, our study results suggest that continuous exposure to a 900-MHz EMF for 1 h throughout adolescence can cause oxidative injury and thickening in the epineurium in the sciatic nerve in male rats.**

**(E) \*Kesari KK, Behari J. Microwave exposure affecting reproductive system in male rats. Appl Biochem Biotechnol 31:495-498, 2010. (LI)**

The object of present study is to investigate the effects of 50 GHz microwave frequency electromagnetic fields on reproductive system of male rats. Male rats of Wistar strain were used in the study. Animals 60 days old were divided into two groups-group I sham exposed and group II experimental (microwave exposed). During exposure, rats were confined in Plexiglas cages with drilled ventilation holes for 2 h a day for 45 days continuously at a specified specific absorption rate of  $8.0 \times 10^{-4}$  W/kg. After the last exposure, the rats were sacrificed immediately and sperms were collected. Antioxidant enzyme (superoxide dismutase (SOD), glutathione peroxidase (GPx), and catalase), histone kinase, apoptosis, and cell cycle were analyzed in sperm cells. Result shows a significant decrease in the level of sperm GPx and SOD activity ( $p \leq 0.05$ ), whereas catalase shows significant increase in exposed group of sperm samples as compared with control ( $p < 0.02$ ). We observed a statistically significant decrease in mean activity of histone kinase as compared to the control ( $p < 0.016$ ). The percentage of cells dividing in a spermatogenesis was estimated by analyzing DNA per cell by flow cytometry. The percentage of apoptosis in electromagnetic field exposed group shows increased ratio as compared to sham exposed ( $p < 0.004$ ). There were no significant differences in the G(0)/G phase; however, a significant decrease ( $p < 0.026$ ) in S phase was obtained. Results also indicate a decrease in percentage of G/M transition phase of cell cycle in exposed group as compared to sham exposed ( $p < 0.019$ ). We conclude that these radiations may have a significant effect on reproductive system of male rats, which may be an indication of male infertility.

**(E) Kesari KK, Behari J. Evidence for mobile phone radiation exposure effects on reproductive pattern of male rats: role of ROS. Electromagn Biol Med. 31(3):213-22, 2012.**

The relationship between radiofrequency electromagnetic fields emitted from mobile phone and infertility is a matter of continuing debate. It is postulated that these radiations may affect the reproduction pattern spell by targeting biochemistry of sperm. In an attempt to expedite the issue, 70 days old Wistar rats ( $n = 6$ ) were exposed to mobile phone radiofrequency (RF) radiation for 2 h per day for 45 days and data compared with sham exposed ( $n = 6$ ) group. A significant decrease ( $P < 0.05$ ) in the level of testosterone

and an increase in caspase-3 activity were found in the RF-exposed animals. Distortions in sperm head and mid piece of sperm mitochondrial sheath were also observed as captured by Transmission Electron Microscope (TEM). In addition, progeny from RF-exposed rats showed significant decreases in number and weight as compared with that of sham-exposed animals. A reduction in testosterone, an increase in caspase-3, and distortion in spermatozoa could be caused by overproduction of reactive oxygen species (ROS) in animals under mobile phone radiation exposure. Our findings on these biomarkers are clear indications of possible health implications of repeated exposure to mobile phone radiation.

**(E) Kesari KK, Kumar S, Behari J. Mobile phone usage and male infertility in Wistar rats. Indian J Exp Biol. 48(10):987-992, 2010.**

A significant decrease in protein kinase C and total sperm count along with increased apoptosis were observed in male Wistar rats exposed to mobile phone frequencies (2 h/day x 35 days at 0.9 W/kg specific absorption rate). The results suggest that a reduction in protein kinase activity may be related to overproduction of reactive oxygen species (ROS) under microwave field exposure. Decrease in sperm count and an increase in apoptosis may be causative factor due to mobile radiation exposure leading to infertility.

**(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.**

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, Kumar S, Behari J. 900-MHz microwave radiation promotes oxidation in rat brain. Electromagn Biol Med. 30(4):219-234, 2011.**

Recently, there have been several reports referring to detrimental effects due to radio frequency electromagnetic fields (RF-EMF) exposure. Special attention was given to investigate the effect of mobile phone exposure on the rat brain. Since the integrative

mechanism of the entire body lies in the brain, it is suggestive to analyze its biochemical aspects. For this, 35-day old Wistar rats were exposed to a mobile phone for 2 h per day for a duration of 45 days where specific absorption rate (SAR) was 0.9 W/Kg. Animals were divided in two groups: sham exposed (n = 6) and exposed group (n = 6). Our observations indicate a significant decrease ( $P < 0.05$ ) in the level of glutathione peroxidase, superoxide dismutase, and an increase in catalase activity. Moreover, protein kinase shows a significant decrease in exposed group ( $P < 0.05$ ) of hippocampus and whole brain. Also, a significant decrease ( $P < 0.05$ ) in the level of pineal melatonin and a significant increase ( $P < 0.05$ ) in creatine kinase and caspase 3 was observed in exposed group of whole brain as compared with sham exposed. Finally, a significant increase in the level of ROS (reactive oxygen species) ( $P < 0.05$ ) was also recorded. The study concludes that a reduction or an increase in antioxidative enzyme activities, protein kinase C, melatonin, caspase 3, and creatine kinase are related to overproduction of reactive oxygen species (ROS) in animals under mobile phone radiation exposure. Our findings on these biomarkers are clear indications of possible health implications.

**(E) Khalil AM, Gagaa MH, Alshamali AM. 8-Oxo-7, 8-dihydro-2'-deoxyguanosine as a biomarker of DNA damage by mobile phone radiation. Hum Exp Toxicol. 31(7):734-740, 2012.**

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, Abu Khadra KM, 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.**

Abstract 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-Oxo-dG 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) Kim JY, Kim HJ, Kim N, Kwon JH, Park MJ. Effects of radiofrequency field exposure on glutamate-induced oxidative stress in mouse hippocampal HT22 cells. *Int J Radiat Biol.* 93(2):249-256, 2017.**

**PURPOSE:** To define the impact of radiofrequency (RF) under in vitro experimental Alzheimer's disease conditions, we investigated the effect of RF radiation on glutamate-induced oxidative stress in mouse hippocampal neuronal HT22 cells. **MATERIALS AND METHODS:** Cell survival rate was measured by MTT and trypan blue exclusion assays. Cell cycle distribution, cell death, and ROS production were analyzed using flow cytometry. Expression of proteins was analyzed by Western blot. **RESULTS:** RF exposure alone had a marginal impact on cell proliferation, however significantly enhanced glutamate-induced cytotoxicity in HT22 cells. Glutamate augmented the subG1 fraction of cell cycle, annexin/propidium iodide positive cell population, and expression of cleaved poly (ADP ribose) polymerase, which were further increased by RF exposure. Glutamate induced reactive oxygen species (ROS) generation and RF exposure further upregulated it. N-acetylcysteine (NAC) treatment completely abrogated glutamate- and RF-induced ROS production followed by cell death and restored cell proliferation in HT22 cells. Finally, glutamate phosphorylated c-Jun N-terminal kinase (JNK) and RF increased this event further. Treatment with NAC and inhibitor of JNK decreased JNK phosphorylation and restored cell proliferation, respectively. **CONCLUSIONS:** Our results demonstrate that RF exposure enhanced glutamate-induced cytotoxicity by further increase of ROS production in HT22 cells.

**(E) Kim MJ, Rhee SJ. Green tea catechins protect rats from microwave-induced oxidative damage to heart tissue. *J Med Food.* 7(3):299-304, 2004.**

We investigated the effects of **green tea** catechin on oxidative damage in microwave-exposed rats. The microwave-exposed rats received one of three diets: catechin-free (MW-0C), 0.25% catechin (MW-0.25C), or 0.5% catechin (MW-0.5C). Rats were sacrificed 6 days after microwave irradiation (2.45 GHz, 15 minutes). Cytochrome P(450) levels in the MW-0C group was increased by 85% compared with normal, but was 11% and 14% lower in the MW-0.25C and MW-0.5C groups than in the MW-0C group. NADPH-cytochrome P(450) reductase activity in the MW-0C group was increased by 29%, compared with the normal

group, but was significantly less in the MW-0.25C and MW-0.5C groups. Superoxide dismutase activity in the MW-0C group was decreased by 34%, compared with the normal group, but in the MW-0.25C and MW-0.5C groups was 19% and 25% higher. The activity of glutathione peroxidase in the MW-0C group was decreased by 28% but remained near normal with catechin supplements. Superoxide radical concentrations in the MW-0C group were increased by 35%, compared with the normal group. However, superoxide radicals in the MW-0.25C and MW-0.5C groups were 11% and 12% lower, respectively, compared with the MW-0C group. Microwave irradiation significantly increased levels of thiobarbituric acid-reactive substances, carbonyl values, and lipofuscin contents, but **green tea** catechin partially overcame the effects of the microwave irradiation. In conclusion, the mixed function oxidase system was activated, the formation of superoxide radical, lipid peroxide, oxidized protein, and lipofuscin was increased, and the antioxidative defense system was weakened in heart tissue of microwave-exposed rats, but the oxidative damage was significantly reduced by catechin supplementation.

**(NE) \*Kismali G, Ozgur E, Guler G, Akcay A, Sel T, Seyhan N. The influence of 1800 MHz GSM-like signals on blood chemistry and oxidative stress in non-pregnant and pregnant rabbits. Int J Radiat Biol. 88(5):414-419, 2012.**

**PURPOSE:** Environmental electromagnetic fields originate from man-made sources, such as mobile phones and base stations, and have led to increasing public concern about their possible adverse health effects. We aimed to investigate the possible effects of radiofrequency radiation (RFR) generated from these devices on oversensitive animals, such as pregnant rabbits. **MATERIALS AND METHODS:** In the present study, the effects of whole body 1800 MHz Global System for Mobile Communications (GSM)-like RFR exposure for 15 min/day for seven days on blood chemistry and lipid peroxidation levels in both non-pregnant and pregnant New Zealand White rabbits were investigated. Thirteen-month-old rabbits were studied in the following four groups: Non-pregnant control, non-pregnant RFR-exposed, pregnant control and pregnant RFR-exposed. **RESULTS:** Lipid peroxidation, namely malondialdehyde (MDA) levels, did not change after RFR exposure. However, blood chemistry parameters, such as cholesterol (CHO), total protein (TP), albumin (ALB), uric acid, creatinin and creatine kinase (CK) and creatine kinase-myocardial band isoenzyme (CK-MB) changed due to both pregnancy and RFR exposure. **CONCLUSION:** Our investigations have been shown that no indication for oxidative stress was detected in the blood of pregnant rabbits upon RF exposure at specific conditions employed in the present study. Minor changes in some blood chemistry parameters were detected but CK-MB and CK increases were found remarkable. Studies on RFR exposure during pregnancy will help establish international standards for the protection of pregnant women from environmental RFR.

**(E) Kivrak EG, Altunkaynak BZ, Alkan I, Yurt KK, Kocaman A, Onger ME. Effects of 900-MHz radiation on the hippocampus and cerebellum of adult rats and attenuation of such effects by folic acid and *Boswellia sacra*. J Microsc Ultrastruct.;5(4):216-224,2017.**

The radiation emitted from mobile phones has various deleterious effects on human health. This study was conducted to evaluate the effects of exposure to the 900-MHz radiation electromagnetic fields (EMF) emitted by mobile phones on Ammon's horn and the dentate gyrus (DG) in the hippocampus and cerebellum of male *Wistar* albino rats. We also investigated the neuroprotective effects of the antioxidants *Boswellia sacra* (BS) and folic acid (FA) against exposure to EMF. Twenty-four adult male rats were randomly divided into four groups of six animals each, an EMF group, an EMF + FA exposure group (EFA), an EMF + BS exposure group (EBS) and a control group (Cont). The EMF, EFA and EBS groups were exposed to 900-MHz EMF radiation inside a tube once daily over 21 days (60 min/day). The Cont group was not exposed to 900-MHz EMF. The results showed that EMF caused a significant decrease in total pyramidal and granular cell numbers in the hippocampus, and DG and in Purkinje cell numbers in the cerebellum in the EMF group compared to the other groups ( $p < 0.05$ ). BS and FA attenuated the neurodegenerative effects of EMF in the hippocampus and cerebellum. Significant differences were also determined between the numbers of neurons in the EFA and EMF groups, and between the EBS and EMF groups ( $p < 0.05$ ). However, there were no significant differences among Cont, EFA and EBS ( $p > 0.05$ ). Our results may contribute to ongoing research into the effects of 900-MHz EMF exposure.

**Kivrak EG, Yurt KK, Kaplan AA, Alkan I, Altun G. Effects of electromagnetic fields exposure on the antioxidant defense system. J Microsc Ultrastruct. 2017 Oct-Dec;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.

**(E) Koohestani NV, Zavareh S, Lashkarbolouki T, Azimipour F. Exposure to cell phone induce oxidative stress in mice preantral follicles during in vitro cultivation: An experimental study. Int J Reprod Biomed (Yazd). 2019 Sep 22;17(9):637-646.**

**BACKGROUND:** Radiations emitting from mobile phones have been proposed to affect people's health, mediated by various mechanisms like induction of oxidative stress.

**OBJECTIVE:** This study aims to investigate the effect of cell phone exposure on the oxidative status of mice preantral follicles (PFs) during in vitro culture.

**MATERIALS AND METHODS:** PFs (n=2580) were isolated mechanically from 16 to 18 day-old NMRI mice (n=50) and divided into control and cell phone-exposed groups. PFs were cultured for 12 days and ovulation was induced using human chorion gonadotropin. The developmental parameters including size, survival, antral cavity formation, ovulation and oocyte maturation were assessed. In parallel, enzymatic antioxidants activities, total antioxidant capacity (TAC), and Malondialdehyde (MDA) levels were evaluated.

**RESULTS:** The diameters and the rates of survival, antrum formation, ovulation, and metaphase II oocytes of exposed PFs to cell phone were significantly lower than those of the control group ( $p \leq 0.001$ ). The PFs exposed to cell phone had significantly lower superoxide dismutase (SOD), glutathione peroxidase (GPX), and catalase (CAT) activity compared with the control group. In the cell phone exposed PFs, the TAC level was significantly lower ( $p \leq 0.001$ ) and MDA levels was significantly higher ( $p \leq 0.001$ ), compared to the those of control group.

**CONCLUSION:** Exposure to cell phone compromised the developmental competence of mice PFs by increasing oxidative stress.

**(E) Koylu H, Mollaoglu H, Ozguner F, Nazyroglu M, Delibab N. Melatonin modulates 900 Mhz microwave-induced lipid peroxidation changes in rat brain. Toxicol Ind Health. 22(5):211-216, 2006.**

Microwaves (MW) from cellular phones may affect biological systems by increasing free radicals, which may enhance lipid peroxidation levels of the brain, thus leading to oxidative damage. Melatonin is synthesized in and secreted by the pineal gland at night and exhibits anti-oxidant properties. Several studies suggest that supplementation with anti-oxidant can influence MW-induced brain damage. The present study was designed to determine the effects of MW on the brain lipid peroxidation system, and the possible protective effects of melatonin on brain degeneration induced by MW. Twenty-eight Sprague-Dawley male rats were randomly divided into three groups as follows: (1) sham-operated control group (N = 8); (2) study 900-MHz MW-exposed group (N = 8); and (3) 900-MHz MW-exposed+melatonin (100 microg/kg sc before daily MW exposure treated group) (N = 10). Cortex brain and hippocampus tissues were removed to study the levels of lipid peroxidation as malonyl dialdehyde. The levels of lipid peroxidation in the brain cortex and hippocampus increased in the MW group compared with the control group, although the levels in the hippocampus were decreased by MW+melatonin administration. The brain cortex lipid peroxidation levels were unaffected by melatonin treatment. We conclude that melatonin may prevent MW-induced oxidative changes in the hippocampus by strengthening the anti-oxidant defense system, by reducing oxidative stress products.

**(E) Kulaber A, Kerimoğlu G, Ersöz Ş, Çolakoğlu S, Odacı E. Alterations of thymic morphology and antioxidant biomarkers in 60-day-old male rats following exposure to a continuous 900 MHz electromagnetic field during adolescence. Biotech Histochem. 92(5):331-337, 2017.**

We investigated changes in thymic tissue of male rats exposed to a 900 megahertz (MHz) electromagnetic field (EMF) on postnatal days 22-59. Three groups of six 21-day-old male Sprague-Dawley rats were allocated as: control (CG), sham (SG) and EMF (EMFG) groups. No procedure was performed on the CG rats. SG rats were placed in a Plexiglas cage for 1 h every day between postnatal days 22 and 59 without exposure to EMF. EMFG rats were placed in the same cage for the same periods as the SG rats and were exposed to 900 MHz EMF. Rats were sacrificed on postnatal day 60. Sections of thymus were stained for histological assessment. Oxidant/antioxidant parameters were investigated biochemically. Malondialdehyde (MDA) levels in EMFG increased compared to the other groups. Extravascular erythrocytes were observed in the medullary/corticomedullary regions in EMFG sections. We found that 900 MHz EMF applied for 1 h/day on postnatal days 22-59 can increase tissue MDA and histopathological changes in male rat thymic tissue.

**(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.**

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) Kuzay D, Ozer C, Sirav B, Canseven AG, Seyhan N. Oxidative effects of extremely low frequency magnetic field and radio frequency radiation on testes tissues of diabetic and healthy rats. Bratisl Lek Listy. 118(5):278-282, 2017.**

With the development of technology, people are increasingly under the exposure of electromagnetic fields. Individuals with chronic diseases such as diabetes are now long-term exposed to Radio Frequency-RF radiation and extremely low frequency (ELF) magnetic fields (MFs). The purpose of this present study is to investigate oxidative effects and antioxidant parameters of ELF MFs and RF radiation on testis tissue in diabetic and healthy rats. Wistar male rats were divided into 10 groups. Intraperitoneal single dose STZ (65 mg/kg) dissolved in citrate buffer (0.1M (pH 4.5)) was injected to diabetes groups. ELF MFs and RF radiation were used as an electromagnetic exposure for 20 min/day, 5 days/week for one month. Testis tissue oxidant malondialdehyde (MDA), and antioxidants glutathione (GSH), and total nitric oxide (NOx) levels were determined. The results of ANOVA and Mann-Whitney tests were compared;  $p < 0.05$  was considered significant. ELF and RF radiation resulted in an increase in testicular tissue MDA and NOX levels ( $p < 0.05$ ), and caused a decrease in GSH levels ( $p < 0.05$ ) in both healthy and diabetic rats, yet more distinctively in diabetic rats. The most pronounced effect was recorded in D-RF + ELF group ( $p < 0.005$ ). Both radiation practices increased the oxidative stress in testis tissue while causing a decrease in antioxidant level which was more distinctive in diabetic rats (Tab. 1, Fig. 3, Ref. 30).

**(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.**

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/cm<sup>2</sup>, 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-phenylnitron (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.

**(NE) Lantow M, Schuderer J, Hartwig C, Simko M. Free Radical Release and HSP70 Expression in Two Human Immune-Relevant Cell Lines after Exposure to 1800 MHz Radiofrequency Radiation. Radiat Res. 165(1):88-94, 2006.**

The goal of this study was to investigate whether radiofrequency (RF) electromagnetic-field (EMF) exposure at 1800 MHz causes production of free radicals and/or expression of heat-shock proteins (HSP70) in human immune-relevant cell systems. Human Mono Mac 6 and K562 cells were used to examine free radical release after exposure to incubator control, sham, RF EMFs, PMA, LPS, heat (40 degrees C) or co-exposure conditions. Several signals were used: continuous-wave, several typical modulations of the Global System for Mobile Communications (GSM): GSM-non DTX (speaking only), GSM-DTX (hearing only), GSM-Talk (34% speaking and 66% hearing) at specific absorption rates (SARs) of 0.5, 1.0, 1.5 and 2.0 W/kg. Heat and PMA treatment induced a significant increase in superoxide radical anions and in ROS production in the Mono Mac 6 cells when compared to sham and/ or incubator conditions. No significant differences in free radical production were detected after RF EMF exposure or in the respective controls, and no additional effects on superoxide radical anion production were detected after co-exposure to RF EMFs+PMA or RF EMFs+LPS. The GSM-DTX signal at 2 W/kg produced a significant difference in free radical production when the data were compared to sham because of the decreasing sham value. This difference disappeared when data were compared to the incubator controls. To determine the involvement of heat-shock proteins as a possible inhibitor of free radical production, we investigated the HSP70 expression level after different RF EMF exposures; no significant effects were detected.

**(NE) Lantow M, Lupke M, Frahm J, Mattsson MO, Kuster N, Simko M. ROS release and Hsp70 expression after exposure to 1,800 MHz radiofrequency**

**electromagnetic fields in primary human monocytes and lymphocytes. Radiat Environ Biophys. 45(1):55-62, 2006.**

The aim of this study is to investigate if 1,800 MHz radiofrequency electromagnetic fields (RF-EMF) can induce reactive oxygen species (ROS) release and/or changes in heat shock protein 70 (Hsp70) expression in human blood cells, using different exposure and co-exposure conditions. Human umbilical cord blood-derived monocytes and lymphocytes were used to examine ROS release after exposure to continuous wave or different GSM signals (GSM-DTX and GSM-Talk) at 2 W/kg for 30 or 45 min of continuous or intermittent (5 min ON/5 min OFF) exposure. The cells were exposed to incubator conditions, to sham, to RF-EMF, or to chemicals in parallel. Cell stimulation with the phorbol ester phorbol-12-myristate-13-acetate (PMA; 1  $\mu$ M) was used as positive control for ROS release. To investigate the effects on Hsp70 expression, the human monocytes were exposed to the GSM-DTX signal at 2 W/kg for 45 min, or to heat treatment (42 degrees C) as positive control. ROS production and Hsp70 expression were determined by flow cytometric analysis. The data were compared to sham and/or to control values and the statistical analysis was performed by the Student's t-test ( $P < 0.05$ ). The PMA treatment induced a significant increase in ROS production in human monocytes and lymphocytes when the data were compared to sham or to incubator controls. After continuous or intermittent GSM-DTX signal exposure (2 W/kg), a significantly different ROS production was detected in human monocytes if the data were compared to sham. However, this significant difference appeared due to the lowered value of ROS release during sham exposure. In human lymphocytes, no differences could be detected if data were compared either to sham or to incubator control. The Hsp70 expression level after 0, 1, and 2 h post-exposure to GSM-DTX signal at 2 W/kg for 1 h did not show any differences compared to the incubator or to sham control.

**(NE) Lee JS, Kim JY, Kim HJ, Kim JC, Lee JS, Kim N, Park MJ. Effects of combined radiofrequency field exposure on amyloid-beta-induced cytotoxicity in HT22 mouse hippocampal neurones. J Radiat Res. 57(6):620-626, 2016.**

Alzheimer's disease (AD) is the most common progressive and irreversible neurodegenerative disease and it is caused by neuronal death in the brain. Recent studies have shown that non-ionizing radiofrequency (RF) radiation has some beneficial cognitive effects in animal models of AD. In this study, we examined the effect of combined RF radiation on amyloid-beta ( $A\beta$ )-induced cytotoxicity in HT22 rat hippocampal neurons. Treatment with  $A\beta$  suppressed HT22 cell proliferation in a concentration-dependent manner. RF exposure did not affect cell proliferation, and also had a marginal effect on  $A\beta$ -induced suppression of growth in HT22 cells. Cell cycle analysis showed that  $A\beta$  decreased the G1 fraction and increased the subG1 fraction, indicating increased apoptosis. Accordingly,  $A\beta$  increased the annexin V/propidium iodide (PI)-positive cell fraction and the degradation of poly (ADP ribose) polymerase and caspase-3 in HT22 cells. However, RF alone and the combination of  $A\beta$  and RF did not affect these events significantly.  $A\beta$  increased reactive oxygen species (ROS) generation, thereby suppressing cell proliferation. This was abrogated by N-acetylcysteine (NAC) treatment, indicating that  $A\beta$ -induced ROS generation is the main cause of suppression of proliferation. NAC also restored  $A\beta$ -induced

annexin V/PI-positive cell populations. However, RF did not have a significant impact on these events. Finally, A $\beta$  stimulated the ataxia telangiectasia and Rad3-related protein/checkpoint kinase 1 DNA single-strand breakage pathway, and enhanced beta-site amyloid precursor protein expression; RF had no effect on them. Taken together, our results demonstrate that RF exposure did not significantly affect the A $\beta$ -induced decrease of cell proliferation, increase of ROS production, or induction of cell death in these cells.

**(E) Lewicka M, Henrykowska GA, Pacholski K, Szczęśny A, Dziedziczak-Buczyńska M, Buczyński A. The impact of electromagnetic radiation of different parameters on platelet oxygen metabolism - in vitro studies. Adv Clin Exp Med. 24(1):31-35, 2015.**

BACKGROUND: Electromagnetic radiation emitted by a variety of devices, e.g. cell phones, computers and microwaves, interacts with the human body in many ways. Research studies carried out in the last few decades have not yet resolved the issue of the effect of this factor on the human body and many questions are left without an unequivocal answer. Various biological and health-related effects have not been fully recognized. Thus further studies in this area are justified. OBJECTIVES: A comparison of changes within catalase enzymatic activity and malondialdehyde concentration arising under the influence of the electromagnetic radiation emitted by car electronics, equipment used in physiotherapy and LCD monitors. MATERIAL AND METHODS: The suspension of human blood platelets at a concentration of  $1 \times 10^9/0.001 \text{ dm}^3$ , obtained from whole blood by manual apheresis, was the study material. Blood platelets were exposed to an electromagnetic field for 30 min in a laboratory stand designed for the reconstruction of the electromagnetic radiation generated by car electronics, physiotherapy equipment and LCD monitors. The changes in catalase activity and malondialdehyde concentration were investigated after the exposure and compared to the control values (unexposed material). RESULTS: An increase in catalase activity and malondialdehyde concentration was observed after 30 min exposure of platelets to EMF regardless of the radiation source. The most significant changes determining the degree of oxidative stress were observed after exposure to the EMF generated by car electronics. CONCLUSIONS: The low frequency electromagnetic fields generated by car electronics, physiotherapy equipment and LCD monitors may be a cause of oxidative stress in the human body and may lead to free radical diseases.

**(E) Li Q, Tian M, Teng J, Gao P, Tang BQ, Wu H. Radio frequency-induced superoxide accumulation affected the growth and viability of *Saccharomyces cerevisiae*. Int Microbiol. 2020 Jan 2. doi: 10.1007/s10123-019-00111-2. [Epub ahead of print]**

With the development of the electric technologies, the biological effects of electromagnetic fields (EMF) were widely studied. However, the results remain controversial and the biophysical mechanisms are still unknown. To our knowledge, little studies pay attention to the radio frequency (RF) of 2.6-5 MHz. In the present study, we investigated the effect of these radio frequencies on the growth and cell viability of *Saccharomyces cerevisiae* at very low power density below 0.1 mT. The result appeared to be time-dependent. The growth of the yeast cells was obviously

affected by the RF-EMF with a 43.5% increase when exposed for 30 h, and the growth-promoting effect decreased along with the radiation time and eventually turned to an inhibiting effect retarding growth by 20.7% at 89 h. The cell viability was improved to 70.1% at 8 h and reduced by 33.5% at 28 h. The superoxide accumulated in exposed cells as radiation time increased which may lead to the inhibition of viability and growth of the cells. However, the efficient frequency, power density, and exposure dosage await further investigation. Nevertheless, the wave band studied in this research is effective to produce biological effect, and therefore, it may provide an optional new radio frequency which is valuable for the development and utilization in therapy technique and medical use.

**(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 Spermatoocyte-Derived Cells Exposed to 1800 MHz Radiofrequency Electromagnetic Fields. Cell Physiol Biochem. 48(1):29-41, 2018.**

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 spermatoocyte-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 AMPK $\alpha$ , increased DNA damage was observed in GC-2 cells following RF-EMFs exposure, and overexpression of AMPK $\alpha$  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.

**(E) Lian HY, Lin KW, Yang C, Cai P. Generation and propagation of yeast prion [URE3] are elevated under electromagnetic field. Cell Stress Chaperones. 23(4):581-594, 2018.**

We studied the effect of 2.0 GHz radio frequency electromagnetic field (RF-EMF) and 50 Hz extremely low frequency electromagnetic field (ELF-EMF) exposure on prion generation and

propagation using two budding yeast strains, NT64C and SB34, as model organisms. Under exposure to RF-EMF or ELF-EMF, the de novo generation and propagation of yeast prions [URE3] were elevated in both strains. The elevation increased over time, and the effects of ELF-EMF occurred in a dose-dependent manner. The transcription and expression levels of the molecular chaperones Hsp104, Hsp70-Ssa1/2, and Hsp40-Ydj1 were not statistically significantly changed after exposure. Furthermore, the levels of ROS, as well as the activities of superoxide dismutase (SOD) and catalase (CAT), were significantly elevated after short-term, but not long-term exposure. This work demonstrated for the first time that EMF exposure could elevate the de novo generation and propagation of yeast prions and supports the hypothesis that ROS may play a role in the effects of EMF on protein misfolding. The effects of EMF on protein folding and ROS levels may mediate the broad effects of EMF on cell function.

**(E) Lin Y, Gao P, Guo Y, Chen Q, Lang H, Guo Q, Miao X, Li J, Zeng L, Guo G. Effects of Long-Term Exposure to L-Band High-Power Microwave on the Brain Function of Male Mice. Biomed Res Int. 2021 Sep 4;2021:2237370. doi: 10.1155/2021/2237370.**

Currently, the impact of electromagnetic field (EMF) exposure on the nervous system is an increasingly arousing public concern. The present study was designed to explore the effects of continuous long-term exposure to L-band (~ 2.0 GHz) high-power microwave (L-HPM) on brain function and related mechanisms. Forty-eight male Institute of Cancer Research (ICR) mice were exposed to L-HPM at various power densities (0.5, 1.0, and 1.5 W/m<sup>2</sup>) and the brain function was examined at different time periods after exposure. The morphology of the brain was examined by hematoxylin-eosin (HE) and deoxynucleotidyl transferase-mediated dUTP nick-end labeling (TUNEL) staining. Furthermore, cholinergic markers, oxidative stress markers, and the expression of c-fos were evaluated to identify a "potential" mechanism. The results showed that exposure to L-HPM at 1.5 W/m<sup>2</sup> can cause generalized injuries in the hippocampus (CA1 and CA3) and cerebral cortex (the first somatosensory cortex) of mice, including cell apoptosis, cholinergic dysfunction, and oxidative damage. Moreover, the deleterious effects were closely related to the power density and exposure time, indicating that long-term and high-power density exposure may be detrimental to the nervous system.

**(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. 89:993-1001, 2013b.**

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) \*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.**

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  $\alpha$ -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 K, Zhang G, Wang Z, Liu Y, Dong J, Dong X, Liu J, Cao J, Ao L, Zhang S. The protective effect of autophagy on mouse spermatocyte derived cells exposure to 1800MHz radiofrequency electromagnetic radiation. Toxicol Lett. 2014 May 8. pii: S0378-4274(14)00195-7. doi: 10.1016/j.toxlet.2014.05.004. [Epub ahead of print]**

The increasing exposure to radiofrequency (RF) radiation emitted from mobile phone use has raised public concern regarding the biological effects of RF exposure on the male reproductive system. Autophagy contributes to maintaining intracellular homeostasis under environmental stress. To clarify whether RF exposure could induce autophagy in the spermatocyte, mouse spermatocyte-derived cells (GC-2) were exposed to 1800MHz Global System for Mobile Communication (GSM) signals in GSM-Talk mode at specific absorption rate (SAR) values of 1 w/kg, 2w/kg or 4w/kg for 24h, respectively. The results indicated that the expression of LC3-II increased in a dose- and time-dependent manner with RF exposure, and showed a significant change at the SAR value of 4w/kg. The autophagosome formation and the occurrence of autophagy were further confirmed by GFP-LC3 transient transfection assay and transmission electron microscopy (TEM) analysis. Furthermore, the conversion of LC3-I to LC3-II was enhanced by co-treatment with Chloroquine (CQ), indicating autophagic flux

could be enhanced by RF exposure. Intracellular ROS levels significantly increased in a dose- and time-dependent manner after cells were exposed to RF. Pretreatment with anti-oxidative NAC obviously decreased the conversion of LC3-I to LC3-II and attenuated the degradation of p62 induced by RF exposure. Meanwhile, phosphorylated extracellular-signal-regulated kinase (ERK) significantly increased after RF exposure at the SAR value of 2w/kg and 4w/kg. Moreover, we observed that RF exposure did not increase the percentage of apoptotic cells, but inhibition of autophagy could increase the percentage of apoptotic cells. These findings suggested that autophagy flux could be enhanced by 1800MHz GSM exposure (4w/kg), which is mediated by ROS generation. Autophagy may play an important role in preventing cells from apoptotic cell death under RF exposure stress.

**(E) Liu Q, Si T, Xu X, Liang F, Wang L, Pan S. Electromagnetic radiation at 900 MHz induces sperm apoptosis through bcl-2, bax and caspase-3 signaling pathways in rats. *Reprod Health.* 12:65, 2015.**

**BACKGROUND:** The decreased reproductive capacity of men is an important factor contributing to infertility. Accumulating evidence has shown that Electromagnetic radiation potentially has negative effects on human health. However, whether radio frequency electromagnetic radiation (RF-EMR) affects the human reproductive system still requires further investigation. Therefore, The present study investigates whether RF-EMR at a frequency of 900 MHz can trigger sperm cell apoptosis and affect semen morphology, concentration, and microstructure. **METHODS:** Twenty four rats were exposed to 900 MHz electromagnetic radiation with a special absorption rate of  $0.66 \pm 0.01$  W/kg for 2 h/d. After 50d, the sperm count, morphology, apoptosis, reactive oxygen species (ROS), and total antioxidant capacity (TAC), representing the sum of enzymatic and nonenzymatic antioxidants, were investigated. Western blotting and reverse transcriptase PCR were used to determine the expression levels of apoptosis-related proteins and genes, including bcl-2, bax, cytochrome c, and capase-3. **RESULTS:** In the present study, the percentage of apoptotic sperm cells in the exposure group was significantly increased by 91.42% compared with the control group. Moreover, the ROS concentration in exposure group was increased by 46.21%, while the TAC was decreased by 28.01%. Radiation also dramatically decreased the protein and mRNA expression of bcl-2 and increased that of bax, cytochrome c, and capase-3. **CONCLUSION:** RF-EMR increases the ROS level and decreases TAC in rat sperm. Excessive oxidative stress alters the expression levels of apoptosis-related genes and triggers sperm apoptosis through bcl-2, bax, cytochrome c and caspase-3 signaling pathways.

**(E) López-Furelos A, Salas-Sánchez AA, Ares-Pena FJ, Leiro-Vidal JM, López-Martín E. Exposure to radiation from single or combined radio frequencies provokes macrophage dysfunction in the RAW 264.7 cell line. *Int J Radiat Biol.* (6):607-618, 2018.**

**PURPOSE:** The aim of this study was to determine whether exposure to radiation from single or multiple radio-frequency (RF) signals at 900 and 2450 MHz would induce effects in the RAW 264.7 cell line. **MATERIALS AND METHODS:** Cell cultures were exposed to single or combined RF for 4, 24, 48, or 72 h in a GTEM electromagnetic test chamber. At the end of the radiation exposure time, viability and cell growth were analyzed by flow cytometry, nitric oxide (NO) production was measured by colorimetry, the expression of HSP70 and TNF- $\alpha$  was ascertained by qPCR, and the phagocytic activity was observed by microscopy. **RESULTS:** NO production increased after 48 h exposure at 2450 MHz, compared with controls. The group subjected to the combined interaction of two RFs showed an increase of HSP70 after 48 h exposure and a significant increase of NO and TNF- $\alpha$  after 72 h. The phagocytic activity of macrophages decreased in all groups as exposure time increased. **CONCLUSIONS:** Our results indicated a decrease in phagocytic activity and an increase in inflammatory, cytoprotective, and cytotoxic responses in macrophages after continuous and combined exposure of multiple RF signals. Multiple RF interact in everyday life, the immune response in humans is unknown.

**(E) Lu YS, Huang BT, Huang YX. Reactive Oxygen Species Formation and Apoptosis in Human Peripheral Blood Mononuclear Cell Induced by 900 MHz Mobile Phone Radiation. Oxid Med Cell Longev. 2012:740280, 2012.**

We demonstrate that reactive oxygen species (ROS) plays an important role in the process of apoptosis in human peripheral blood mononuclear cell (PBMC) which is induced by the radiation of 900 MHz radiofrequency electromagnetic field (RFEMF) at a specific absorption rate (SAR) of  $\sim 0.4$  W/kg when the exposure lasts longer than two hours. The apoptosis is induced through the mitochondrial pathway and mediated by activating ROS and caspase-3, and decreasing the mitochondrial potential. The activation of ROS is triggered by the conformation disturbance of lipids, protein, and DNA induced by the exposure of GSM RFEMF. Although human PBMC was found to have a self-protection mechanism of releasing carotenoid in response to oxidative stress to lessen the further increase of ROS, the imbalance between the antioxidant defenses and ROS formation still results in an increase of cell death with the exposure time and can cause about 37% human PBMC death in eight hours.

**(E) Luo YP, Ma HR, Chen JW, Li JJ, Li CX. [Effect of American Ginseng Capsule on the liver oxidative injury and the Nrf2 protein expression in rats exposed by electromagnetic radiation of frequency of cell phone.] [Article in Chinese]. Zhongguo Zhong Xi Yi Jie He Za Zhi. 34(5):575-580, 2014. (In Chinese)**

**OBJECTIVE:** To observe the effect of American Ginseng Capsule (AGC) on the liver oxidative injury and the Nrf2 protein expression in the liver tissue of rats exposed by 900 MHz cell phone electromagnetic radiation. **METHODS:** Totally 40 male SD rats were randomly divided into the normal control group, the model group, the Shuifei Jibin Capsule (SJC) group, and the AGC group, 10 in each group. Rats in the normal control group were not irradiated. Rats in the rest three groups were exposed by imitated 900 MHz cellular phone for

4 h in 12 consecutive days. Meanwhile, rats in the SJC group and the AGC group were intragastrically administrated with suspension of SJC and AGC (1 mL/200 g body weight) respectively. Normal saline was administered to rats in the normal control group and the model group. The histomorphological changes of the liver tissue were observed by HE staining. Contents of malonic dialdehyde (MDA), superoxide dismutase (SOD), glutathione (GSH), and glutathione peroxidase (GSH-PX) were detected by colorimetry. The Nrf2 protein expression of hepatocytes was detected by immunohistochemical assay and Western blot. RESULTS: Compared with the normal control group, hepatocyte nucleus was atrophied or partially disappeared, the contents of liver MDA and Nrf2 protein obviously increased (P <0.05, P <0.01); contents of liver SOD and GSH decreased (P <0.05) in the model group. Compared with the model group, karyopyknosis was obviously attenuated and approached to the normal level in the SJC group and the AGC group. The contents of liver MDA and Nrf2 protein expression decreased (P <0.05), and the contents of liver SOD, GSH, and GSH-PX obviously increased (P <0.05) in the SJC group. The contents of liver MDA and the Nrf2 protein expression decreased (P <0.05), and contents of SOD and GSH obviously increased in the AGC group (P <0.01, P <0.05). CONCLUSIONS: The electromagnetic radiation induced by 900 MHz cell phone could affect the expression of Nrf2 protein, induce oxidative injury, and induce abnormal morphology of liver cells. SJC and AGC could promote the morphological recovery of the liver cells. Its mechanism might be related to affecting the expression of Nrf2 protein and attenuating oxidative damage of liver cells.

**(cancer) (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(1-2):54-58, 2009.**

The objective of the study was to investigate effects of 872MHz 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 60min 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(6):417-424, 2010.**

The aim of the present study was to investigate possible cooperative effects of radiofrequency (RF) radiation and ferrous chloride (FeCl<sub>2</sub>) 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: (1) Sham exposure (control), (2) RF radiation, (3) Chemical treatment, (4) 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<sub>2</sub> 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) Ma HR, Ma ZH, Wang GY, Song CM, Ma XL, Cao XH, Zhang GH. Impacts of exposure to 900 MHz mobile phone radiation on liver function in rats. Zhongguo Ying Yong Sheng Li Xue Za Zhi. 31(6):567-571, 2015.**

**OBJECTIVE:** To study the impacts of exposure to electromagnetic radiation (EMR) on liver function in rats. **METHODS:** Twenty adult male Sprague-Dawley rats were randomly divided into normal group and radiated group. The rats in normal group were not radiated, those in radiated group were exposed to EMR 4 h/ d for 18 consecutive days. Rats were sacrificed immediately after the end of the experiment. The serum levels of alanine aminotransferase (ALT) and aspartate aminotransferase (AST), and those of malondialdehyde (MDA) and glutathione (GSH) in liver tissue were evaluated by colorimetric method. The liver histopathological changes were observed by hematoxylin and eosin staining and the protein expression of bax and bcl- 2 in liver tissue were detected by immunohistochemical method. Terminal-deoxynucleotidyl transferase mediated nick and labelling (TUNEL) method was used for analysis of apoptosis in liver. **RESULTS:** Compared with the normal rats, the serum levels of ALT and AST in the radiated group had no obvious changes ( $P > 0.05$ ), while the contents of MDA increased ( $P < 0.01$ ) and those of GSH decreased ( $P < 0.01$ ) in liver tissues. The histopathology examination showed diffuse hepatocyte swelling and vacuolation, small pieces and focal necrosis. The immunohistochemical results displayed that the expression of the bax protein was higher and that of bcl-2 protein was lower in radiated group. The hepatocyte apoptosis rates in radiated group was higher than that in normal group (all  $P < 0.01$ ). **CONCLUSION:** The exposure to **900 MHz** mobile phone 4 h/d for 18 days could induce the liver histological changes, which may be partly due to the apoptosis and oxidative stress induced in liver tissue by electromagnetic radiation.

**(E) \*Maaroufi K, Save E, Poucet B, Sakly M, Abdelmelek H, Had-Aissouni L. Oxidative stress and prevention of the adaptive response to chronic iron overload in the brain of young adult rats exposed to a 150 kilohertz electromagnetic field. Neuroscience. 186:39-47, 2011.**

Iron surcharge may induce an oxidative stress-based decline in several neurological functions. In addition, electromagnetic fields (EMF) of frequencies up to about 100 kHz, emitted by electric/electronic devices, have been suggested to enhance free radical production through an iron dependent pathway. The purpose of this study was therefore to determine a possible relationship between iron status, exposure to EMF, and brain oxidative stress in young adult rats. Samples were micro-dissected from prefrontal cortex, hippocampus, striatum, and cerebellum after chronic saline or iron overload (IO) as well as after chronic sham exposure or exposure to a 150 kHz EMF or after combining EMF exposure with IO. The brain samples were used to monitor oxidative stress-induced lipid peroxidation and activity of the antioxidant enzymes superoxide dismutase and catalase. While IO did not induce any oxidative stress in young adult rats, it stimulated antioxidant defenses in the cerebellum and prefrontal cortex in particular. On the contrary, EMF exposure stimulated lipid peroxidation mainly in the cerebellum, without affecting antioxidant defenses. When EMF was coapplied with IO, lipid peroxidation was further increased as compared to EMF alone while the increase in antioxidant defenses triggered by the sole IO was abolished. These data suggest that EMF exposure may be harmful in young adults by impairing the antioxidant defenses directed at preventing iron-induced oxidative stress.

**(NE) Maaroufi K, Had-Aissouni L, Melon C, Sakly M, Abdelmelek H, Poucet B, Save E. Spatial learning, monoamines and oxidative stress in rats exposed to 900MHz electromagnetic field in combination with iron overload. Behav Brain Res. 258:80-89, 2014. (AS, CE, BE, CH)**

The increasing use of mobile phone technology over the last decade raises concerns about the impact of high frequency electromagnetic fields (EMF) on health. More recently, a link between EMF, iron overload in the brain and neurodegenerative disorders including Parkinson's and Alzheimer's diseases has been suggested. Co-exposure to EMF and brain iron overload may have a greater impact on brain tissues and cognitive processes than each treatment by itself. To examine this hypothesis, Long-Evans rats submitted to 900MHz exposure or combined 900MHz EMF and iron overload treatments were tested in various spatial learning tasks (navigation task in the Morris water maze, working memory task in the radial-arm maze, and object exploration task involving spatial and non spatial processing). Biogenic monoamines and metabolites (dopamine, serotonin) and oxidative stress were measured. Rats exposed to EMF were impaired in the object exploration task but not in the navigation and working memory tasks. They also showed alterations of monoamine content in several brain areas but mainly in the hippocampus. Rats that received combined treatment did not show greater behavioral and neurochemical deficits than EMF-exposed rats. None of the two treatments produced global oxidative stress. These results show that there is an impact of EMF on the brain and cognitive processes but this impact is revealed only in a task exploiting spontaneous exploratory activity. In contrast, there are no synergistic effects between EMF and a high content of iron in the brain.

**(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.**

**BACKGROUND:** The daily use by people of wireless communication devices has increased exponentially in the last decade, begetting concerns regarding its potential health hazards. **METHODS:** *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. **RESULTS:** 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 ( $\geq 1.25x$ ,  $P < 0.05$ ) and associated with multiple and critical biological processes, such as basic metabolism and cellular subroutines related to stress response and apoptotic death. **CONCLUSION:** 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) Mailankot M, Kunnath AP, Jayalekshmi H, Koduru B, Valsalan R. Radio frequency electromagnetic radiation (RF-EMR) from GSM (0.9/1.8GHz) mobile phones induces oxidative stress and reduces sperm motility in rats. Clinics (Sao Paulo). 64(6):561-565, 2009.**

INTRODUCTION: Mobile phones have become indispensable in the daily lives of men and women around the globe. As cell phone use has become more widespread, concerns have mounted regarding the potentially harmful effects of RF-EMR from these devices.

OBJECTIVE: The present study was designed to evaluate the effects of RF-EMR from mobile phones on free radical metabolism and sperm quality. MATERIALS AND

METHODS: Male albino Wistar rats (10-12 weeks old) were exposed to RF-EMR from an active GSM (0.9/1.8 GHz) mobile phone for 1 hour continuously per day for 28 days.

Controls were exposed to a mobile phone without a battery for the same period. The phone was kept in a cage with a wooden bottom in order to address concerns that the effects of exposure to the phone could be due to heat emitted by the phone rather than to RF-EMR alone. Animals were sacrificed 24 hours after the last exposure and tissues of interest were harvested. RESULTS: One hour of exposure to the phone did not significantly change facial temperature in either group of rats. No significant difference was observed in total sperm count between controls and RF-EMR exposed groups. However, rats exposed to RF-EMR exhibited a significantly reduced percentage of motile sperm. Moreover, RF-EMR exposure resulted in a significant increase in lipid peroxidation and low GSH content in the testis and epididymis. CONCLUSION: Given the results of the present study, we speculate that RF-EMR from mobile phones negatively affects semen quality and may impair male fertility.

**(E) \*Manta AK, Stravopodis DJ, Papassideri IS, Margaritis LH. Reactive oxygen species elevation and recovery in Drosophila bodies and ovaries following short-term and long-term exposure to DECT base EMF. Electromagn Biol Med. 33(2):118-131, 2014. (LI)**

Abstract The objective of this study was to approach the basic mechanism(s) underlying reported ovarian apoptotic cell death and fecundity decrease induced by nonionizing radiation (NIR) in *Drosophila melanogaster*. ROS (Reactive Oxygen Species) levels were measured in the bodies and the ovaries of (sexually mature) 4-day-old flies, following exposure for 0.5, 1, 6, 24 and 96 h to a wireless DECT (Digital Enhanced Cordless Telephone) base radiation (1.88-1.90 GHz). Electrical field intensity was 2.7 V/m, measured within the fly vials and calculated SAR (Specific Absorption Rate) value = 0.009 W/Kg. Male and female bodies showed twofold increase in ROS levels ( $p < 0.001$ ) after 6 h of exposure, slightly increasing with more irradiation (24 and 96 h).

Ovaries of exposed females had a quick response in ROS increase after 0.5 h (1.5-fold,  $p < 0.001$ ), reaching 2.5-fold after 1 h with no elevation thereafter at 6, 24 and 96 h. ROS levels returned to normal, in the male and the female bodies 24 h after 6 h of exposure of the flies ( $p < 0.05$ ) and in the ovaries 4 h after 1 h exposure of the females ( $p < 0.05$ ). It is postulated that the pulsed (at 100 Hz rate and 0.08 ms duration) idle state of the DECT base radiation is capable of inducing free radical formation albeit the very low SAR, leading rapidly to accumulation of ROS in a level-saturation manner under continuous exposure, or in a recovery manner after interruption of radiation, possibly due to activation of the antioxidant machinery of the organism.

**(E) Marconi A, Tasteyre A, de Seze R, Fogel P, Simoneau G, Conti M, Sarbach C, Young SS, Gilbert J-E, Thomas Y. Journal Scientific Exploration. 29 (3): 449-465, 2015.**

A multidisciplinary project was conducted to study the possible biological impact of mobile phone emissions. As part of that project, we conducted a pilot study on 18 human volunteers, with the treatment being GSM mobile phone exposure. The volunteers were randomized and the study was a double-blind, crossover design. Two categories of oxidative stress biomarkers were followed and measured in blood and exhaled air: those assessing oxidative attacks of cell membrane lipids (malondialdehyde, exhaled alkanes, aldehydes, and isoprene) and those accounting for the organism's antioxidant defense systems (superoxyde dismutase, glutathion Peroxydase, and exhaled halogenated alkanes). The overall entropy of the system with and without GSM exposure was then calculated for each volunteer, using a statistical approach based on the global entropic difference of raw data. A significant modulation of organization of the biomarkers after 30 minutes of mobile phone exposure was found, as evidenced by a decreased entropy of the dataset associated to the emitting mobile phone condition. While these results illustrate neither deleterious effects nor the innocuity of mobile phone use, they nonetheless constitute evidence of actual interactions of these wavelengths with complex biological systems. These results will need to be confirmed in larger, future studies.

**(E) Marjanovic AM, Pavicic I, Trosic I, Cell oxidation–reduction imbalance after modulated radiofrequency radiation. Electromagn Biol Med. 34(4):381-386, 2015.**

Aim of this study was to evaluate an influence of modulated radiofrequency field (RF) of 1800 MHz, strength of 30 V/m on oxidation–reduction processes within the cell. The assigned RF field was generated within Gigahertz Transversal Electromagnetic Mode cell equipped by signal generator, modulator, and amplifier. Cell line V79, was irradiated for 10, 30, and 60 min, specific absorption rate was calculated to be 1.6 W/kg. Cell metabolic activity and viability was determined by MTT assay. In order to define total protein content, colorimetric method was used. Concentration of oxidised proteins was evaluated by enzyme-linked immunosorbent assay. Reactive oxygen species (ROS) marked with fluorescent probe 2',7'-dichlorofluorescein diacetate were measured by means of plate reader device. In comparison with control cell samples, metabolic activity and total protein content in exposed cells did not differ significantly. Concentrations of carbonyl derivates, a product of protein oxidation, insignificantly but continuously increase with duration of exposure. In exposed samples, ROS level significantly ( $p < 0.05$ ) increased after 10 min of exposure. Decrease in ROS level was observed after 30-min treatment indicating antioxidant defence mechanism activation. In

conclusion, under the given laboratory conditions, modulated RF radiation might cause impairment in cell oxidation–reduction equilibrium within the growing cells.

**(E) Marjanovic Cermak AM, Pavicic I, Tariba Lovakovic B, Pizent A, Trosic I. In vitro non-thermal oxidative stress response after 1800 MHz radiofrequency radiation. Gen Physiol Biophys. 36(4): 407-414, 2017.**

In this study possible connection between radiofrequency exposure (RF) and development of oxidative stress was investigated by measuring impairment in cellular oxidation-reduction balance immediately after RF exposure. Fibroblast cells V79 were exposed for 10, 30 and 60 minutes to 1800 MHz RF radiation. Electric field strength was 30 V/m and specific absorption rate (SAR) was calculated to be 1.6 W/kg. Electromagnetic field was generated within Gigahertz Transversal Electromagnetic Mode cell (GTEM) equipped by signal generator, amplifier and modulator. Cell viability was determined by CCK-8 colorimetric assay and level of reactive oxygen species (ROS) was detected by dihydroethidium staining. Reduced glutathione (GSH) and glutathione peroxidase (GSH-Px) were used to assess cell antioxidant activity while lipid oxidative damage was evaluated measuring concentration of malondialdehyde. Viability of V79 cells remained within normal physiological values regardless of exposure time. Increased level of superoxide radicals was detected after 60-min exposure. Significantly higher GSH level was observed immediately after 10-min exposure with higher but insignificant activity of GSH-Px. Lipid oxidative damage in exposed cell samples was not observed. Short-term RF exposure revealed transient oxidation-reduction imbalance in fibroblast cells following adaptation to applied experimental conditions.

**(cancer) (E) Marjanovic Cermak AM, Pavicic I, Trosic I. Oxidative stress response in SH-SY5Y cells exposed to short-term 1800 MHz radiofrequency radiation. J Environ Sci Health A Tox Hazard Subst Environ Eng. 53(2):132-138, 2018.**

The exact mechanism that could explain the effects of radiofrequency (RF) radiation exposure at non-thermal level is still unknown. Increasing evidence suggests a possible involvement of reactive oxygen species (ROS) and development of oxidative stress. To test the proposed hypothesis, human neuroblastoma cells (SH-SY5Y) were exposed to 1800 MHz short-term RF exposure for 10, 30 and 60 minutes. Electric field strength within Gigahertz Transverse Electromagnetic cell (GTEM) was 30 V m<sup>-1</sup> and specific absorption rate (SAR) was calculated to be 1.6 W kg<sup>-1</sup>. Cellular viability was measured by MTT assay and level of ROS was determined by fluorescent probe 2',7'-dichlorofluorescein diacetate. Concentrations of malondialdehyde and protein carbonyls were used to assess lipid and protein oxidative damage and antioxidant activity was evaluated by measuring concentrations of total glutathione (GSH). After radiation exposure, viability of irradiated cells remained within normal physiological values. Significantly higher ROS level was observed for every radiation exposure time. After 60 min of exposure, the applied radiation caused significant lipid and protein damage. The highest GSH concentration was detected after 10 minute-exposure. The results of our study showed enhanced susceptibility of SH-SY5Y cells for development of oxidative stress even after short-term RF exposure.

**(E) Masoumi A, Karbalaeei N, Mortazavi SMJ, Shabani M. Radiofrequency radiation emitted from Wi-Fi (2.4 GHz) causes impaired insulin secretion and increased oxidative stress in rat pancreatic islets. Int J Radiat Biol. 94(9):850-857, 2018.**

**PURPOSE:** There is a great concern regarding the possible adverse effects of electromagnetic radiation (EMR). This study investigated the effects of EMR induced by Wi-Fi (2.45 GHz) on insulin secretion and antioxidant redox systems in the rat pancreas. **MATERIALS AND METHODS:** Adult male Sprague-Dawley rats in the weight range of 230-260 g were divided into control, sham, Wi-Fi exposed groups. After long-term exposure (4 h/day for 45 days) to Wi-Fi EMR, plasma levels of glucose and insulin during intraperitoneal glucose tolerance test were measured. Islet insulin secretion and content, lipid peroxidation, and antioxidant status in pancreas of rats were determined. **RESULTS:** Our data showed that the weight gain in the Wi-Fi exposed group was significantly lower than the control group ( $p < .05$ ). Wi-Fi (2.45 GHz)-exposed group showed hyperglycemia. Plasma insulin level and glucose-stimulated insulin secretion from pancreatic islet were significantly reduced in the Wi-Fi-exposed group. EMR emitted from Wi-Fi caused a significant increase in lipid peroxidation and a significant decrease in GSH level, SOD, and GPx activities of the pancreas. **CONCLUSIONS:** These data showed that EMR of Wi-Fi leads to hyperglycemia, increased oxidative stress, and impaired insulin secretion in the rat pancreatic islets.

**(E) Marzook EA, Abd El Moneim AE, Elhadary AA. Protective role of sesame oil against mobile phone base station-induced oxidative stress. J Rad Res Appl Sci 7(1):1-6, 2014.**

The present study was undertaken to shed the light on the environmental threats associated with the wireless revolution and the health hazards associated with exposure to mobile base station (MBS). Besides, studying the possible protective role of sesame oil (SO) as an antioxidant against oxidative stress. Therefore, the present work was designed to study the effect of chronic exposure to electromagnetic radiations (EMR), produced by a cellular tower for mobile phone and the possible protective role of sesame oil on glutathione reductase (GSH-Rx), superoxide dismutase (SOD), catalase (CAT), total testosterone and lipid profile (total cholesterol (Tch), triglycerides (TG), low density lipoprotein cholesterol (LDL-c) and high density lipoprotein cholesterol (HDL-c) in male albino rats. Rats were arranged into four groups: the control unexposed, the exposed untreated and the exposed treated groups (1.5 and 3 ml oil). Exposed groups were subjected to electromagnetic field at frequency of 900 MHz, for 24 h/day for 8 weeks, at the same time both treated groups were supplied with oral injection of sesame oil three times per week. At the end of the experiment, blood samples were obtained for determination of the above mentioned variables in serum. The results obtained revealed that TG and testosterone were raised significantly over control in all groups and the significant increase in oil groups occurred in dose dependent manner. SOD and CAT activities were reduced significantly in exposed rats than control and increased significantly in sesame oil groups as the dose of oil increased. Total cholesterol only showed remarkable reduction in the group treated with 3 ml sesame oil. Also, in this latter group, significant elevation of GSH-Rx was recorded. Changes in serum HDL-c and LDL-c followed an

opposite trend in exposed and sesame oil groups reflecting their affectation by EMR or sesame oil. In conclusion, all results of the current study proved that sesame oil can be used as an edible oil to attenuate the oxidative stress which could be yielded as a result of chronic exposure to EMR.

**(NE) McNamee JP, Grybas VS, Qutob SS, Bellier PV Effects of 1800 MHz radiofrequency fields on signal transduction and antioxidant proteins in human A172 glioblastoma cells. Int J Radiat Biol 2021 May 28;1-34. doi: 10.1080/09553002.2021.1934751. Online ahead of print.**

**Purpose:** To assess the effects of 1800 MHz radiofrequency electromagnetic field (RF-EMF) exposure on the expression of signal transduction and antioxidant proteins in a human-derived A172 glioblastoma cell line. **Materials and methods:** Adherent human-derived A172 glioblastoma cells ( $1.0 \times 10^5$  cells per 35 mm culture dish, containing 2 mL DMEM media) were exposed to 1800 MHz continuous-wave (CW) or GSM-modulated RF fields, in the presence or absence of serum for 5, 30 or 240 min at a specific absorption rate (SAR) of 0 (sham) or 2.0 W/kg. Concurrent negative (vehicle) and positive controls (1  $\mu\text{g}/\text{mL}$  anisomycin) were included in each experiment. Cell lysates were collected immediately after exposure, stabilized by protease and phosphatase inhibitors in lysis buffer, then frozen and maintained at  $-80^\circ\text{C}$  until analysis. The relative expression levels of phosphorylated- and total-signal transduction proteins (CREB, JNK, NF- $\kappa\text{B}$ , ERK1/2, Akt, p70S6K, STAT3 and STAT5) and antioxidant proteins (SOD1, SOD2, CAT, TRX1, PRX2) were assessed using Milliplex magnetic bead array panels and a MagPix Multiplex imaging system. **Results:** In cells exposed to 1800 MHz continuous-wave RF-EMF with the presence of serum in the culture medium, CAT expression was statistically significantly decreased after a 30 min exposure, total JNK was decreased at both 30 and 240 min of exposure, STAT3 was decreased after 240 min of exposure and phosphorylated-CREB expression was decreased after 30 min of exposure. In cells exposed to 1800 MHz GSM-modulated RF-EMF in serum-free cultures, the expression level of total STAT5 was decreased after 30 and 240 min of exposure. These observed changes were detected sporadically across time-points, culture conditions and RF-EMF exposure conditions indicating the likelihood of false positive events. When cells were treated with anisomycin for 15 min as a positive control, dramatic increases in the expression of phosphorylated signalling proteins were observed in both serum-starved and serum-fed A172 cells, with larger fold change increases in the serum-free cultures. No statistically significant differences in the expression levels of SOD1, SOD2 or TRX1 were observed under any tested conditions after exposure to RF-EMF. **Conclusions:** The current study found no consistent evidence of changes in the expression of antioxidant proteins (SOD1, SOD2, CAT or TRX2) or a variety of signal transductions proteins (CREB, JNK, NF- $\kappa\text{B}$ , ERK1/2, Akt, p70S6K, STAT3, STAT5) in a human-derived glioblastoma A172 cell line in response to exposure to 1800 MHz continuous-wave or GSM-modulated RF-EMF for 5, 30 or 240 min in either serum-free or serum-containing cultures.

**(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 Electromagn Biol Med. 33(2):81-91, 2014.**

Abstract 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<sup>2</sup>) 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, Banerjee BD, Tripathi AK, Abegaonkar MP. Microwave radiation induced oxidative stress, cognitive impairment and inflammation in brain of Fischer rats. Indian J Exp Biol. 50(12):889-896, 2012. (LI)**

Public concerns over possible adverse effects of microwave radiation emitted by mobile phones on health are increasing. To evaluate the intensity of oxidative stress, cognitive impairment and inflammation in brain of Fischer rats exposed to microwave radiation, male Fischer-344 rats were exposed to 900 MHz microwave radiation (SAR = 5.953 x 10<sup>(-4)</sup> W/kg) and 1800 MHz microwave radiation (SAR = 5.835 x 10<sup>(-4)</sup> W/kg) for 30 days (2 h/day). Significant impairment in cognitive function and induction of oxidative stress in brain tissues of microwave exposed rats were observed in comparison with sham exposed groups. Further, significant increase in level of cytokines (IL-6 and TNF-alpha) was also observed following microwave exposure. Results of the present study indicated that increased oxidative stress due to microwave exposure may contribute to cognitive impairment and inflammation in brain.

**(E) Meral I, Mert H, Mert N, Deger Y, Yoruk I, Yetkin A, Keskin S. Effects of 900-MHz electromagnetic field emitted from cellular phone on brain oxidative stress and some vitamin levels of guinea pigs. Brain Res. 1169:120-124, 2007.**

This study was designed to demonstrate the effects of 900-MHz electromagnetic field (EMF) emitted from cellular phone on brain tissue and also blood malondialdehyde (MDA), glutathione (GSH), retinol (vitamin A), vitamin D(3) and tocopherol (vitamin E) levels, and catalase (CAT) enzyme activity of guinea pigs. Fourteen male guinea pigs, weighing 500-800 g were randomly divided into one of two experimental groups: control and treatment (EMF-exposed), each containing seven animals. Animals in treatment

group were exposed to 890- to 915-MHz EMF (217-Hz pulse rate, 2-W maximum peak power, SAR 0.95 w/kg) of a cellular phone for 12 h/day (11-h 45-min stand-by and 15-min spiking mode) for 30 days. Control guinea pigs were housed in a separate room without exposing EMF of a cellular phone. Blood samples were collected through a cardiac puncture and brains were removed after decapitation for the biochemical analysis at the end of the 30 days of experimental period. It was found that the MDA level increased ( $P<0.05$ ), GSH level and CAT enzyme activity decreased ( $P<0.05$ ), and vitamins A, E and D(3) levels did not change ( $P>0.05$ ) in the brain tissues of EMF-exposed guinea pigs. In addition, MDA, vitamins A, D(3) and E levels, and CAT enzyme activity increased ( $P<0.05$ ), and GSH level decreased ( $P<0.05$ ) in the blood of EMF-exposed guinea pigs. It was concluded that electromagnetic field emitted from cellular phone might produce oxidative stress in brain tissue of guinea pigs. However, more studies are needed to demonstrate whether these effects are harmful or/and affect the neural functions.

**(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. 2021 Sep;15(3):172-178. doi: 10.18502/jfrh.v15i3.7134.**

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  $0.045 \mu\text{w}/\text{cm}^2$  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) Mojez MR, Gaeini AA, Choobineh S, Sheykhloovand M. Hippocampal Oxidative Stress Induced by Radiofrequency Electromagnetic Radiation and the Neuroprotective Effects of Aerobic Exercise in Rats: A Randomized Control Trial. J Phys Act Health 2021 Oct 25;1-7. doi: 10.1123/jpah.2021-0213. Online ahead of print.**

**Background:** The present study determined whether 4 weeks of moderate aerobic exercise improves antioxidant capacity on the brain of rats against oxidative stress caused by radiofrequency electromagnetic radiation emitted from cell phones. **Methods:** Responses of malondialdehyde, catalase, glutathione peroxidase, and superoxide dismutase, as well as the number of hippocampal dead cells, were examined. Male Wistar rats (10-12 wk old) were

randomly assigned to 1 of 4 groups (N = 8): (1) moderate aerobic exercise (EXE) (2 × 15-30 min at 1215 m/min speed with 5 min of active recovery between sets), (2) exposure to 900/1800 MHz radiofrequency electromagnetic waves 3 hours per day (RAD), (3) EXE + RAD, and (4) exposure to an experimental phone without battery. **Results:** Following the exposure, the number of the hippocampal dead cells was significantly higher in group RAD compared with groups EXE, EXE + RAD, and control group. Malondialdehyde concentration in group RAD was significantly higher than that of groups EXE, EXE + RAD, and control group. Also, the activity of catalase, glutathione peroxidase, and superoxide dismutase in groups EXE, EXE + RAD, and control group was significantly higher compared with those of the exposure group. **Conclusion:** This study demonstrated that moderate aerobic exercise enhances hippocampal antioxidant capacity against oxidative challenge in the form of radiofrequency electromagnetic waves.

**(E) Monselise EB, Levkovitz A, Gottlieb HE, Kost D. Bioassay for assessing cell stress in the vicinity of radio-frequency irradiating antennas. J Environ Monit. 13(7):1890-1896, 2011.**

The 24 h exposure of water plants (etiolated duckweed) to RF-EMF between 7.8 V m<sup>-1</sup> and 1.8 V m<sup>-1</sup>, generated by AM 1.287 MHz transmitting antennas, resulted in alanine accumulation in the plant cells, a phenomenon we have previously shown to be a universal stress signal. The magnitude of the effect corresponds qualitatively to the level of RF-EMF exposure. In the presence of 10 mM vitamin C, alanine accumulation is completely suppressed, suggesting the involvement of free radicals in the process. A unique biological connection has thus been made between exposure to RF-EMF and cell stress, in the vicinity of RF transmitting antennas. This simple test, which lasts only 24 h, constitutes a useful bioassay for the quick detection of biological cell stress caused in the vicinity of RF irradiating antennas.

**(E) Morimoto S, Takahashi T, Shimizu K, Kanda T, Okaishi K, Okuro M, Murai H, Nishimura Y, Nomura K, Tsuchiya H, Ohashi I, Matsumoto M. Electromagnetic fields inhibit endothelin-1 production stimulated by thrombin in endothelial cells. J Int Med Res. 33(5):545-554, 2005.**

Electromagnetic field (EMF) radiation has been found to induce arteriolar dilatation, but the mechanism of action remains largely unknown. This study investigated the effect of EMF radiation on the production of endothelin-1 (ET-1), a potent vasoconstrictor, by cultured endothelial cells. EMF radiation reduced ET-1 basal levels in human umbilical vein and microvascular endothelial cells, but failed to reduce ET-1 basal levels in bovine and human aortic endothelial cells. EMF radiation significantly inhibited thrombin-stimulated ET-1 production in all four endothelial cell types in a dose-dependent manner. EMF radiation significantly inhibited thrombin-induced endothelin-1 mRNA expression in all four cell types. The inhibitory effect of EMF radiation on ET-1 production was abolished by the nitric oxide synthase inhibitor NG-monomethyl-L-arginine (10<sup>-3</sup> mol/l). These results demonstrate that EMF radiation modulates ET-1 production in cultured vascular endothelial cells and the inhibitory effect of EMF radiation is, at least partly, mediated through a nitric oxide-related pathway.

**(E) Mortazavi SMJ, Mostafavi-Pour Z, Daneshmand M, Zal F, Zare R, Mosleh-Shirazi MA. Adaptive Response Induced by Pre-Exposure to 915 MHz Radiofrequency: A Possible Role for Antioxidant Enzyme Activity. J Biomed Phys Eng. 7(2):137-142, 2017.**

**BACKGROUND:** Over the past few years, the rapid use of high frequency electromagnetic fields like **mobile phones** has raised global concerns about the negative health effects of its use. Adaptive response is the ability of a **cell** or tissue to better resist stress damage by prior exposure to a lesser amount of stress. This study aimed to assess whether radiofrequency radiation can induce adaptive response by changing the antioxidant balance. **MATERIALS AND METHODS:** In order to assess RF-induced adaptive response in tissues, we evaluated the level of GSH and the activity of GR in liver. 50 rats were divided into 5 groups. Three groups were pre-exposed to 915 MHz RF radiation, 4 hours per day for one week at different powers, as low, medium and high. 24 hours after the last exposure to radiation, they were exposed to 4 Gy sublethal dose of gamma radiation and then sacrificed after 5 hours. Their livers were removed, washed and were kept at -80o C until used. **RESULTS:** Our finding showed that pre-exposure to 915 MHz radiofrequency radiation with specific power could induce adaptive response in liver by inducing changes in the activity and level of antioxidant enzymes. **CONCLUSION:** It can be concluded that pre-exposure to microwave radiation could increase the level of GSH and the activity of GR enzyme, although these increases were seen just in low power group, and the GR activity was indicated in medium power group. This increase protects tissue from oxidative damage induced by sublethal dose of gamma radiation.

**(E) Motawi TK, Darwish HA, Moustafa YM, Labib MM. Biochemical Modifications and Neuronal Damage in Brain of Young and Adult Rats After Long-Term Exposure to Mobile Phone Radiations. Cell Biochem Biophys. 70:845-855, 2014.**

This study investigated the effect of exposure to mobile phone radiations on oxidative stress and apoptosis in brain of rats. Rats were allocated into six groups (three young and three adult). Groups 1 and 4 were not subjected to the radiation source and served as control groups. In groups 2 and 5, the mobile phones were only connected to the global system for mobile communication, while in groups 3 and 6, the option of calling was in use. Microwaves were generated by a mobile test phone (SAR = 1.13 W/kg) during 60 days (2 h/day). Significant increments in conjugated dienes, protein carbonyls, total oxidant status, and oxidative stress index along with a significant reduction of total antioxidant capacity levels were evident after exposure. Bax/Bcl-2 ratio, caspase-3 activity, and tumor necrosis factor-alpha level were enhanced, whereas no DNA fragmentation was detected. The relative brain weight of young rats was greatly affected, and histopathological examination reinforced the neuronal damage. The study highlights the detrimental effects of mobile phone radiations on brain during young and adult ages. The interaction of these radiations with brain is via dissipating its antioxidant status and/or triggering apoptotic cell death.

**(E) Moustafa YM, Moustafa RM, Belacy A, Abou-El-Ela SH, Ali FM. Effects of acute exposure to the radiofrequency fields of cellular phones on plasma lipid peroxide and antioxidant activities in human erythrocytes. J Pharm Biomed Anal 26(4):605-608, 2001.**

Radiofrequency fields of cellular phones may affect biological systems by increasing free radicals, which appear mainly to enhance lipid peroxidation, and by changing the antioxidase activities of human blood thus leading to oxidative stress. To test this, we have investigated the effect of acute exposure to radiofrequency fields of commercially available cellular phones on some parameters indicative of oxidative stress in 12 healthy adult male volunteers. Each volunteer put the phone in his pocket in standby position with the keypad facing the body. The parameters measured were lipid peroxide and the activities of superoxide dismutase (SOD), total glutathione peroxidase (GSH-Px) and catalase. The results obtained showed that the plasma level of lipid peroxide was significantly increased after 1, 2 and 4 h of exposure to radiofrequency fields of the cellular phone in standby position. Moreover, the activities of SOD and GSH-Px in human erythrocytes showed significant reduction while the activity of catalase in human erythrocytes did not decrease significantly. These results indicate that acute exposure to radiofrequency fields of commercially available cellular phones may modulate the oxidative stress of free radicals by enhancing lipid peroxidation and reducing the activation of SOD and GSH-Px, which are free radical scavengers. Therefore, these results support the interaction of radiofrequency fields of cellular phones with biological systems.

**(NE) Nakatani-Enomoto S, Okutsu M, Suzuki S, Suganuma R, Groiss SJ, Kadowaki S, Enomoto H, Fujimori K, Ugawa Y. Effects of 1950 MHz W-CDMA-like signal on human spermatozoa. Bioelectromagnetics. 2016 Jun 11. doi: 10.1002/bem.21985. [Epub ahead of print]**

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) Narayanan SN, Kumar RS, Kedage V, Nalini K, Nayak S, Bhat PG. Evaluation of oxidant stress and antioxidant defense in discrete brain regions of rats exposed to 900 MHz radiation. Bratisl Lek Listy. 115(5):260-266, 2014.**

AIM: In the current study, the effects of 900 MHz radio-frequency electromagnetic radiation (RF-EMR) on levels of thiobarbituric acid-reactive substances (TBARS), total antioxidants (TA), and glutathione S-transferase (GST) activity in discrete brain regions

were studied in adolescent rats. MATERIALS AND METHODS: Thirty-six male Wistar rats (6-8 weeks old) were allotted into three groups (n = 12 in each group). Control group (1) remained undisturbed in their home cage; sham group (2) was exposed to mobile phone in switch off mode for four weeks; RF-EMR-exposed group (3) was exposed to 900 MHz of RF-EMR (1 hr/day with peak power density of 146.60  $\mu\text{W}/\text{cm}^2$ ) from an activated Global System for Mobile communication (GSM) mobile phone (kept in silent mode; no ring tone and no vibration) for four weeks. On 29th day, behavioral analysis was done. Followed by this, six animals from each group were sacrificed and biochemical parameters were studied in amygdala, hippocampus, frontal cortex, and cerebellum. RESULTS: Altered behavioral performances were found in RF-EMR-exposed rats. Additionally, elevated TBARS level was found with all brain regions studied. RF-EMR exposure significantly decreased TA in the amygdala and cerebellum but its level was not significantly changed in other brain regions. GST activity was significantly decreased in the hippocampus but, its activity was unaltered in other brain regions studied. CONCLUSION: RF-EMR exposure for a month induced oxidative stress in rat brain, but its magnitude was different in different regions studied. RF-EMR-induced oxidative stress could be one of the underlying causes for the behavioral deficits seen in rats after RF-EMR exposure (Fig. 5, Ref. 37).

**(E) Narayanan SN, Lukose ST, Arun G, Mohapatra N, Pamala J, Concessao PL, Jetti R, Kedage V, Nalini K, Bhat PG. Modulatory effect of 900 MHz radiation on biochemical and reproductive parameters in rats. Bratisl Lek Listy. 119(9):581-587, 2018.**

In the present study, the effect of 900 MHz radiation exposure on blood biochemical and reproductive parameters was evaluated in adolescent rats. Male albino Wistar rats (8-10 weeks of age) were exposed to 900 MHz radiation (1hr/day, power density - 146.60  $\mu\text{W}/\text{cm}^2$ ) from a mobile phone for 28 days. On 29th day the animals were euthanized and malondialdehyde (MDA), total antioxidants (TA) levels and Glutathione-S transferase (GST) activity were studied in the blood. Reproductive parameters such as total sperm count, percentage of non-motile sperms, and sperm morphology were determined. Testes sections were stained with H(et)E staining and their cellular integrity was evaluated. Caspase-3 activity in the testes was also determined. MDA concentration was increased but TA levels and GST activity were not found to be different in 900 MHz group compared to controls. Sperm motility was found to be slightly reduced in 900 MHz group. Percentage of abnormal sperm was significantly elevated in 900 MHz group. Additionally, loss of germ cells particularly spermatocytes and spermatids was found in the testes of 900 MHz group. Testes caspase-3 activity was slightly elevated in 900 MHz exposed rats. Chronic 900 MHz exposure induced oxidative damage in the blood and lead to alterations in reproductive parameters in rats (Fig. 4, Ref. 33).

**(E) Naziroğlu M, Gümral N. Modulator effects of L-carnitine and selenium on wireless devices (2.45 GHz)-induced oxidative stress and electroencephalography records in brain of rat. Int J Radiat Biol. 85(8):680-689, 2009.**

**PURPOSE:** Electromagnetic radiation (EMR) from wireless devices may affect biological systems by increasing free radicals. The present study was designed to determine the effects of 2.45 GHz EMR on the brain antioxidant redox system and electroencephalography (EEG) records in rat. The possible protective effects of selenium and L-carnitine were also tested and compared to untreated controls. **MATERIALS AND METHODS:** Thirty rats were equally divided into five different groups, namely Group A(1): Cage control, Group A(2): Sham control, group B: 2.45 GHz EMR, group C: 2.45 GHz EMR + selenium, group D: 2.45 GHz EMR + L-carnitine. Groups B, C and D were exposed to 2.45 GHz EMR during 60 min/day for 28 days. End of the experiments, EEG records and the brain cortex samples were taken. **RESULTS:** The cortex brain vitamin A ( $p < 0.05$ ), vitamin C ( $p < 0.01$ ) and vitamin E ( $p < 0.05$ ) concentrations values were lower in group B than in group A1 and A2 although their concentrations were increased by selenium and L-carnitine supplementation. Lipid peroxidation, levels were lower in group C ( $p < 0.05$ ) and D ( $p < 0.01$ ) than in group B where as reduced glutathione levels were higher in group C ( $p < 0.05$ ) than in group A1, A2 and B. However, B-carotene levels did not change in the five groups. **CONCLUSIONS:** L-carnitine and selenium seem to have protective effects on the 2.45 GHz-induced decrease of the vitamins by supporting antioxidant redox system. L-carnitine on the vitamin concentrations seems to more protective affect than in selenium.

**(cancer) (E) \*Nazıroğlu M, Ciğ B, Doğan S, Uğuz AC, Dilek S, Faouzi D. 2.45-Gz wireless devices induce oxidative stress and proliferation through cytosolic Ca<sup>2+</sup> influx in human leukemia cancer cells. Int J Radiat Biol. 88(6):449-456, 2012a.**

**PURPOSE:** Electromagnetic radiation from wireless devices may affect biological systems by increasing free radicals. The present study was designed to determine the effects of 2.45 GHz radiation on the antioxidant redox system, calcium ion signaling, cell count and viability in human leukemia 60 cells. **MATERIALS AND METHODS:** Twelve cell cultures were equally divided into two main groups as controls ( $n = 6$ ) and irradiated ( $n = 6$ ) and then subdivided into four different subgroups depending on the duration of exposure, namely 1, 2, 12 and 24 hours. The samples were analyzed immediately after the experimental period. **RESULTS:** The extent of lipid peroxidation, cytosolic free Ca<sup>2+</sup> and cell numbers were higher in 2.45 GHz groups than in the controls. The increase of cytosolic free Ca<sup>2+</sup> concentrations was radiation time-dependent and was highest at 24-h exposure. The reduced glutathione, glutathione peroxidase, vitamin C and cell viability values did not show any changes in any of the experimental groups. 2-aminoethyl diphenylborinate inhibits Ca<sup>2+</sup> ions influx by blockage of the transient receptor potential melastatin 2. **CONCLUSIONS:** 2.45 GHz electromagnetic radiation appears to induce proliferative effects through oxidative stress and Ca<sup>2+</sup> influx although blocking of transient receptor potential melastatin 2 channels by 2-aminoethyl diphenylborinate seems to counteract the effects on Ca<sup>2+</sup> ions influx.

**(E) \*Nazıroğlu M, Çelik Ö, Özgül C, Çiğ B, Doğan S, Bal R, Gümral N, Rodríguez AB, Pariente JA. Melatonin modulates wireless (2.45 GHz)-induced oxidative injury through TRPM2 and voltage gated Ca(2+) channels in brain and dorsal root ganglion in rat. Physiol Behav. 105(3):683-692, 2012b.**

We aimed to investigate the protective effects of melatonin and 2.45 GHz electromagnetic radiation (EMR) on brain and dorsal root ganglion (DRG) neuron antioxidant redox system, Ca(2+) influx, cell viability and electroencephalography (EEG) records in the rat. Thirty two rats were equally divided into four different groups namely group A1: Cage control, group A2: Sham control, group B: 2.45 GHz EMR, group C: 2.45 GHz EMR+melatonin. Groups B and C were exposed to 2.45 GHz EMR during 60 min/day for 30 days. End of the experiments, EEG records and the brain cortex and DRG samples were taken. Lipid peroxidation (LP), cell viability and cytosolic Ca(2+) values in DRG neurons were higher in group B than in groups A1 and A2 although their concentrations were increased by melatonin, 2-aminoethyldiphenyl borinate (2-APB), diltiazem and verapamil supplementation. Spike numbers of EEG records in group C were lower than in group B. Brain cortex vitamin E concentration was higher in group C than in group B. In conclusion, Melatonin supplementation in DRG neurons and brain seems to have protective effects on the 2.45 GHz-induced increase Ca(2+) influx, EEG records and cell viability of the hormone through TRPM2 and voltage gated Ca(2+) channels.

**(NE) Nazırođlu M, Özkan FF, Hapil SR, Ghazizadeh V, Çiđ B. Epilepsy but not mobile phone frequency (900 MHz) induces apoptosis and calcium entry in hippocampus of epileptic rat: involvement of TRPV1 channels. J Membr Biol. 248(1):83-91, 2015.**

Electromagnetic radiation (EMR) and epilepsy are reported to mediate the regulation of apoptosis and oxidative stress through Ca(2+) influx. Results of recent reports indicated that EMR can increase temperature and oxidative stress of body cells, and TRPV1 channel is activated by noxious heat, oxidative stress, and capsaicin (CAP). We investigated the effects of mobile phone (900 MHz) EMR exposure on Ca(2+) influx, apoptosis, oxidative stress, and TRPV1 channel activations in the hippocampus of pentylenetetrazol (PTZ)-induced epileptic rats. Freshly isolated hippocampal neurons of twenty-one rats were used in study within three groups namely control, PTZ, and PTZ + EMR. The neurons in the three groups were stimulated by CAP. Epilepsy was induced by PTZ administration. The neurons in PTZ + EMR group were exposed to the 900 MHz EMR for 1 h. The apoptosis, mitochondrial membrane depolarization, intracellular reactive oxygen species (ROS), and caspase-3 and caspase-9 values were higher in PTZ and PTZ + EMR groups than in control. However, EMR did not add additional increase effects on the values in the hippocampal neurons. Intracellular-free Ca(2+) concentrations in fura-2 analyses were also higher in PTZ + CAP group than in control although their concentrations were decreased by TRPV1 channel blocker, capsazepine. However, there were no statistical changes on the Ca(2+) concentrations between epilepsy and EMR groups. In conclusion, apoptosis, mitochondrial, ROS, and Ca(2+) influx via TRPV1 channel were increased in the hippocampal neurons by epilepsy induction although the mobile phone did not change the values. The results indicated that TRPV1 channels in hippocampus may possibly be a novel target for effective target of epilepsy.

**(cancer) (Review) Nazırođlu M, Tokat S, Demirci S. Role of melatonin on electromagnetic radiation-induced oxidative stress and Ca2+ signaling molecular pathways in breast cancer. J Recept Signal Transduct Res. 32(6):290-297, 2012c.**

AIMS: Exposure to electromagnetic radiation (EMR) may increase breast cancer risk by inducing oxidative stress and suppressing the production of melatonin. Aim of the present review is to discuss the mechanisms and risk factors of EMR and oxidative stress-induced breast cancer, to summarize the controlled studies evaluating measures for prevention, and to conclude with evidence-based strategies for prevention. MATERIALS: Review of the relevant literature and results from our recent basic studies, as well as critical analyses of published systematic reviews were obtained from the Pubmed and the Science Citation Index. RESULTS: It has been proposed that chronic exposure to EMR may increase the risk of breast cancer by suppressing the production of melatonin; this suppression may affect the development of breast cancer either by increasing levels of circulation of estrogen or through over production of free oxygen radicals. Most epidemiological studies have also indicated overall effect of EMR exposure in premenopausal women, particularly for estrogen receptor positive breast tumors. Enhanced voltage-dependent Ca(2+) current and impaired inhibitory G-protein function, and derangement of intracellular organelles with a Ca(2+) buffering effect, such as endoplasmic reticulum and mitochondria have been also shown to contribute to disturbed Ca(2+) signaling in breast cancer. CONCLUSION: Melatonin may modulate breast cancer through modulation of enhanced oxidative stress and Ca(2+) influx in cell lines. However, there is not enough evidence on increased risk of breast cancer related to EMR exposure.

**(Review) Nazıroğlu M, Yüksel M, Köse SA, Özkaya MO. Recent reports of Wi-Fi and mobile phone-induced radiation on oxidative stress and reproductive signaling pathways in females and males. J Membr Biol. 246(12):869-875, 2013.**

Environmental exposure to electromagnetic radiation (EMR) has been increasing with the increasing demand for communication devices. The aim of the study was to discuss the mechanisms and risk factors of EMR changes on reproductive functions and membrane oxidative biology in females and males. It was reported that even chronic exposure to EMR did not increase the risk of reproductive functions such as increased levels of neoantigens abort. However, the results of some studies indicate that EMR induced endometriosis and inflammation and decreased the number of follicles in the ovarium or uterus of rats. In studies with male rats, exposure caused degeneration in the seminiferous tubules, reduction in the number of Leydig cells and testosterone production as well as increases in luteinizing hormone levels and apoptotic cells. In some cases of male and female infertility, increased levels of oxidative stress and lipid peroxidation and decreased values of antioxidants such as melatonin, vitamin E and glutathione peroxidase were reported in animals exposed to EMR. In conclusion, the results of current studies indicate that oxidative stress from exposure to Wi-Fi and mobile phone-induced EMR is a significant mechanism affecting female and male reproductive systems. However, there is no evidence to this date to support an increased risk of female and male infertility related to EMR exposure.

**(E) Nirwane A, Sridhar V, Majumdar A. Neurobehavioural Changes and Brain Oxidative Stress Induced by Acute Exposure to GSM900 Mobile Phone Radiations in Zebrafish (Danio rerio). Toxicol Res. 2016 Apr;32(2):123-32. doi: 10.5487/TR.2016.32.2.123. Epub 2016 Apr 30.**

The impact of mobile phone (MP) radiation on the brain is of specific interest to the scientific community and warrants investigations, as MP is held close to the head. Studies on humans and rodents revealed hazards MP radiation associated such as brain tumors, impairment in cognition, hearing etc. Melatonin (MT) is an important modulator of CNS functioning and is a neural antioxidant hormone. Zebrafish has emerged as a popular model organism for CNS studies. Herein, we evaluated the impact of GSM900MP (GSM900MP) radiation exposure daily for 1 hr for 14 days with the SAR of 1.34W/Kg on neurobehavioral and oxidative stress parameters in zebrafish. Our study revealed that, GSM900MP radiation exposure, significantly decreased time spent near social stimulus zone and increased total distance travelled, in social interaction test. In the novel tank dive test, the GSM900MP radiation exposure elicited anxiety as revealed by significantly increased time spent in bottom half; freezing bouts and duration and decreased distance travelled, average velocity, and number of entries to upper half of the tank. Exposed zebrafish spent less time in the novel arm of the Y-Maze, corroborating significant impairment in learning as compared to the control group. Exposure decreased superoxide dismutase (SOD), catalase (CAT) activities whereas, increased levels of reduced glutathione (GSH) and lipid peroxidation (LPO) was encountered showing compromised antioxidant defense. Treatment with MT significantly reversed the above neurobehavioral and oxidative derangements induced by GSM900MP radiation exposure. This study traced GSM900MP radiation exposure induced neurobehavioral aberrations and alterations in brain oxidative status. Furthermore, MT proved to be a promising therapeutic candidate in ameliorating such outcomes in zebrafish.

**(E) Odacı E, Unal D, Mercantepe T, Topal Z, Hancı H, Türedi S, Erol H, Mungan S, Kaya H, Colakoğlu S. Pathological effects of prenatal exposure to a 900 MHz electromagnetic field on the 21-day-old male rat kidney. Biotech Histochem. 2014 Aug 27:1-9. [Epub ahead of print]**

We investigated the effects on kidney tissue of 900 megahertz (MHz) EMF applied during the prenatal period. Pregnant rats were exposed to 900 MHz EMF, 1 h/day, on days 13-21 of pregnancy; no procedure was performed on control group pregnant rats or on mothers or newborns after birth. On postnatal day 21, kidney tissues of male rat pups from both groups were examined by light and electron microscopy. Malondialdehyde (MDA), superoxide dismutase (SOD), catalase (CAT) and glutathione levels also were investigated. Light microscopy revealed some degenerative changes in the tubule epithelium, small cystic formations in the primitive tubules and large cysts in the cortico-medullary or medullary regions in the experimental group. Electron microscopy revealed a loss of peritubular capillaries and atypical parietal layer epithelial cells in the experimental group. Biochemical analysis showed significantly increased MDA levels in the experimental group and decreased SOD and CAT levels. EMF applied during the prenatal period can caused pathological changes in kidney tissue in 21-day-old male rats owing to oxidative stress and decreased antioxidant enzyme levels.

**(E) Odacı E, Özyılmaz C. Exposure to a 900 MHz electromagnetic field for one hour a day over 30 days does change the histopathology and biochemistry of the rat testis. Int J Radiat Biol. 2015 Mar 19:1-20. [Epub ahead of print]**

**PURPOSE:** This study investigated the effect of exposure to a 900-megahertz (MHz) electromagnetic field (EMF) on the rat testicle. **MATERIALS AND METHODS:** Twenty-four adult male rats were divided into control, sham and EMF groups. The EMF group rats were exposed to 900-MHz EMF (1 h / 30 day), and testicles were extracted at the end of the experiment. Malondialdehyde, superoxide dismutase, catalase and glutathione levels and apoptotic index and histopathological damage scores were compared. **RESULTS:** Histopathologically, EMF group rats exhibited vacuoles in seminiferous tubules basal membrane and edema in the intertubular space. Seminiferous tubule diameters and germinal epithelium thickness were both smaller, and apoptotic index was higher, in the EMF group than in the other groups. Malondialdehyde, superoxide dismutase, catalase and glutathione values in the EMF group decreased significantly compared to those of the control group. **CONCLUSIONS:** The results show that exposure to 900-MHz EMF causes alterations in adult rat testicular morphology and biochemistry.

**(E) Okatan DÖ, Kulaber A, Kerimoglu G, Odacı E. Altered morphology and biochemistry of the female rat liver following 900 megahertz electromagnetic field exposure during mid to late adolescence. Biotech Histochem. 2019 Aug;94(6):420-428.**

Despite their benefits, technological devices such as cell phones may also have deleterious effects on human health. Considerable debate continues concerning the effects of the electromagnetic field (EMF) emitted during cell phone use on human health. We investigated the effects of exposure to 900 megahertz (MHz) EMF during mid to late adolescence on the rat liver. Control (ContGr), sham (ShmGr) and EMF (EMFGr) groups of female rats were established. We exposed the EMFGr rats daily to 900 MHz EMF on postnatal days 35-59. ShmGr rats underwent sham procedures. No procedure was performed on ContGr rats. Rats were sacrificed on postnatal day 60 and the livers were extracted. One part of the liver was stained with Masson's trichrome or hematoxylin and eosin. The remaining tissue was used to measure oxidative stress markers including malondialdehyde, glutathione, catalase, superoxide dismutase, 8-hydroxydeoxyguanosine (8-OHdG) and nitrotyrosine. Total antioxidant status and total oxidant status were used to calculate the oxidative stress index. We found normal hepatic morphology in the ContGr and ShmGr groups. The EMFGr group exhibited occasional irregularities in the radial arrangement of hepatocytes, cytoplasmic vacuolization, hemorrhage, sinusoid expansion, hepatocyte morphology and edema. Biochemical analysis revealed that 8-OHdG and SOD levels in EMFGr decreased significantly compared to the ContGr and ShmGr groups. Exposure to a continuous 900 MHz EMF for 1 h daily during mid to late adolescence may cause histopathological and biochemical alterations in hepatic tissue.

**(E) \*Oksay T, Naziroğlu M, Doğan S, Güzel A, Gümral N, Koşar PA. Protective effects of melatonin against oxidative injury in rat testis induced by wireless (2.45 GHz) devices. Andrologia. 46:65-72, 2014.**

Wireless devices have become part of everyday life and mostly located near reproductive organs while they are in use. The present study was designed to determine the possible protective effects of melatonin on oxidative stress-dependent testis injury induced by 2.45-GHz electromagnetic radiation (EMR). Thirty-two rats were equally divided into four different groups, namely cage control (A1), sham control (A2), 2.45-GHz EMR (B)

and 2.45-GHz EMR+melatonin (C). Group B and C were exposed to 2.45-GHz EMR during 60 min day(-1) for 30 days. Lipid peroxidation levels were higher in Group B than in Group A1 and A2. Melatonin treatment prevented the increase in the lipid peroxidation induced by EMR. Also reduced glutathione (GSH) and glutathione peroxidase (GSH-Px) levels in Group D were higher than that of exposure group. Vitamin A and E concentrations decreased in exposure group, and melatonin prevented the decrease in vitamin E levels. In conclusion, wireless (2.45 GHz) EMR caused oxidative damage in testis by increasing the levels of lipid peroxidation and decreasing in vitamin A and E levels. Melatonin supplementation prevented oxidative damage induced by EMR and also supported the antioxidant redox system in the testis.

**(E) Oktem F, Ozguner F, Mollaoglu H, Koyu A, Uz E. Oxidative Damage in the Kidney Induced by 900-MHz-Emitted Mobile Phone: Protection by Melatonin. Arch Med Res. 36(4):350-355, 2005.**

BACKGROUND: The mobile phones emitting 900-MHz electromagnetic radiation (EMR) may be mainly absorbed by kidneys because they are often carried in belts. Melatonin, the chief secretory product of the pineal gland, was recently found to be a potent free radical scavenger and antioxidant. The aim of this study was to examine 900-MHz mobile phone-induced oxidative stress that promotes production of reactive oxygen species (ROS) on renal tubular damage and the role of melatonin on kidney tissue against possible oxidative damage in rats. METHODS: The animals were randomly grouped as follows: 1) sham-operated control group and 2) study groups: i) 900-MHz EMR exposed (30 min/day for 10 days) group and ii) 900-MHz EMR exposed+melatonin (100 mug kg(-1) s.c. before the daily EMR exposure) treated group. Malondialdehyde (MDA), an index of lipid peroxidation), and urine N-acetyl-beta-d-glucosaminidase (NAG), a marker of renal tubular damage were used as markers of oxidative stress-induced renal impairment. Superoxide dismutase (SOD), catalase (CAT), and glutathione peroxidase (GSH-Px) activities were studied to evaluate the changes of antioxidant status. RESULTS: In the EMR-exposed group, while tissue MDA and urine NAG levels increased, SOD, CAT, and GSH-Px activities were reduced. Melatonin treatment reversed these effects as well. In this study, the increase in MDA levels of renal tissue and in urine NAG and also the decrease in renal SOD, CAT, GSH-Px activities demonstrated the role of oxidative mechanism induced by 900-MHz mobile phone exposure, and melatonin, via its free radical scavenging and antioxidant properties, ameliorated oxidative tissue injury in rat kidney. CONCLUSIONS: These results show that melatonin may exhibit a protective effect on mobile phone-induced renal impairment in rats.

**(E) Olgar Y, Hidisoglu E, Celen MC, Yamasan BE, Yargicoglu P, Ozdemir S. 2.1 GHz electromagnetic field does not change contractility and intracellular Ca<sup>2+</sup> transients but decreases  $\beta$ -adrenergic responsiveness through nitric oxide signaling in rat ventricular myocytes. Int J Radiat Biol. 2015 Jul 1:1-23. [Epub ahead of print]**

PURPOSE: Due to the increasing use of wireless technology in developing countries, particularly mobile phones, the influence of electromagnetic fields (EMF) on biologic systems has become the subject of an intense debate. Therefore, in this study we investigated the effect of 2.1 GHz EMF on contractility and beta-adrenergic ( $\beta$ -AR) responsiveness of ventricular myocytes. MATERIALS AND METHODS: Rats were randomized to the following groups:

sham rats (SHAM) and rats exposed to 2.1 GHz EMF for 2 hours/day for 10 weeks (EM-10). Sarcomere shortening and  $\text{Ca}^{2+}$  transients were recorded in isolated myocytes loaded with Fura2-AM and electrically stimulated at 1 Hz, while L-type  $\text{Ca}^{2+}$  currents ( $I_{\text{CaL}}$ ) were measured using whole-cell patch clamping at  $36\pm 1^\circ\text{C}$ . Cardiac nitric oxide (NO) levels were measured in tissue samples using a colorimetric assay kit. RESULTS: Fractional shortening and amplitude of the matched  $\text{Ca}^{2+}$  transients were not changed in EM-10 rats. Although the isoproterenol-induced ( $10^{-6}$  M)  $I_{\text{CaL}}$  response was reduced in rats exposed to EMF, basal  $I_{\text{CaL}}$  density in myocytes was similar between the two groups ( $p < 0.01$ ). Moreover, EMF exposure led to a significant increase in nitric oxide levels in rat heart ( $p < 0.02$ ). CONCLUSIONS: Long-term exposure to 2.1 GHz EMF decreases  $\beta$ -AR responsiveness of ventricular myocytes through NO signaling.

**(E) Oral B, Guney M, Ozguner F, Karahan N, Mungan T, Comlekci S, Cesur G. Endometrial Apoptosis Induced by a 900-MHz Mobile Phone: Preventive Effects of Vitamins E and C. Adv Ther. 23(6):957-973, 2006.**

Numerous reports have described the effects induced by an electromagnetic field (EMF) in various cellular systems. The purposes of this study were to examine oxidative stress that promotes production of reactive oxygen species induced by a 900-megahertz (MHz) mobile phone and the possible ameliorating effects of vitamins E and C on endometrial tissue against EMF-induced endometrial impairment and apoptosis in rats. Animals were randomly grouped as follows: (1) sham-operated control group ( $n=8$ ), (2) 900 MHz EMF-exposed group ( $n=8$ ; 30 min/d for 30 d), and (3) 900 MHz EMF-exposed group, treated with vitamins E and C ( $n=8$ ; 50 mg/kg intramuscularly and 20 mg/kg body weight intraperitoneally before daily EMF exposure). Malondialdehyde (an index of lipid peroxidation) was used as a marker of oxidative stress-induced endometrial impairment; Bcl-2, Bax, caspase-3, and caspase-8 were assessed immunohistochemically. In this study, increased malondialdehyde levels in endometrial tissue and apoptosis illustrated the role of the oxidative mechanism induced by exposure to a 900-MHz mobile phone-like device and vitamins E and C; via free radical scavenging and antioxidant properties, oxidative tissue injury and apoptosis were ameliorated in rat endometrium. In conclusion, exposure to 900-MHz radiation emitted by mobile phones may cause endometrial apoptosis and oxidative stress, but treatment with vitamins E and C can diminish these changes and may have a beneficial effect in preventing endometrial changes in rats.

**(cancer) (E) Osera C, Amadio M, Falone S, Fassina L, Magenes G, Amicarelli F, Ricevuti G, Govoni S, Pascale A. Pre-exposure of neuroblastoma cell line to pulsed electromagnetic field prevents H<sub>2</sub>O<sub>2</sub>-induced ROS production by increasing mnSOD activity. Bioelectromagnetics. 2015 Feb 23. doi: 10.1002/bem.21900. [Epub ahead of print]**

Electromagnetic fields (EMFs) have been linked to increased risk of cancers and neurodegenerative diseases; however, EMFs can also elicit positive effects on biological systems, and redox status seems crucially involved in EMF biological effects. This study aimed to assess whether a short and repeated pulsed EMF (PEMF) could trigger adaptive responses against an oxidative insult in a neuronal cellular model. We found that a 40 min

overall (four times a week, 10 min each) pre-exposure to PEMF did not affect major physiological parameters and led to a significant increase of Mn-dependent superoxide dismutase activity in the human neuroblastoma SH-SY5Y cell line. In addition, we found PEMF-pre-exposed cells exhibited decreased reactive oxygen species production following a 30 min H<sub>2</sub>O<sub>2</sub> challenge, with respect to non pre-exposed cells. Our findings might provide new insights on the role played by short and repeated PEMF stimulations in the enhancement of cellular defenses against oxidative insults. Although studies in normal neuronal cells would be useful to further confirm our hypothesis, we suggest that specific PEMF treatments may have potential biological repercussions in diseases where oxidative stress is implicated.

**(E) Othman H, Ammari M, Sakly M, Abdelmelek H. Effects of prenatal exposure to WIFI signal (2.45GHz) on postnatal development and behavior in rat: Influence of maternal restraint. Behav Brain Res. 326:291-302, 2017.**

The present study was carried out to investigate the potential combined influence of maternal restraint stress and 2.45GHz WiFi signal exposure on postnatal development and behavior in the offspring of exposed rats. 24 pregnant albino Wistar rats were randomly assigned to four groups: Control, WiFi-exposed, restrained and both WiFi-exposed and restrained groups. Each of WiFi exposure and restraint occurred 2h/day along gestation till parturition. The pups were evaluated for physical development and neuromotor maturation. Moreover, elevated plus maze test, open field activity and stationary beam test were also determined on postnatal days 28, 30 and 31, respectively. After behavioral tests, the rats were anesthetized and their brains were removed for biochemical analysis. Our main findings showed no detrimental effects on gestation progress and outcomes at delivery in all groups. Subsequently, WiFi and restraint, per se and mainly in concert altered physical development of pups with slight differences between genders. Behaviorally, the gestational WiFi irradiation, restraint and especially the associated treatment affected the neuromotor maturation mainly in male progeny. At adult age, we noticed anxiety, motor deficit and exploratory behavior impairment in male offspring co-exposed to WiFi radiation and restraint, and in female progeny subjected to three treatments. The biochemical investigation showed that, all three treatments produced global oxidative stress in brain of both sexes. As for serum biochemistry, phosphorus, magnesium, glucose, triglycerides and calcium levels were disrupted. Taken together, prenatal WiFi radiation and restraint, alone and combined, provoked several behavioral and biochemical impairments at both juvenile and adult age of the offspring.

**(E) Othman H, Ammari M, Rtibi K, Bensaid N, Sakly M, Abdelmelek H. Postnatal development and behavior effects of in-utero exposure of rats to radiofrequency waves emitted from conventional WiFi devices. Environ Toxicol Pharmacol. 52:239-247, 2017.**

The present work investigated the effects of prenatal exposure to radiofrequency waves of conventional WiFi devices on postnatal development and behavior of rat offspring. Ten Wistar albino pregnant rats were randomly assigned to two groups (n=5). The experimental group was exposed to a 2.45GHz WiFi signal for 2h a day throughout gestation period. Control females were subjected to the same conditions as treated group without applying WiFi radiations. After delivery, the offspring was

tested for physical and neurodevelopment during its 17 postnatal days (PND), then for anxiety (PND 28) and motricity (PND 40-43), as well as for cerebral oxidative stress response and cholinesterase activity in brain and serum (PND 28 and 43). Our main results showed that the in-utero WiFi exposure impaired offspring neurodevelopment during the first seventeen postnatal days without altering emotional and motor behavior at adult age. Besides, prenatal WiFi exposure induced cerebral oxidative stress imbalance (increase in malondialdehyde level (MDA) and hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>) levels and decrease in catalase (CAT) and superoxide dismutase (SOD) activities) at 28 but not 43days old, also the exposure affected acetylcholinesterase activity at both cerebral and seric levels. Thus, the current study revealed that maternal exposure to WiFi radiofrequencies led to various adverse neurological effects in the offspring by affecting neurodevelopment, cerebral stress equilibrium and cholinesterase activity.

**(E) Othman H, Ammari M , Sakly M, Abdelmelek H. Effects of repeated restraint stress and WiFi signal exposure on behavior and oxidative stress in rats. *Metab Brain Dis.* 32(5):1459-1469, 2017.**

Today, due to technology development and aversive events of daily life, Human exposure to both radiofrequency and stress is unavoidable. This study investigated the co-exposure to repeated restraint stress and WiFi signal on cognitive function and oxidative stress in brain of male rats. Animals were divided into four groups: Control, WiFi-exposed, restrained and both WiFi-exposed and restrained groups. Each of WiFi exposure and restraint stress occurred 2 h (h)/day during 20 days. Subsequently, various tests were carried out for each group, such as anxiety in elevated plus maze, spatial learning abilities in the water maze, cerebral oxidative stress response and cholinesterase activity in brain and serum. Results showed that WiFi exposure and restraint stress, alone and especially if combined, induced an anxiety-like behavior without impairing spatial learning and memory abilities in rats. At cerebral level, we found an oxidative stress response triggered by WiFi and restraint, per se and especially when combined as well as WiFi-induced increase in acetylcholinesterase activity. Our results reveal that there is an impact of WiFi signal and restraint stress on the brain and cognitive processes especially in elevated plus maze task. In contrast, there are no synergistic effects between WiFi signal and restraint stress on the brain.

**(E) Oyewopo AO, Olaniyi SK, Oyewopo CI, Jimoh AT. Radiofrequency electromagnetic radiation from cell phone causes defective testicular function in male Wistar rats. *Andrologia.* 2017 Dec;49(10). doi: 10.1111/and.12772. Epub 2017 Mar 6**

Cell phones have become an integral part of everyday life. As cell phone usage has become more widespread, concerns have increased regarding the harmful effects of radiofrequency electromagnetic radiation from these devices. The current study was undertaken to investigate the effects of the emitted radiation by cell phones on testicular histomorphometry and biochemical analyses. Adult male Wistar rats weighing 180-200 g were randomly allotted to control, group A (switched off mode exposure), group B (1-hr exposure), group C (2-hr exposure) and group D (3-hr exposure). The animals were

exposed to radiofrequency electromagnetic radiation of cell phone for a period of 28 days. Histomorphometry, biochemical and histological investigations were carried out. The histomorphometric parameters showed no significant change ( $p < .05$ ) in the levels of germinal epithelial diameter in all the experimental groups compared with the control group. There was no significant change ( $p < .05$ ) in cross-sectional diameter of all the experimental groups compared with the control group. Group D rats showed a significant decrease ( $p < .05$ ) in lumen diameter compared with group B rats. There was an uneven distribution of germinal epithelial cells in groups B, C and D. However, there was degeneration of the epithelia cells in group D when compared to the control and group B rats. Sera levels of malondialdehyde (MDA) and superoxide dismutase (SOD), which are markers of reactive oxygen species, significantly increased (MDA) and decreased (SOD), respectively, in all the experimental groups compared with the control group. Also sera levels of gonadotropic hormones (FSH, LH and testosterone) significantly decreased ( $p < .05$ ) in groups C and D compared with the control group. The study demonstrates that chronic exposure to radiofrequency electromagnetic radiation of cell phone leads to defective testicular function that is associated with increased oxidative stress and decreased gonadotropic hormonal profile.

**(E) Özdemir E, Çömelekoğlu U, Degirmenci E, Bayrak G, Yildirim M, Ergenoglu t T, Yilmaz BC, Yalin S, Koyuncu DD, Ozbay E. The effect of 4.5 G (LTE Advanced-Pro network) mobile phone radiation on the optic nerve. Cutan Ocul Toxicol 2021 Sep;40(3):198-206.**

**Purpose:** Rapid development in mobile phone technologies increase the average mobile phone usage duration. This increase also triggers exposure to radiofrequency radiation (RF), which is a risk factor for the health. In this study, it was aimed to investigate the effect of mobile phone working with LTE-Advanced Pro (4.5 G) mobile network on the optic nerve, which is responsible for the transmission of visual information. **Material and methods:** Thirty-two rats divided into two groups as control (no RF, sham exposure) and experimental (RF exposure using a mobile phone with LTE-Advanced Pro network; 2 hours/day, 6 weeks). The visual evoked potential (VEP) was recorded and determined amplitudes and latencies of VEP waves. Optic nerve malondialdehyde level, catalase and superoxide dismutase activities were determined. Furthermore, ultrastructural and morphometric changes of optic nerve were evaluated **Results:** In VEP recordings, the mean VEP amplitudes of experimental group were significantly lower than control group. In ultrastructural evaluation, myelinated nerve fibres and glial cells were observed in normal histologic appearance both in sham and experimental group. However, by performing morphometric analysis, in the experimental group, axonal diameter and myelin thickness were shown to be lower and the G-ratio was higher than in the sham group. In the experimental group, malondialdehyde level was significantly higher and superoxide dismutase and catalase activities were significantly lower than sham group. There was a high correlation between VEP wave amplitudes and oxidative stress markers. **Conclusion:** Findings obtained in this study support optic nerve damage. These results point out an important risk that may decrease the quality of life.

**(E) Ozguner F, Aydin G, Mollaoglu H, Gokalp O, Koyu A, Cesur G. Prevention of mobile phone induced skin tissue changes by melatonin in rat: an experimental study. Toxicol Ind Health. 20(6-10):133-139, 2004.**

Most of the mobile phones in Turkey emit 900 MHz radiation which is mainly absorbed by the skin and, to a lesser extent, muscle. The aim of this study was to investigate the effects the 900 MHz electromagnetic irradiation emitted by these devices on the induction of histopathologic changes in skin and the effect of melatonin (Mel) on any of these changes. Thirty male Wistar-Albino rats were used in the study. The experimental groups were composed of: a nontreated control group, an irradiated group (IR) without Mel and an irradiated with Mel treatment group (IR + Mel). 900 MHz radiation was applied to IR group for 10 days (30 min/day). The IR + Mel group received 10 mg/kg per day melatonin in tap water for 10 days before irradiation. At the end of the tenth day, the skin graft was excized from the thoraco-abdominal area. Histopathologic changes in skin were analyzed. In the IR group, increased thickness of stratum corneum, atrophy of epidermis, papillomatosis, basal cell proliferation, increased granular cell layer (hypergranulosis) in epidermis and capillary proliferation, impairment in collagen tissue distribution and separation of collagen bundles in dermis were all observed compared to the control group. Most of these changes, except hypergranulosis, were prevented with melatonin treatment. In conclusion, exposure to 900 MHz radiation emitted by mobile phones caused mild skin changes. Furthermore, melatonin treatment can reduce these changes and may have a beneficial effect to prevent 900 MHz mobile phone-induced rat skin changes.

**(E) Ozguner F, Oktem F, Ayata A, Koyu A, Yilmaz HR. A novel antioxidant agent caffeic acid phenethyl ester prevents long-term mobile phone exposure-induced renal impairment in rat. Prognostic value of malondialdehyde, N-acetyl-beta-D-glucosaminidase and nitric oxide determination. Mol Cell Biochem. 277(1-2):73-80, 2005.**

Caffeic acid phenethyl ester (CAPE), a flavonoid like compound, is one of the major components of honeybee propolis. It has been used in folk medicine for many years in Middle East countries. It was found to be a potent free radical scavenger and antioxidant recently. The aim of this study was to examine long-term applied 900 MHz emitting mobile phone-induced oxidative stress that promotes production of reactive oxygen species (ROS) and, was to investigate the role of CAPE on kidney tissue against the possible electromagnetic radiation (EMR)-induced renal impairment in rats. In particular, the ROS such as superoxide and nitric oxide (NO) may contribute to the pathophysiology of EMR-induced renal impairment. Malondialdehyde (MDA, an index of lipid peroxidation) levels, urinary N-acetyl-beta-D-glucosaminidase (NAG, a marker of renal tubular injury) and nitric oxide (NO, an oxidant product) levels were used as markers of oxidative stress-induced renal impairment and the success of CAPE treatment. The activities of superoxide dismutase (SOD), catalase (CAT), and glutathione peroxidase (GSH-Px) in renal tissue were determined to evaluate the changes of antioxidant status. The rats used in the study were randomly grouped (10 each) as follows: i) Control group (without stress and EMR), ii) Sham-operated rats stayed without exposure to EMR (exposure device off), iii) Rats exposed to 900 MHz EMR (EMR group), and iv) A 900 MHz EMR exposed + CAPE treated group (EMR + CAPE group). In the EMR exposed

group, while tissue MDA, NO levels and urinary NAG levels increased ( $p < 0.0001$ ), the activities of SOD, CAT, and GSH-Px in renal tissue were reduced ( $p < 0.001$ ). CAPE treatment reversed these effects as well ( $p < 0.0001$ ,  $p < 0.001$  respectively). In conclusion, the increase in NO and MDA levels of renal tissue, and in urinary NAG with the decrease in renal SOD, CAT, GSH-Px activities demonstrate the role of oxidative mechanisms in 900 MHz mobile phone-induced renal tissue damage, and CAPE, via its free radical scavenging and antioxidant properties, ameliorates oxidative renal damage. These results strongly suggest that CAPE exhibits a protective effect on mobile phone-induced and free radical mediated oxidative renal impairment in rats.

**(E) Ozguner F, Oktem F, Armagan A, Yilmaz R, Koyu A, Demirel R, Vural H, Uz E. Comparative analysis of the protective effects of melatonin and caffeic acid phenethyl ester (CAPE) on mobile phone-induced renal impairment in rat. Mol Cell Biochem. 276(1-2):31-37, 2005.**

Melatonin and caffeic acid phenethyl ester (CAPE), a component of honeybee propolis, were recently found to be potent free radical scavengers and antioxidants. There are a number of reports on the effects induced by electromagnetic radiation (EMR) in various cellular systems. Mechanisms of adverse effects of EMR indicate that reactive oxygen species may play a role in the biological effects of this radiation. The present study was carried out to compare the protective effects of melatonin and CAPE against 900 MHz EMR emitted mobile phone-induced renal tubular injury. Melatonin was administered whereas CAPE was given for 10 days before the exposure. Urinary N-acetyl-beta-D-glucosaminidase (NAG, a marker of renal tubular injury) and malondialdehyde (MDA, an index of lipid peroxidation), were used as markers of oxidative stress-induced renal impairment in rats exposed to EMR. Superoxide dismutase (SOD), catalase (CAT), and glutathione peroxidase (GSH-Px) activities were studied to evaluate the changes of antioxidant status in renal tissue. Urinary NAG and renal MDA were increased in EMR exposed rats while both melatonin and CAPE caused a significant reduction in the levels of these parameters. Likewise, renal SOD and GSH-Px activities were decreased in EMR exposed animals while melatonin caused a significant increase in the activities of these antioxidant enzymes but CAPE did not. Melatonin caused a significant decrease in urinary NAG activity and MDA levels which were increased because of EMR exposure. CAPE also reduced elevated MDA levels in EMR exposed renal tissue, but the effect of melatonin was more potent than that of CAPE. Furthermore, treatment of EMR exposed rats with melatonin increased activities of SOD and GSH-Px to higher levels than those of control rats. In conclusion, melatonin and CAPE prevent renal tubular injury by reducing oxidative stress and protect the kidney from oxidative damage induced by 900 MHz mobile phone. Nevertheless, melatonin seems to be a more potent antioxidant compared with CAPE in kidney.

**(E) Ozguner F, Altinbas A, Ozaydin M, Dogan A, Vural H, Kisioglu AN, Cesur G, Yildirim NG. Mobile phone-induced myocardial oxidative stress: protection by a novel antioxidant agent caffeic acid phenethyl ester. Toxicol Ind Health. 21(9):223-230, 2005.**

Electromagnetic radiation (EMR) or radiofrequency fields of cellular mobile phones may affect biological systems by increasing free radicals, which appear mainly to enhance lipid

peroxidation, and by changing the antioxidant defense systems of human tissues, thus leading to oxidative stress. Mobile phones are used in close proximity to the heart, therefore 900 MHz EMR emitting mobile phones may be absorbed by the heart. Caffeic acid phenethyl ester (CAPE), one of the major components of honeybee propolis, was recently found to be a potent free radical scavenger and antioxidant, and is used in folk medicine. The aim of this study was to examine 900 MHz mobile phone-induced oxidative stress that promotes production of reactive oxygen species (ROS) and the role of CAPE on myocardial tissue against possible oxidative damage in rats. Thirty rats were used in the study. Animals were randomly grouped as follows: sham-operated control group (N: 10) and experimental groups: (a) group II: 900 MHz EMR exposed group (N: 10); and (b) group III: 900 MHz EMR exposed+CAPE-treated group (N: 10). A 900 MHz EMR radiation was applied to groups II and III 30 min/day, for 10 days using an experimental exposure device. Malondialdehyde (MDA, an index of lipid peroxidation), and nitric oxide (NO, a marker of oxidative stress) were used as markers of oxidative stress-induced heart impairment. Superoxide dismutase (SOD), catalase (CAT), and glutathione peroxidase (GSH-Px) activities were studied to evaluate the changes of antioxidant status. In the EMR exposed group, while tissue MDA and NO levels increased, SOD, CAT and GSH-Px activities were reduced. CAPE treatment in group III reversed these effects. In this study, the increased levels of MDA and NO and the decreased levels of myocardial SOD, CAT and GSH-Px activities demonstrate the role of oxidative mechanisms in 900 MHz mobile phone-induced heart tissue damage, and CAPE, via its free radical scavenging and antioxidant properties, ameliorates oxidative heart injury. These results show that CAPE exhibits a protective effect on mobile phone-induced and free radical mediated oxidative heart impairment in rats.

**(E) Ozguner F, Bardak Y, Comlekci S. Protective effects of melatonin and caffeic acid phenethyl ester against retinal oxidative stress in long-term use of mobile phone: A comparative study. Mol Cell Biochem. 282(1-2):83-88, 2006.**

There are numerous reports on the effects of electromagnetic radiation (EMR) in various cellular systems. Melatonin and caffeic acid phenethyl ester (CAPE), a component of honeybee propolis, were recently found to be potent free radical scavengers and antioxidants. Mechanisms of adverse effects of EMR indicate that reactive oxygen species may play a role in the biological effects of this radiation. The present study was carried out to compare the efficacy of the protective effects of melatonin and CAPE against retinal oxidative stress due to long-term exposure to 900 MHz EMR emitting mobile phones. Melatonin and CAPE were administered daily for 60 days to the rats prior to their EMR exposure during our study. Nitric oxide (NO, an oxidant product) levels and malondialdehyde (MDA, an index of lipid peroxidation), were used as markers of retinal oxidative stress in rats following to use of EMR. Superoxide dismutase (SOD), catalase (CAT), and glutathione peroxidase (GSH-Px) activities were studied to evaluate the changes of antioxidant status in retinal tissue. Retinal levels of NO and MDA increased in EMR exposed rats while both melatonin and CAPE caused a significant reduction in the levels of NO and MDA. Likewise, retinal SOD, GSH-Px and CAT activities decreased in EMR exposed animals while melatonin and CAPE caused a significant increase in the activities of these antioxidant enzymes. Treatment of EMR exposed rats with melatonin or CAPE increased the activities of SOD, GSH-Px and CAT to higher levels than those of control rats. In conclusion, melatonin and CAPE reduce retinal oxidative

stress after long-term exposure to 900 MHz emitting mobile phone. Nevertheless, there was no statistically significant difference between the efficacies of these two antioxidants against to EMR induced oxidative stress in rat retina. The difference was in only GSH-Px activity in rat retina. Melatonin stimulated the retinal GSH-Px activity more efficiently than CAPE did.

**(E) Ozgur E, Güler G, Seyhan N. Mobile phone radiation-induced free radical damage in the liver is inhibited by the antioxidants n-acetyl cysteine and epigallocatechin-gallate. Int J Radiat Biol. 86(1):935-945, 2010.**

Purpose: To investigate oxidative damage and antioxidant enzyme status in the liver of guinea pigs exposed to mobile phone-like radiofrequency radiation (RFR) and the potential protective effects of N-acetyl cysteine (NAC) and epigallocatechin-gallate (EGCG) on the oxidative damage. Materials and methods: Nine groups of guinea pigs were used to study the effects of exposure to an 1800-MHz Global System for Mobile Communications (GSM)-modulated signal (average whole body Specific Absorption Rate (SAR) of 0.38 W/kg, 10 or 20 min per day for seven days) and treatment with antioxidants. Results: Significant increases in malondialdehyde (MDA) and total nitric oxide (NO(x)) levels and decreases in activities of superoxide dismutase (SOD), myeloperoxidase (MPO) and glutathione peroxidase (GSH-Px) were observed in the liver of guinea pigs after RFR exposure. Only NAC treatment induces increase in hepatic GSH-Px activities, whereas EGCG treatment alone attenuated MDA level. Extent of oxidative damage was found to be proportional to the duration of exposure ( $P < 0.05$ ). Conclusion: Mobile phone-like radiation induces oxidative damage and changes the activities of antioxidant enzymes in the liver. The adverse effect of RFR may be related to the duration of mobile phone use. NAC and EGCG protect the liver tissue against the RFR-induced oxidative damage and enhance antioxidant enzyme activities.

**(E) \*Ozgur E, Kismali G, Guler G, Akcay A, Ozkurt G, Sel T, Seyhan N. Effects of Prenatal and Postnatal Exposure to GSM-Like Radiofrequency on Blood Chemistry and Oxidative Stress in Infant Rabbits, an Experimental Study. Cell Biochem Biophys. 67(2):743-751, 2013.**

We aimed to investigate the potential hazardous effects of prenatal and/or postnatal exposure to 1800 MHz GSM-like radiofrequency radiation (RFR) on the blood chemistry and lipid peroxidation levels of infant rabbits. A total of 72 New Zealand female and male white rabbits aged 1-month were used. Thirty-six female and 36 male were divided into four groups which were composed of nine infants: (i) Group 1 were the sham exposure (control), (ii) Group 2 were exposed to RFR, 15 min daily for 7 days in the prenatal period (between 15th and 22nd days of the gestational period) (prenatal exposure group). (iii) Group 3 were exposed to RFR 15 min/day (14 days for male, whereas 7 days for female) after they reached 1-month of age (postnatal exposure group). (iv) Group 4 were exposed to RFR for 15 min daily during 7 days in the prenatal period (between 15th and 22nd days of the gestational period) and 15 min/day (14 days for male, whereas 7 days for female) after they reached 1-month of age (prenatal and postnatal exposure group). Results showed that serum lipid peroxidation level in both female and male rabbits changed due to the RFR exposure. However, different parameters of the

blood biochemistry were affected by exposure in male and female infants. Consequently, the whole-body 1800 MHz GSM-like RFR exposure may lead to oxidative stress and changes on some blood chemistry parameters. Studies on RFR exposure during prenatal and postnatal periods will help to establish international standards for the protection of pregnant and newborns from environmental RFR.

**(E) Ozgur E, Sahin D, Tomruk A, Guler G, Sepici-Dinçel A, Altan N, Seyhan N. The Effects of N-acetyl-L-cysteine and Epigallocatechin-3-gallate on Liver Tissue Protein Oxidation and Antioxidant Enzyme Levels After the Exposure to Radio Frequency Radiation. Int J Radiat Biol. 2014 Sep 24:1-19. [Epub ahead of print]**

**PURPOSE:** The widespread and sustained use of mobile and cordless phones causes unprecedented increase of radiofrequency radiation (RFR). The aim of this experimental study was to investigate the effect of 900 MHz Global System for Mobile Communications (GSM) modulated RFR (average whole body Specific Absorption Rate (SAR) of 0,4 W/kg, 10 or 20 min daily for consecutive 7 days) to the liver tissue of guinea pigs and the protective effects of antioxidant treatments. **MATERIALS and METHODS:** Adult male guinea pigs were randomly divided into nine groups as; Group I (Sham/saline), Group II (Sham/EGCG), Group III (Sham/NAC), Group IV (10-min RF-exposure/saline), Group V (20-min RF-exposure/saline), Group VI (10-min RF-exposure/EGCG), Group VII (20-min RF-exposure/EGCG), Group VIII (10-min RF-exposure/NAC), Group IX (20-min RF-exposure/NAC). Protein oxidation (PCO), advanced oxidation protein products (AOPP) and antioxidant enzyme activities of superoxide dismutase (SOD) were evaluated after the exposure and the treatments with N-acetylcysteine (NAC) and (-)-epigallocatechin-3-gallate (EGCG). **RESULTS and CONCLUSIONS:** Significant decreases in the activities of SOD were observed in the liver of guinea pigs after RFR exposure. Protein damage did not change due to RFR exposure. On the other hand, only NAC treatment induces increase PCO levels, whereas EGCG treatment alone elevated the level of AOPP. Due to antioxidants have pro-oxidant behavior, the well decided doses and treatment time tables of NAC and EGCG is needed.

**(E) \*Ozorak A, Nazıroğlu M, Celik O, Yüksel M, Özçelik D, Ozkaya MO, Cetin H, Kahya MC, Kose SA. Wi-Fi (2.45 GHz)- and Mobile Phone (900 and 1800 MHz)-Induced Risks on Oxidative Stress and Elements in Kidney and Testis of Rats During Pregnancy and the Development of Offspring. Biol Trace Elem Res. 156(1-3):221-229, 2013.**

The present study was designed to determine the effects of both Wi-Fi (2.45 GHz)- and mobile phone (900 and 1800 MHz)-induced electromagnetic radiation (EMR) on oxidative stress and trace element levels in the kidney and testis of growing rats from pregnancy to 6 weeks of age. Thirty-two rats and their 96 newborn offspring were equally divided into four different groups, namely, control, 2.45 GHz, 900 MHz, and 1800 MHz groups. The 2.45 GHz, 900 MHz, and 1,800 MHz groups were exposed to EMR for 60 min/day during pregnancy and growth. During the fourth, fifth, and sixth weeks of the experiment, kidney and testis samples were taken from decapitated rats. Results from the fourth week showed that the level of lipid peroxidation in the kidney and testis and the copper, zinc, reduced glutathione (GSH),

glutathione peroxidase (GSH-Px), and total antioxidant status (TAS) values in the kidney decreased in the EMR groups, while iron concentrations in the kidney as well as vitamin A and vitamin E concentrations in the testis increased in the EMR groups. Results for fifth-week samples showed that iron, vitamin A, and  $\beta$ -carotene concentrations in the kidney increased in the EMR groups, while the GSH and TAS levels decreased. The sixth week results showed that iron concentrations in the kidney and the extent of lipid peroxidation in the kidney and testis increased in the EMR groups, while copper, TAS, and GSH concentrations decreased. There were no statistically significant differences in kidney chromium, magnesium, and manganese concentrations among the four groups. In conclusion, Wi-Fi- and mobile phone-induced EMR caused oxidative damage by increasing the extent of lipid peroxidation and the iron level, while decreasing total antioxidant status, copper, and GSH values. Wi-Fi- and mobile phone-induced EMR may cause precocious puberty and oxidative kidney and testis injury in growing rats.

**(E) Özsoğacı NP, Ergün DD, Durmuş S, Tunçdemir M, Uzun H, Gelişgen R, Özçelik D Selenium supplementation ameliorates electromagnetic field-induced oxidative stress in the HEK293 cells. J Trace Elem Med Biol 2018;50:572-579.**

There is a widespread use of 2.4 GHz electromagnetic radiation emitting devices especially in communication and education. Recent studies show the adverse effects of electromagnetic fields (EMF) such as oxidative stress, cellular damage and apoptosis on tissues. Selenium (Se) has antioxidant properties by inhibiting oxidative damage being within the structure of antioxidant enzymes like glutathione peroxidase (GSH-Px) and it has also regulatory function for cell cycle and apoptosis. The aim of this study was to investigate the effect of Se on 2.4 GHz frequency EMF exposed human embryonic kidney cells (HEK293) by means of alterations in apoptotic and oxidative stress parameters. Our study was planned as control, EMF, 100 nM Se + EMF, 200 nM Se + EMF groups. EMF groups were exposed to 2.4 GHz EMF for 1 h, element groups were incubated with two different doses of Se added cell culture medium for 48 h before EMF exposure. MDA levels were significantly higher whereas SOD and GSH-Px activities were significantly lower in EMF compared to control. 100 and 200 nM Se + EMF application decreased MDA levels, increased SOD and GSH-Px activities than EMF. Apoptosis and caspase-3 were statistically significantly higher but bcl-2 was lower in EMF than control. Apoptosis and caspase-3 were lower in 100 and 200 nM Se + EMF, although bcl-2 were higher than EMF. In conclusion, Se has protective effects against 2.4 GHz EMF-induced oxidative stress by reducing lipid peroxidation, regulating SOD and GSH-Px activity. Also, Se has inhibitory effect on 2.4 GHz EMF induced apoptosis by increasing the expression of anti-apoptotic protein bcl-2 and suppressing apoptosis regulatory protein caspase-3.

**(E) Özsoğacı NP, Ergün DD, Tunçdemir M, Özçelik D. Protective Effects of Zinc on 2.45 GHz Electromagnetic Radiation-Induced Oxidative Stress and Apoptosis in HEK293 Cells. Biol Trace Elem Res. 194:368-378, 2020.**

Several epidemiological studies have shown that exposure to electromagnetic radiation (EMR) can be harmful to human health. The purpose of this study was to examine oxidative parameters and apoptosis induced by EMR in human kidney embryonic cells (HEK293) and to investigate whether zinc (Zn) has protective effect on EMR-induced apoptosis in HEK293 cells. For our experiment, HEK293 cells were divided into four main groups, control, EMR, 50  $\mu$ M Zn + EMR, and 100  $\mu$ M Zn + EMR. HEK293 cells of EMR groups were exposed to 2.45 GHz EMR for 1 h. In Zn groups, HEK293 cells were incubated with different concentrations of Zn for 48 h before EMR exposure. Oxidative stress parameters were determined by spectrophotometric method; bcl-2 and caspase-3 were assessed immunohistochemically and TUNEL method was performed for apoptotic activity. EMR group had higher malondialdehyde (MDA) level and lower superoxide dismutase (SOD) activity compared with control group. In Zn-applied groups, MDA was decreased and SOD activity was increased compared with EMR group. The number of the apoptotic cells and caspase-3 immunopositive cells at EMR group was increased significantly compared with the control group, whereas bcl-2 was decreased. Besides, Zn-treated groups showed a significant reduction in the number of apoptotic cells and caspase-3 from that of EMR group, whereas there was an increase in bcl-2 immunopositivity. Our findings show that EMR caused oxidative stress and apoptotic activation in HEK293 cells. Zn seems to have protective effects on the EMR by increasing SOD activity and bcl-2 immunopositivity, decreasing lipid peroxidation and caspas-3 immunopositivity.

**(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):373-384, 2017.**

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.**

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.

**(E) Paredi P, Kharitonov SA, Hanazawa T, Barnes PJ, Local vasodilator response to mobile phones. Laryngoscope 111(1):159-162, 2001.**

**OBJECTIVES:** The use of mobile phones with the resulting generation of potentially harmful electromagnetic fields (EMF) is the focus of public interest. Heat generation and the activation of the inducible form of nitric oxide (NO) synthase may be possible causes of the biological effects of EMF exposure. We investigated if a mobile telephone conversation can modify skin temperature, NO, and nasal resistance. **METHODS:** We studied the effect of an EMF (900 MHz) generated by a commercially available cellular phone during a 30-minute telephone conversation on skin temperature, nasal NO measured by chemiluminescence, and nasal minimal cross-sectional area (MCA) measured by rhinometry. Eleven normal subjects (mean age +/- standard error of mean [SEM], 32 +/- 5 y; 10 male) were studied. **RESULTS:** There was a similar and significant increase in skin temperature of the nostril and occipital area on the same side as the telephone (maximal increase 2.3 +/- 0.2 degrees C at 6 min) as well as a tendency for higher nasal NO levels (maximal increase 12.9 +/- 4.9% at 10 min), whereas the

MCA was significantly reduced (maximal decrease  $-27 \pm 6\%$  at 15 min). Such changes were not recorded when an earpiece was used to avoid the direct exposure to the electromagnetic field. There were no changes in the skin temperature and nasal NO measured on the opposite side to the mobile phone, whereas the MCA was significantly increased ( $38 \pm 10\%$ ).

CONCLUSIONS: Exposure to EMF produced by a mobile phone produces biological effects that can be easily measured. Microwaves may increase skin temperature and therefore cause vasodilation and reduce MCA. Further studies are needed to study the long-term effects of mobile phone use and the relation among NO production, vasodilation, and temperature.

**(E) Pastacı Özsoğacı N, Düzgün Ergün D, Durmuş S, Tunçdemir M, Uzun H, Erdoğan Gelişgen R, Özçelik D. Selenium supplementation ameliorates electromagnetic field-induced oxidative stress in the HEK293 cells. J Trace Elem Med Biol.:572-579, 2018.**

There is a widespread use of 2.4 GHz electromagnetic radiation emitting devices especially in communication and education. Recent studies show the adverse effects of electromagnetic fields (EMF) such as oxidative stress, cellular damage and apoptosis on tissues. Selenium (Se) has antioxidant properties by inhibiting oxidative damage being within the structure of antioxidant enzymes like glutathione peroxidase (GSH-Px) and it has also regulatory function for cell cycle and apoptosis. The aim of this study was to investigate the effect of Se on 2.4 GHz frequency EMF exposed human embryonic kidney cells (HEK293) by means of alterations in apoptotic and oxidative stress parameters. Our study was planned as control, EMF, 100 nM Se + EMF, 200 nM Se + EMF groups. EMF groups were exposed to 2.4 GHz EMF for 1 h, element groups were incubated with two different doses of Se added cell culture medium for 48 h before EMF exposure. MDA levels were significantly higher whereas SOD and GSH-Px activities were significantly lower in EMF compared to control. 100 and 200 nM Se + EMF application decreased MDA levels, increased SOD and GSH-Px activities than EMF. Apoptosis and caspase-3 were statistically significantly higher but bcl-2 was lower in EMF than control. Apoptosis and caspase-3 were lower in 100 and 200 nM Se + EMF, although bcl-2 were higher than EMF. In conclusion, Se has protective effects against 2.4 GHz EMF-induced oxidative stress by reducing lipid peroxidation, regulating SOD and GSH-Px activity. Also, Se has inhibitory effect on 2.4 GHz EMF induced apoptosis by increasing the expression of anti-apoptotic protein bcl-2 and suppressing apoptosis regulatory protein caspase-3.

**(E) Payez A, Ghanati F, Behmanesh M, Abdolmaleki P, Hajnorouzi A, Rajabbeigi E. Increase of seed germination, growth and membrane integrity of wheat seedlings by exposure to static and a 10-KHz electromagnetic field. Electromagn Biol Med. 32(4):417-429, 2013.**

There is a large body of experimental data demonstrating various effects of magnetic field (MF) on plants growth and development. Although the mechanism(s) of perception of MF by plants is not yet elucidated, there is a possibility that like other stimuli, MF exerts its effects on plants by changing membrane integrity and

conductance of its water channels, thereby influencing growth characteristics. In this study, the seeds of wheat (*Triticum aestivum* L. cv. Kavir) were imbibed in water overnight and then treated with or without a 30-mT static magnetic field (SMF) and a 10-kHz electromagnetic field (EMF) for 4 days, each 5 h. Water uptake of seeds reduced 5 h of the treatment with EMF but did not show changes in SMF treatment. Exposure to both magnetic fields did not affect germination percent of the seeds but increased the speed of germination, compared to the control group. Treatment with EMF significantly reduced seedling length and subsequently vigor index I, while SMF had no effects on these parameters. Both treatments significantly increased vigor index II, compared to the control group. These treatments also remarkably increased catalase activity and proline contents of seedlings but reduced the activity of peroxidase, the rate of lipid peroxidation and electrolyte leakages of membranes. The results suggest promotional effects of EMFs on membrane integrity and growth characteristics of wheat seedlings.

**(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.**

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) Pilla AA. Electromagnetic fields instantaneously modulate nitric oxide signaling in challenged biological systems. *Biochem Biophys Res Commun.* 426(3):330-333, 2012.**

This study shows that a non-thermal pulse-modulated RF signal (PRF), configured to modulate calmodulin (CaM) activation via acceleration of Ca<sup>2+</sup> binding kinetics, produced

an immediate nearly 3-fold increase in nitric oxide (NO) from dopaminergic MN9D cultures ( $P < 0.001$ ). NO was measured electrochemically in real-time using a NO selective membrane electrode, which showed the PRF effect occurred within the first seconds after lipopolysaccharide (LPS) challenge. Further support that the site of action of PRF involves CaM is provided in human fibroblast cultures challenged with low serum and exposed for 15min to the identical PRF signal. In this case a CaM antagonist W-7 could be added to the culture 3h prior to PRF exposure. Those results showed the PRF signal produced nearly a two-fold increase in NO, which could be blocked by W-7 ( $P < 0.001$ ). To the authors' knowledge this is the first report of a real-time effect of non-thermal electromagnetic fields (EMF) on NO release from challenged cells. The results provide mechanistic support for the many reported bioeffects of EMF in which NO plays a role. Thus, in a typical clinical application for acute post operative pain, or chronic pain from, e.g., osteoarthritis, EMF therapy could be employed to modulate the dynamics of NO via Ca/CaM-dependent constitutive nitric oxide synthase (cNOS) in the target tissue. This, in turn, would modulate the dynamics of the signaling pathways the body uses in response to the various phases of healing after physical or chemical insult or injury.

**(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.**

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) Postaci I, Coskun O, Senol N, Aslankoc R, Comlekci S. The physiopathological effects of quercetin on oxidative stress in radiation of 4.5 g mobile phone exposed liver tissue of rat. Bratisl Lek Listy 2018;119(8):481-489.**

Objective: The study was aimed to evaluate the physiopathological consideration of the effects of electromagnetic field (EMF) from the radiation of 4.5 G mobile phones on the liver tissue

of rats and quercetin (Qu) applied as an antioxidant for reducing these effects. Methods: Male Wistar-Albino rats were divided into four groups with 8 rats in each group. Group 1 (control group), Group 2 (sham group), Group 3 (EMF group) and Group 4 (EMF + Qu). From the animals sacrificed at the end of the 30th day; liver tissues were taken for histopathological and immunohistochemical examinations. Results: In the liver tissue of the electromagnetic field group; dilatation of sinusitis was determined to be higher than in the sham group. It was concluded that the concentration of caspase-3 and TNF- $\alpha$  immunopositive cells was in the EMF group (+3) level and also the immunostaining was stronger, it caused an increase in malondialdehyde level, the difference between the groups was statistically significant, in terms of superoxide dismutase, catalase activities, the difference was not significant. Conclusion: It was determined that 2600 MHz EMF exposure caused damage to the liver, 100 mg/kg/day quercetin was not sufficient to prevent this damage (Tab. 5, Fig. 15, Ref. 27).

**(NE) Poulletier de Gannes F, Haro E, Hurtier A, Taxile M, Ruffié G, Billaudel B, Veyret B, Lagroye I. Effect of exposure to the edge signal on oxidative stress in brain cell models. Radiat Res. 175(2):225-230, 2011.**

In this study we investigated the effect of the Enhanced Data rate for GSM Evolution (EDGE) signal on cells of three human brain cell lines, SH-SY5Y, U87 and CHME5, used as models of neurons, astrocytes and microglia, respectively, as well as on primary cortical neuron cultures. SXC-1800 waveguides (IT'IS-Foundation, Zürich, Switzerland) were modified for in vitro exposure to the EDGE signal radiofrequency (RF) radiation at 1800 MHz. Four exposure conditions were tested: 2 and 10 W/kg for 1 and 24 h. The production of reactive oxygen species (ROS) was measured by flow cytometry using the dichlorofluorescein diacetate (DCFH-DA) probe at the end of the 24-h exposure or 24 h after the 1-h exposure. Rotenone treatment was used as a positive control. All cells tested responded to rotenone treatment by increasing ROS production. These findings indicate that exposure to the EDGE signal does not induce oxidative stress under these test conditions, including 10 W/kg. Our results are in agreement with earlier findings that RF radiation alone does not increase ROS production.

**(E) Qin F, Yuan H, Nie J, Cao Y, Tong J. [Effects of nano-selenium on cognition performance of mice exposed in 1800 MHz radiofrequency fields]. Wei Sheng Yan Jiu. 43(1):16-21, 2014. [Article in Chinese]**

OBJECTIVE: To study the effects of nano-selenium (NSe) on cognition performance of mice exposed to 1800 MHz radiofrequency fields (RF). METHODS: Male mice were randomly divided into four groups, control and nano-Se low, middle and high dose groups (L, M, H). Each group was sub-divided into three groups, RF 0 min, RF 30 min and RF 120 min. Nano-se solution (2, 4 and 8 microg/ml) were administered to mice of L, M, H groups by intragastric injection respectively, 0.5 ml/d for 50 days, the control group were administered with distilled water. At the 21st day, the mice in RF subgroup were exposed to 208 microW/cm<sup>2</sup> 1800 MHz radiofrequency fields (0, 30 and 120 min/d respectively) for 30 days. The cognitive ability of the mice were tested with Y-maze. Further, the levels of MDA, GABA, Glu, Ach and the activities of CAT and GSH-Px in cerebra were measured. RESULTS: Significant impairments in learning and memory ( $P < 0.05$ ) were observed in the RF 120 min group, and

with reduction of the Ach level and the activities of CAT and GSH-Px and increase of the content of GABA, Glu and MDA in cerebrum. NSe enhanced cognitive performance of RF mice, decreased GABA, Glu and MDA levels, increased Ach levels, GSH-Px and CAT activities. CONCLUSION: NSe could improve cognitive impairments of mice exposed to RF, the mechanism of which might involve the increasing antioxidation, decreasing free radical content and the changes of cerebra neurotransmitters.

**(E) Qin F, Shen T, Cao H, Qian J, Zou D, Ye M, Pei H. CeO<sub>2</sub>NPs relieve radiofrequency radiation, improve testosterone synthesis, and clock gene expression in Leydig cells by enhancing antioxidation. Int J Nanomedicine. 14:4601-4611, 2019.**

**Introduction:** The ratio of Ce<sup>3+</sup>/Ce<sup>4+</sup> in their structure confers unique functions on cerium oxide nanoparticles (CeO<sub>2</sub>NPs) containing rare earth elements in scavenging free radicals and protecting against oxidative damage. The potential of CeO<sub>2</sub>NPs 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<sub>2</sub>NPs to identify the optimum concentration for cell proliferation. The cells were pretreated with the optimum dose of CeO<sub>2</sub>NPs for 24 hrs and then exposed to 1,800 MHz RF at a power density of 200.27 μW/cm<sup>2</sup> (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 *Rora*). **Results:** Our preliminary result showed that 128 μg/mL CeO<sub>2</sub>NPs 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 *Rora*. Pretreatment of the cells with 128 μg/mL CeO<sub>2</sub>NPs 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<sub>2</sub>NPs 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<sub>2</sub>NPs against RF in the male reproductive system are required.

**(E) Ragy MM. Effect of exposure and withdrawal of 900-MHz-electromagnetic waves on brain, kidney and liver oxidative stress and some biochemical parameters in male rats. Electromagn Biol Med. 2014 Apr 8. [Epub ahead of print]**

Increasing use of mobile phones in daily life with increasing adverse effects of electromagnetic radiation (EMR), emitted from mobile on some physiological processes, cause many concerns about their effects on human health. Therefore, this work was designed to study the effects of exposure to mobile phone emits 900-MHz EMR on the brain, liver and kidney of male albino rats. Thirty male adult rats were randomly divided into four groups (10

each) as follows: control group (rats without exposure to EMR), exposure group (exposed to 900-MHz EMR for 1 h/d for 60 d) and withdrawal group (exposed to 900-MHz electromagnetic wave for 1 h/d for 60 d then left for 30 d without exposure). EMR emitted from mobile phone led to a significant increase in malondialdehyde (MDA) levels and significant decrease total antioxidant capacity (TAC) levels in brain, liver and kidneys tissues. The sera activity of alanine transaminase (ALT), aspartate aminotransferase (AST), urea, creatinine and corticosterone were significantly increased ( $p < 0.05$ ), while serum catecholamines were insignificantly higher in the exposed rats. These alterations were corrected by withdrawal. In conclusion, electromagnetic field emitting from mobile phone might produce impairments in some biochemicals changes and oxidative stress in brain, liver and renal tissue of albino rats.

**(E) Şahin D, Özgür E, Güler G, Tomruk A, Ünlü İ, Sepici-Dinçel A, Seyhan N. The 2100MHz radiofrequency radiation of a 3G-mobile phone and the DNA oxidative damage in brain. J Chem Neuroanat. 75 (Pt B):94-98, 2016.**

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) Saikhedkar N, Bhatnagar M, Jain A, Sukhwal P, Sharma C, Jaiswal N. Effects of mobile phone radiation (900 MHz radiofrequency) on structure and functions of rat brain. Neurol Res. 36:1072-1079, 2014.**

Objectives: The goals of this study were: (1) to obtain basic information about the effects of long-term use of mobile phone on cytological makeup of the hippocampus in rat brain (2) to evaluate the effects on antioxidant status, and (3) to evaluate the effects on cognitive behavior particularly on learning and memory. Methods: Rats (age 30 days,  $120 \pm 5$  g) were exposed to 900 MHz radio waves by means of a mobile hand set for 4 hours per day for 15 days. Effects on anxiety, spatial learning, and memory were studied using open field test, elevated plus maze, Morris water maze (MWM), and classic maze test. Effects on brain antioxidant status were also studied. Cresyl violet staining was done to assess the neuronal damage. Result: A significant change in behavior, i.e., more anxiety and poor learning was shown by test animals

as compared to controls and sham group. A significant change in level of antioxidant enzymes and non-enzymatic antioxidants, and increase in lipid peroxidation were observed in test rats. Histological examination showed neurodegenerative cells in hippocampal sub regions and cerebral cortex. Discussion: Thus our findings indicate extensive neurodegeneration on exposure to radio waves. Increased production of reactive oxygen species due to exhaustion of enzymatic and non-enzymatic antioxidants and increased lipid peroxidation are indicating extensive neurodegeneration in selective areas of CA1, CA3, DG, and cerebral cortex. This extensive neuronal damage results in alterations in behavior related to memory and learning.

**(E) Salah MB, Abdelmelek H, Abderraba M. Effects of olive leave extract on metabolic disorders and oxidative stress induced by 2.45 GHz WIFI signals. Environ Toxicol Pharmacol. 36(3):826-834, 2013.**

We investigated the effect of olive leaves extract administration on glucose metabolism and oxidative response in liver and kidneys of rats exposed to radio frequency (RF). The exposure of rats to RF (2.45 GHz, 1h/day during 21 consecutive days) induced a diabetes-like status. Moreover, RF decreased the activities of glutathione peroxidase (GPx, -33.33% and -49.40%) catalase (CAT, -43.39% and -39.62%) and the superoxide dismutase (SOD, -59.29% and -68.53%) and groups thiol amount (-62.68% and -34.85%), respectively in liver and kidneys. Indeed, exposure to RF increased the malondialdehyde (MDA, 29.69% and 51.35%) concentration respectively in liver and kidneys. Olive leaves extract administration (100 mg/kg, ip) in RF-exposed rats prevented glucose metabolism disruption and restored the activities of GPx, CAT and SOD and thiol group amount in liver and kidneys. Moreover, olive leave extract administration was able to bring down the elevated levels of MDA in liver but not in kidneys. Our investigations suggested that RF exposure induced a diabetes-like status through alteration of oxidative response. Olive leaves extract was able to correct glucose metabolism disorder by minimizing oxidative stress induced by RF in rat tissues.

**(E) Santini SJ, Cordone V, Falone S, Mijit M, Tatone C, Amicarelli F, Di Emidio G. Role of Mitochondria in the Oxidative Stress Induced by Electromagnetic Fields: Focus on Reproductive Systems. Oxid Med Cell Longev. 2018 Nov 8;2018:5076271.**

Modern technologies relying on wireless communication systems have brought increasing levels of electromagnetic field (EMF) exposure. This increased research interest in the effects of these radiations on human health. There is compelling evidence that EMFs affect cell physiology by altering redox-related processes. Considering the importance of redox *milieu* in the biological competence of oocyte and sperm, we reviewed the existing literature regarding the effects of EMFs on reproductive systems. Given the role of mitochondria as the main source of reactive oxygen species (ROS), we focused on the hypothesis of a mitochondrial basis of EMF-induced reproductive toxicity. MEDLINE, Web of Science, and Scopus database were examined for peer-reviewed original articles by searching for the following keywords: "extremely low frequency electromagnetic fields (ELF-EMFs)," "radiofrequency (RF)," "microwaves," "Wi-Fi," "mobile phone," "oxidative stress," "mitochondria," "fertility," "sperm," "testis," "oocyte," "ovarian follicle," and "embryo."

These keywords were combined with other search phrases relevant to the topic. Although we reported contradictory data due to lack of uniformity in the experimental designs, a growing body of evidence suggests that EMF exposure during spermatogenesis induces increased ROS production associated with decreased ROS scavenging activity. Numerous studies revealed the detrimental effects of EMFs from mobile phones, laptops, and other electric devices on sperm quality and provide evidence for extensive electron leakage from the mitochondrial electron transport chain as the main cause of EMF damage. In female reproductive systems, the contribution of oxidative stress to EMF-induced damages and the evidence of mitochondrial origin of ROS overproduction are reported, as well. In conclusion, mitochondria seem to play an important role as source of ROS in both male and female reproductive systems under EMF exposure. Future and more standardized studies are required for a better understanding of molecular mechanisms underlying EMF potential challenge to our reproductive system in order to improve preventive strategies.

**(E) Saygin M, Asci H, Ozmen O, Cankara FN, Dincoglu D, Ilhan I. Impact of 2.45 GHz microwave radiation on the testicular inflammatory pathway biomarkers in young rats: The role of gallic acid. Environ Toxicol. 2015 Aug 13. doi: 10.1002/tox.22179. [Epub ahead of print]**

The aim of this study was to investigate electromagnetic radiation (EMR) transmitted by wireless devices (2.45 GHz), which may cause physiopathological or ultrastructural changes, in the testes of rats. We addressed if the supplemental gallic acid (GA) may reduce these adverse effects. Six-week-old male Sprague Dawley rats were used in this study. Forty eight rats were equally divided into four groups, which were named: Sham, EMR only (EMR, 3 h day<sup>-1</sup> for 30 days), EMR + GA (30 mg/kg/daily), and GA (30 mg/kg/daily) groups. Malondialdehyde (MDA) and total oxidant status (TOS) levels increased ( $p = 0.001$  for both) in EMR only group. TOS and oxidative stress index (OSI) levels decreased in GA treated group significantly ( $p = 0.001$  and  $p = 0.045$ , respectively). Total antioxidant status (TAS) activities decreased in EMR only group and increased in GA treatment group ( $p = 0.001$  and  $p = 0.029$ , respectively). Testosterone and vascular endothelial growth factor (VEGF) levels decreased in EMR only group, but this was not statistically significant. Testosterone and VEGF levels increased in EMR+GA group, compared with EMR only group ( $p = 0.002$ ), and also increased in GA group compared with the control and EMR only group ( $p = 0.044$  and  $p = 0.032$ , respectively). Prostaglandin E<sub>2</sub> (PGE<sub>2</sub>) and calcitonin gene related peptide (CGRP) staining increased in tubules of the testes in EMR only group ( $p < 0.001$  for both) and decreased in tubules of the testes in EMR+GA group ( $p < 0.001$  for all parameters). In EMR only group, most of the tubules contained less spermatozoa, and the spermatozoon counts decreased in tubules of the testes. All these findings and the regenerative reaction, characterized by mitotic activity, increased in seminiferous tubules cells of the testes in EMR+GA group ( $p < 0.001$ ). Long term EMR exposure resulted in testicular physiopathology via oxidative damage and inflammation. GA may have ameliorative effects on the prepubertal rat testes physiopathology.

**Schuermann D, Mevissen M Manmade Electromagnetic Fields and Oxidative Stress-Biological Effects and Consequences for Health. Int J Mol Sci 2021 Apr 6;22(7):3772.(Review)**

Concomitant with the ever-expanding use of electrical appliances and mobile communication systems, public and occupational exposure to electromagnetic fields (EMF) in the extremely-low-frequency and radiofrequency range has become a widely debated environmental risk factor for health. Radiofrequency (RF) EMF and extremely-low-frequency (ELF) MF have been classified as possibly carcinogenic to humans (Group 2B) by the International Agency for Research on Cancer (IARC). The production of reactive oxygen species (ROS), potentially leading to cellular or systemic oxidative stress, was frequently found to be influenced by EMF exposure in animals and cells. In this review, we summarize key experimental findings on oxidative stress related to EMF exposure from animal and cell studies of the last decade. The observations are discussed in the context of molecular mechanisms and functionalities relevant to health such as neurological function, genome stability, immune response, and reproduction. Most animal and many cell studies showed increased oxidative stress caused by RF-EMF and ELF-MF. In order to estimate the risk for human health by manmade exposure, experimental studies in humans and epidemiological studies need to be considered as well.

**(E) Sefidbakht Y, Moosavi-Movahedi AA, Hosseinkhani S, Khodaghali F, Torkzadeh-Mahani M, Foolad F, Faraji-Dana R. Effects of 940 MHz EMF on bioluminescence and oxidative response of stable luciferase producing HEK cells. Photochem Photobiol Sci. 13:1082-1092, 2014.**

The effects of mobile phone frequency electromagnetic field (RF-EMF, 940 MHz) on a stable cell line (HEK293T) harbouring the firefly luciferase gene were evaluated. A waveguide exposure system with 1 W input power provided the mean specific absorption rate of  $\approx 0.09 \text{ W kg}^{-1}$  in 35 mm Petri dishes. The effects of exposure duration (15, 30, 45, 60 and 90 min) on luciferase activity and oxidative response elements were investigated. Endogenous luciferase activity was reduced after 30 and 45 min of continuous exposure, while after 60 min, the exposed cell lysate showed higher luciferase activity compared with the non-exposed control. Reactive oxygen species (ROS) generation was highest in the 30 min exposed cells as studied by 2',7'-dichlorodihydrofluorescein diacetate (DCFH-DA) fluorescence. The observed boost in ROS was then followed by a sharp rise in catalase (CAT) and superoxide dismutase (SOD) activity and elevation of glutathione (GSH) during the 45 min exposure. Decrease in lipid peroxidation (malondialdehyde, MDA) was meaningful for the 45 and 60 min exposed cells. Therefore, it appears that an increase in the activity of luciferase after 60 min of continuous exposure could be associated with a decrease in ROS level caused by activation of the oxidative response. This ability in cells to overcome oxidative stress and compensate the luciferase activity could also be responsible for the adaptive response mechanism detected in ionizing radiation studies with RF-EMF pre-treatments.

**(E) 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 Hypertrophy. Indian J Otolaryngol Head Neck Surg. 2019 Nov;71(Suppl 3):1810-1815, 2019.**

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:  $OSI = (TOS/TAS) \times 100$ ). 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) Sepehrimanesh M, Kazemipour N, Saeb M, Nazifi S. Analysis of rat testicular proteome following 30-days exposure to 900 MHz electromagnetic field radiation. Electrophoresis. 2014 Aug 21. doi: 10.1002/elps.201400273. [Epub ahead of print]**

The use of electromagnetic field (EMF) generating apparatuses such as cell phones is increasing, and has caused an interest in the investigations of its effects on human health. We analyzed proteome in preparations from the whole testis in adult male Sprague-Dawley rats exposed for 1, 2 or 4 h/d for 30 consecutive days to 900 MHz EMF radiation, simulating a range of possible human cell phone use. Subjects were sacrificed immediately after the end of the experiment and testes fractions were solubilized and separated via high resolution 2-dimensional electrophoresis, and gel patterns were scanned, digitized and processed. Thirteen of the proteins which found only in sham or in exposure groups were identified by MALDI-TOF/TOF-MS. Among them, heat shock proteins, superoxide dismutase, peroxiredoxin-1 and other proteins related to misfolding of proteins and/or stress were identified. These results demonstrate significant effects of radio-frequency modulated electromagnetic fields (RF-EMF) exposure on proteome, particularly in protein species in the rodent testis, and suggest that a 30 d exposure to EMF radiation induces non-thermal stress in testicular tissue. The functional implication of the identified proteins was discussed.

**(E) Shahin S, Mishra V, Singh SP, Chaturvedi CM. 2.45-GHz microwave irradiation adversely affects reproductive function in male mouse, *Mus musculus* by inducing oxidative and nitrosative stress. Free Radic Res. 48(5):511-525, 2014.**

Electromagnetic radiations are reported to produce long-term and short-term biological effects, which are of great concern to human health due to increasing use of devices emitting EMR especially microwave (MW) radiation in our daily life. In view of the unavoidable use

of MW emitting devices (microwaves oven, mobile phones, Wi-Fi, etc.) and their harmful effects on biological system, it was thought worthwhile to investigate the long-term effects of low-level MW irradiation on the reproductive function of male Swiss strain mice and its mechanism of action. Twelve-week-old mice were exposed to non-thermal low-level 2.45-GHz MW radiation (CW for 2 h/day for 30 days, power density = 0.029812 mW/cm<sup>2</sup>) and SAR = 0.018 W/Kg). Sperm count and sperm viability test were done as well as vital organs were processed to study different stress parameters. Plasma was used for testosterone and testis for 3 $\beta$  HSD assay. Immunohistochemistry of 3 $\beta$  HSD and nitric oxide synthase (i-NOS) was also performed in testis. We observed that MW irradiation induced a significant decrease in sperm count and sperm viability along with the decrease in seminiferous tubule diameter and degeneration of seminiferous tubules. Reduction in testicular 3 $\beta$  HSD activity and plasma testosterone levels was also noted in the exposed group of mice. Increased expression of testicular i-NOS was observed in the MW-irradiated group of mice. Further, these adverse reproductive effects suggest that chronic exposure to nonionizing MW radiation may lead to infertility via free radical species-mediated pathway.

**(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.**

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 3 $\beta$ - and 17 $\beta$ -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) 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.**

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<sup>2</sup>), 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 S, Singh SP, Chaturvedi CM. Mobile phone (1800MHz) radiation impairs female reproduction in mice, *Mus musculus*, through stress induced inhibition of ovarian and uterine activity. Reprod Toxicol. 73:41-60, 2017.**

Present study investigated the long-term effects of mobile phone (1800MHz) radiation in stand-by, dialing and receiving modes on the female reproductive function (ovarian and uterine histo-architecture, and steroidogenesis) and stress responses (oxidative and nitrosative stress). We observed that mobile phone radiation induces significant elevation in ROS, NO, lipid peroxidation, total carbonyl content and serum corticosterone coupled with significant decrease in antioxidant enzymes in hypothalamus, ovary and uterus of mice. Compared to control group, exposed mice exhibited reduced number of developing and mature follicles as well as corpus lutea. Significantly decreased serum levels of pituitary gonadotrophins (LH, FSH), sex steroids (E2 and P4) and expression of SF-1, StAR, P-450scc, 3 $\beta$ -HSD, 17 $\beta$ -HSD, cytochrome P-450 aromatase, ER- $\alpha$  and ER- $\beta$  were observed in all the exposed groups of mice, compared to control. These findings suggest that mobile phone radiation induces oxidative and nitrosative stress, which affects the reproductive performance of female mice.

**(E) Sharma A, Kesari KK, Saxena VK, Sisodia R. Ten gigahertz microwave radiation impairs spatial memory, enzymes activity, and histopathology of developing mice brain. Mol Cell Biochem. 2017 May 3. doi: 10.1007/s11010-017-3051-8. [Epub ahead of print]**

For decades, there has been an increasing concern about the potential hazards of non-ionizing electromagnetic fields that are present in the environment and alarming as a major pollutant or electro-pollutant for health risk and neuronal diseases. Therefore, the objective of the present study was to explore the effects of 10 GHz microwave radiation on developing mice brain. Two weeks old mice were selected and divided into two groups (i) sham-exposed and (ii) microwave-exposed groups. Animals were exposed for 2 h/day for 15 consecutive days. After the completion of exposure, within an hour, half of the animals were autopsied immediately and others were allowed to attain 6 weeks of age for the follow-up study. Thereafter results were recorded in terms of various biochemical, behavioral, and histopathological parameters. Body weight result showed significant changes immediately after treatment, whereas non-significant changes were observed in mice attaining 6 weeks of age. Several other endpoints like brain weight, lipid peroxidation, glutathione, protein, catalase, and superoxide dismutase were also found significantly ( $p < 0.05$ ) altered in mice whole brain. These significant differences were found immediately after exposure and also in follow-up on attaining 6 weeks of age in microwave exposure group. Moreover, statistically significant ( $p < 0.001$ ) effect was investigated in spatial memory of the animals, in learning to locate the position of platform in Morris water maze test. Although in probe trial test, sham-exposed animals spent more time in searching for platform into the target quadrant than in opposite or other quadrants. Significant alteration in histopathological parameters (qualitative and quantitative) was also observed in CA1 region of the hippocampus, cerebral cortex, and ansiform lobule of cerebellum. Results from the present study concludes that the brain of 2 weeks aged mice was very sensitive to microwave exposure as observed immediately after exposure and during follow-up study at 6 weeks of age.

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

**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, 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.**

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. Oxidative damage in the liver and brain of the rats exposed to frequency-dependent radiofrequency electromagnetic exposure: Biochemical and histopathological evidence. Free Radic Res 55:535-546, 2021.**

The study aimed to discover a link between the liver and brain's functional status due to frequency dependent-radiofrequency electromagnetic radiation (RF-EMR). 40 Wistar rats were randomly classified as control (sham-exposed) and EMR exposed groups. Animals were exposed to 900, 1800, and 2100 MHz with the specific absorption rate (SAR) 0.434 (W/Kg), 0.433 (W/Kg), and 0.453 (W/Kg) respectively. Animal exposure was limited at 1 hour/day, 5 days/week for 1 month with a restricted power density (900 MHz- 11.638 $\mu$ W/m<sup>2</sup>, 1800- 11.438  $\mu$ W/m<sup>2</sup> and 2100 MHz frequency- 8.237  $\mu$ W/m<sup>2</sup>). Exposure at various frequencies showed a frequency-dependent change in the body weight and hematologic parameters (RBCs, WBCs, platelets, hemoglobin, and hematocrit) as compared with the control group (P  $\leq$  0.01)(P  $\leq$  0.001). A significant elevation in serum transaminases and bilirubin, urea, uric acid, and creatinine was noted, whereas albumin significantly decreased after EMR exposure (P  $\leq$  0.01)(P  $\leq$  0.001). The blood glucose, lipid peroxidation, triglycerides, and cholesterol were elevated while adenosine triphosphatases, acetylcholinesterase, and tissue antioxidants

such as glutathione, superoxide dismutase, catalase, glutathione reductase, glutathione Peroxidase, glutathione-S-transferase, and glucose-6-phosphate dehydrogenases were decreased significantly ( $P \leq 0.001$ ). Histopathological observations of the liver showed centrilobular mononuclear cell infiltration and swelling in sinusoidal spaces, while in the brain degenerated pyramidal and Purkinje neurons were seen. Furthermore, Substantial evidence was found that the brain is more susceptible to oxidative mutilation compare to the liver of exposed animals. In conclusion, RF-EMR exposure showed oxidative damage to the liver, increasing the incidence of brain damage in a frequency-dependent manner. Highlights EMR exposure showed frequency-dependent toxicity. Alterations in blood profile and modifications in the serological markers. Increasing lipid peroxidation indicating membrane damage. Inhibition of acetylcholinesterase activity affecting cholinergic neurotransmission. EMR exposure resulted in the loss of cellular energy and production of excess amounts of ROS thereby altering several antioxidant enzymes. Histopathological evidence of severe degenerative changes in the liver and brain.

**(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* 2022 May 11. doi: 10.1007/s00709-022-01768-9. Online ahead of print.**

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) Sharma VP, Singh HP, Kohli RK, Batish DR. Mobile phone radiation inhibits *Vigna radiata* (mung bean) root growth by inducing oxidative stress. *Sci Total Environ.* 407(21):5543-5547, 2009.**

During the last couple of decades, there has been a tremendous increase in the use of cell phones. It has significantly added to the rapidly increasing EMF smog, an unprecedented type of pollution consisting of radiation in the environment, thereby prompting the scientists to study the effects on humans. However, not many studies have been conducted to explore the effects of cell **phone** EMFr on growth and biochemical changes in plants. We investigated whether EMFr from cell phones inhibit growth of *Vigna radiata* (mung bean) through induction of conventional stress responses. Effects of cell **phone** EMFr (power density: 8.55 microW cm<sup>-2</sup>); 900 MHz band width; for 1/2, 1, 2, and 4 h) were determined by measuring the generation of reactive oxygen species (ROS) in terms of malondialdehyde and hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>) content, root oxidizability and changes in levels of antioxidant enzymes. Our results showed that cell **phone** EMFr significantly inhibited the germination (at > or =2 h), and radicle and plumule growths (> or =1 h) in mung bean in a time-dependent manner. Further, cell **phone** EMFr enhanced MDA content (indicating lipid peroxidation), and increased H<sub>2</sub>O<sub>2</sub> accumulation and root oxidizability in mung bean roots, thereby inducing oxidative stress and **cellular** damage. In response to EMFr, there was a significant upregulation in the activities of scavenging enzymes, such as superoxide dismutases, ascorbate peroxidases, guaiacol peroxidases, catalases and glutathione reductases, in mung bean roots. The study concluded that cell **phone** EMFr inhibit root growth of mung bean by inducing ROS-generated oxidative stress despite increased activities of antioxidant enzymes.

(E)Sharma, VP., Singh, HP., Kohli, RK. Effect of mobile phone EMF on biochemical changes in emerging seedlings of *Phaseolus aureus* Roxb. *Ecoscan* 3:211–214, 2009.

During the last decade or so, there has been a widespread increase in the usage of cell phones resulting in increased electromagnetic radiations in the environment, sometimes referred to as electromagnetic smog. An enhanced level of radiations in the environment has further increased the exposure risks to living organisms. Most of the studies pertaining to ill-effects or risk-analysis of cell phone radiations have been conducted on animals, including humans or microorganisms (Busljeta et al., 2004; Meral et al., 2007). It has been reported that in animals electromagnetic radiations affect cell proliferation, enzyme activity, cell-membrane permeability, and even reproductive capacity (Busljeta et al., 2004; Dimitris et al., 2004; Meral et al., 2007). In spite of these reports indicating ill-effects of mobile phone radiations on physiology and biochemistry in animal systems, little work has been done in plant systems. Ssawastin (1930) was first who observed the effects of magnetic field on plant growth. Further, most of the available reports pertain largely to the effect of electromagnetic fields generated from electrical installations or radio frequency that is lower than mobile phone frequency (Hart and Marino, 1977; Johnson et al., 1979; Soja et al., 2003). Nevertheless, there have been some studies with simulated systems emitting same frequency / wavelength of radiations as that of mobile phones. For example, Tkalec et al., (2005) reported that electromagnetic radiations of 900 MHz inhibit growth of *Lemna minor* after 2 h of exposure. With this background in mind, we conducted a study to explore the effect of electromagnetic radiations on radicle elongation and changes in amounts of proteins, and carbohydrates, and activities of proteases, amylases, and peroxidases in *Phaseolus aureus*

**(E) Sharma VP, Singh HP, Batish DR, Kohli RK. Cell phone radiations affect early growth of *Vigna radiata* (mung bean) through biochemical alterations. Z Naturforsch C. 65(1-2):66-72, 2010.**

The indiscriminate use of wireless technologies, particularly of cell phones, has increased the health risks among living organisms including plants. We investigated the impact of cell phone electromagnetic field (EMF) radiations (power density, 8.55 microW cm<sup>-2</sup>) on germination, early growth, proteins and carbohydrate contents, and activities of some enzymes in *Vigna radiata*. Cell phone EMF radiations significantly reduced the seedling length and dry weight of *V. radiata* after exposure for 0.5, 1, 2, and 4 h. Furthermore, the contents of proteins and carbohydrates were reduced in EMF-exposed plants. However, the activities of proteases, alpha-amylases, beta-amylases, polyphenol oxidases, and peroxidases were enhanced in EMF-exposed radicles indicating their role in providing protection against EMF-induced stress. The study concludes that cell phone EMFs impair early growth of *V. radiata* seedlings by inducing biochemical changes.

**(E) Shehu A, Mohammed A, Magaji RA, Muhammad MS. Exposure to mobile phone electromagnetic field radiation, ringtone and vibration affects anxiety-like behaviour and oxidative stress biomarkers in albino wistar rats. Metab Brain Dis. 2015 Nov 7. [Epub ahead of print]**

Research on the effects of Mobile phone radio frequency emissions on biological systems has been focused on noise and vibrations as auditory stressors. This study investigated the potential effects of exposure to mobile phone electromagnetic field radiation, ringtone and vibration on anxiety-like behaviour and oxidative stress biomarkers in albino wistar rats. Twenty five male wistar rats were randomly divided into five groups of 5 animals each: group I: exposed to mobile phone in switched off mode (control), group II: exposed to mobile phone in silent mode, group III: exposed to mobile phone in vibration mode, group IV: exposed to mobile phone in ringtone mode, group V: exposed to mobile phone in vibration and ringtone mode. The animals in group II to V were exposed to 10 min call (30 missed calls for 20 s each) per day for 4 weeks. Neurobehavioural studies for assessing anxiety were carried out 24 h after the last exposure and the animals were sacrificed. Brain samples were collected for biochemical evaluation immediately. Results obtained showed a significant decrease ( $P < 0.05$ ) in open arm duration in all the experimental groups when compared to the control. A significant decrease ( $P < 0.05$ ) was also observed in catalase activity in group IV and V when compared to the control. In conclusion, the results of the present study indicates that 4 weeks exposure to electromagnetic radiation, vibration, ringtone or both produced a significant effect on anxiety-like behavior and oxidative stress in young wistar rats.

**(E) Shivashankara AR, Joy J, Sunitha V, Rai MP, Rao S, Nambranathayil S, Baliga MS. Effect of cell phone use on salivary total protein, enzymes and oxidative stress markers in young adults: a pilot study. J Clin Diagn Res. 9(2):BC19-22, 2015.**

INTRODUCTION: The present study aimed to assess the levels of salivary enzymes, protein and oxidant-antioxidant system in young college-going cell phone users. MATERIALS AND METHODS: The cell users (students) were categorized in to two groups - less mobile users and high mobile users, based on the duration and frequency of cell use. Unstimulated whole

saliva samples of the volunteers were analysed for amylase, lactate dehydrogenase (LDH), malondialdehyde (MDA) and glutathione (GSH). RESULTS: High mobile users had significantly higher levels of amylase ( $p = 0.001$ ), LDH ( $p = 0.002$ ) and MDA ( $p = 0.002$ ) in saliva, when compared to less mobile users. The marginal decrease in salivary total proteins, GSH and flow rate were statistically not significant ( $p > 0.05$ ). CONCLUSION: Significant changes in salivary enzymes and MDA suggest adverse effect of high use of cell phones on cell health.

**(E) Shokri M, Shamsaei ME, Malekshah AK, Amiri FT. The protective effect of melatonin on radiofrequency electromagnetic fields of mobile phone-induced testicular damage in an experimental mouse model. *Andrologia*. 2020 Oct 11;e13834. doi: 10.1111/and.13834.**

Radiofrequency electromagnetic radiation (RF-EMR) from mobile devices has undesirable effects on the male reproductive organs. Melatonin with antioxidant potential can help to prevent these damages. Therefore, the aim of this study was to evaluate the protective effect of melatonin on testicular damage induced by RF-EMR of mobile phone. In this experimental study, 32 adult male BALB/c mice were divided randomly into four groups: control, melatonin (2 mg/kg, for 30 consecutive days, intraperitoneally), RF-EMR (900 MHz, 100 to 300 MT, 54 to 160 W/m) (4 hr per day, whole body) and melatonin + RF-EMR groups. One day after the last prescription were evaluated oxidative stress parameters, testosterone level and histopathological assays of the testis. EMR of mobile phone led to the induction of oxidative stress, testicular tissue damage and decreased testosterone. Treatment with melatonin improved oxidative stress parameters such as MDA and GSH, and testis injury score, increased the thickness of the germinal epithelial thickness and diameter of the seminiferous tubule, and decreased testosterone hormone in the EMR-exposed mice, and these differences were significant ( $p < .05$ ). Data showed that melatonin with its antioxidant property can decrease oxidative damage induced by RF-EMR of mobile phones on testis tissue.

**(E) Sieroń K, Knapik K, Onik G, Romuk E, Birkner E, Kwiatek S, Sieroń A. Electromagnetic Fields Modify Redox Balance in the Rat Gastrointestinal Tract. *Front Public Health* 2021 Sep 13;9:710484.**

**Objective:** The aim of the study was to assess the influence of electromagnetic fields with divergent physical properties on the prooxidative and antioxidative balances in homogenates of the tongue, salivary glands, esophagus, stomach, and small and large intestines of rats. **Material and Methods:** Forty rats were randomly divided into four equal groups, namely, a control group, a group exposed to low-frequency electromagnetic fields (LF-EMFs; frequency: 50 Hz; intensity: 10 kV/m; magnetic induction: 4.3 pT), a group exposed to radiofrequency electromagnetic fields (RF-EMFs) emitted by mobile phones (frequency: 900 MHz), and a group exposed simultaneously to LF-EMFs and RF-EMFs emitted by mobile phones. After 28 consecutive days of the experiment, the following pro- and antioxidative markers were assessed in the gastrointestinal tract homogenates: superoxide dismutase (SOD) and its two isoenzymes (Mn-SOD, Cu,Zn-SOD) catalase (CAT), glutathione peroxidase (GPx), glutathione reductase (GR), glutathione S-transferase (GST), total antioxidative capacity (TAC), total oxidative status (TOS), and malondialdehyde (MDA). **Results:** In rats

exposed to LF-EMFs, higher concentrations of the markers of prooxidant processes, MDA or TOS, were observed in the salivary glands, esophagus, and small intestine homogenates in comparison with the control group. Additionally, in the group of rats opposite to the control, antioxidant activity was observed. The main differences included a higher activity of Cu,Zn-SOD in homogenates of the tongue, salivary glands, and esophagus as well as decreased activity of CAT in homogenates of the tongue, esophagus, and small intestine. In animals exposed to RF-EMFs, the concentration of TOS was higher in the large intestine than in control rats. The main difference of antioxidant activity was presented by decreased Cu,Zn-SOD in homogenates of the salivary glands, stomach, small and large intestine as well as CAT in homogenates of the tongue, esophagus, stomach, and small and large intestine. Moreover, in rats exposed simultaneously to LF-EMFs and RF-EMFs, a lower concentration of TOS was observed. Antioxidant activity was presented by a decreased activity of CAT in homogenates of the tongue, esophagus, stomach, and small and large intestine in comparison to the control group. **Conclusion:** Among those applied in the study, electromagnetic fields of a low-frequency caused the most significant disturbances of oxidative stress in the rat gastrointestinal tract.

**(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. 2015 Dec 21:1-9. [Epub ahead of print]**

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.

**(NE) Simkó M, Hartwig C, Lantow M, Lupke M, Mattsson MO, Rahman Q, Rollwitz J. Hsp70 expression and free radical release after exposure to non-thermal radio-frequency electromagnetic fields and ultrafine particles in human Mono Mac 6 cells. Toxicol Lett. (1):73-82, 2006.**

The contemporary urban environment has become increasingly complex in its composition, leading to discussions regarding possible novel health effects. Two factors that recently have received considerable attention are ultrafine particles (UFP; <0.1 microm) produced by combustion processes and emissions from wireless communication devices like mobile phones that emit in the radio-frequency (RF) part of the spectrum. Several studies have shown biological effects of both these exposures in various cell systems. Here we investigate if exposure to UFP (12-14 nm, 100 microg/ml) and RF-electromagnetic fields (EMF; 2 W/kg specific absorption rate (SAR); continuous wave (CW) or modulated (217Hz or GSM-nonDTX)), alone or in combination influences levels of the superoxide radical anion or the stress protein heat-shock protein (Hsp70) in the human monocyte cell line Mono Mac 6. Heat treatment (42-43 degrees C, 1h) was used as positive control for both stress reaction and for heat development in the RF exposure setup. Our results clearly show that Mono Mac 6 cells are capable to internalise UFP, and that this phagocytic activity is connected to an increased release of free radicals. This increase (40-45% above negative control) is stronger than the effect of heat treatment. On the other hand, none of the employed RF exposures showed any effects on free radical levels. Co-exposure of RF and UFP did not potentiate the UFP effect either. Our investigations showed a significantly increased Hsp70 expression level by heat treatment in a time-dependent manner, whereas UFP, RF, or UFP+RF were without any effect. Therefore, we conclude that in the investigated Mono Mac 6 cells, RF exposure alone or in combination with UFP cannot influence stress-related responses.

**(E) Singh KV, Gautam R, Meena R, Nirala JP, Jha SK, Rajamani P. Effect of mobile phone radiation on oxidative stress, inflammatory response, and contextual fear memory in Wistar rat. Environ Sci Pollut Res Int 2020 Jun;27(16):19340-19351.**

In the present lifestyle, we are continuously exposed to radiofrequency electromagnetic field (RF-EMF) radiation generated mainly by mobile phones (MP). Among other organs, our brain and hippocampus in specific, is the region where effect of any environmental perturbation is most pronounced. So, this study was aimed to examine changes in major parameters (oxidative stress, level of pro-inflammatory cytokines (PICs), hypothalamic-pituitary-adrenal (HPA) axis hormones, and contextual fear conditioning) which are linked to hippocampus directly or indirectly, upon exposure to mobile phone radiofrequency electromagnetic field (MP-RF-EMF) radiation. Exposure was performed on young adult male Wistar rats for 16 weeks continuously (2 h/day) with MP-RF-EMF radiation having frequency, power density, and specific absorption rate (SAR) of 1966.1 MHz, 4.0 mW/cm<sup>2</sup>, and 0.36 W/kg, respectively. Another set of animals kept in similar conditions without any radiation exposure serves as control. Towards the end of exposure period, animals were tested for fear memory and then euthanized to measure hippocampal oxidative stress, level of circulatory PICs, and stress hormones. We observed significant increase in hippocampal oxidative stress (p < 0.05) and elevated level of circulatory PICs viz. IL-1beta (p < 0.01), IL-6 (p < 0.05), and TNF-alpha (p < 0.001) in experimental animals upon exposure to MP-RF-EMF radiation. Adrenal gland weight (p < 0.001) and level of stress hormones viz. adrenocorticotrophic hormone (ACTH) (p < 0.01) and corticosterone (CORT) (p < 0.05) were also found to increase significantly in MP-RF-EMF radiation-exposed animals as compared with control. However, alteration in contextual fear memory was not significant enough. In conclusion, current study shows that chronic exposure to MP-RF-EMF radiation emitted from mobile phones may induce oxidative

stress, inflammatory response, and HPA axis deregulation. However, changes in hippocampal functionality depend on the complex interplay of several opposing factors that got affected upon MP-RF-EMF exposure.

**(E) Singh HP, Sharma VP, Batish DR, Kohli RK. Cell phone electromagnetic field radiations affect rhizogenesis through impairment of biochemical processes. Environ Monit Assess. 184(4):1813-1821, 2012.**

Indiscriminate adoption and use of cell phone technology has tremendously increased the levels of electromagnetic field radiations (EMFr) in the natural environment. It has raised the concerns among the scientists regarding the possible risks of EMFr to living organisms. However, not much has been done to assess the damage caused to plants that are continuously exposed to EMFr present in the environment. The present study investigated the biochemical mechanism of interference of 900 MHz cell phone EMFr with root formation in mung bean (*Vigna radiata* syn. *Phaseolus aureus*) hypocotyls, a model system to study rhizogenesis in plants. Cell phone EMFr enhanced the activities of proteases (by 1.52 to 2.33 times), polyphenol oxidases (by 1.5 to 4.3 times), and peroxidases (by 1.5 to 2.0 times) in mung bean hypocotyls over control. Further, EMFr enhanced malondialdehyde (an indicator of lipid peroxidation), hydrogen peroxide, and proline content, indicating a reactive oxygen species-mediated oxidative damage in hypocotyls. It was confirmed by the upregulation in the activities of antioxidant enzymes (superoxide dismutase, ascorbate peroxidase, guaiacol peroxidase, catalase, and glutathione reductase) suggesting their possible role in providing protection against EMFr-induced oxidative damage. The study concluded that cell phone radiations affect the process of rhizogenesis through biochemical alterations that manifest as oxidative damage resulting in root impairment.

**(E) Singh KV, Gautam R, Meena R, Nirala JP, Jha SK, Rajamani P. Effect of mobile phone radiation on oxidative stress, inflammatory response, and contextual fear memory in Wistar rat. Environ Sci Pollut Res Int. 2020 Mar 24. doi: 10.1007/s11356-020-07916-z.**

In the present lifestyle, we are continuously exposed to radiofrequency electromagnetic field (RF-EMF) radiation generated mainly by mobile phones (MP). Among other organs, our brain and hippocampus in specific, is the region where effect of any environmental perturbation is most pronounced. So, this study was aimed to examine changes in major parameters (oxidative stress, level of pro-inflammatory cytokines (PICs), hypothalamic-pituitary-adrenal (HPA) axis hormones, and contextual fear conditioning) which are linked to hippocampus directly or indirectly, upon exposure to mobile phone radiofrequency electromagnetic field (MP-RF-EMF) radiation. Exposure was performed on young adult male Wistar rats for 16 weeks continuously (2 h/day) with MP-RF-EMF radiation having frequency, power density, and specific absorption rate (SAR) of 1966.1 MHz, 4.0 mW/cm<sup>2</sup>, and 0.36 W/kg, respectively. Another set of animals kept in similar conditions without any radiation exposure serves as control. Towards the end of exposure period, animals were tested for fear memory and then euthanized to measure hippocampal oxidative stress, level of circulatory PICs, and stress

hormones. We observed significant increase in hippocampal oxidative stress ( $p < 0.05$ ) and elevated level of circulatory PICs viz. IL-1beta ( $p < 0.01$ ), IL-6 ( $p < 0.05$ ), and TNF-alpha ( $p < 0.001$ ) in experimental animals upon exposure to MP-RF-EMF radiation. Adrenal gland weight ( $p < 0.001$ ) and level of stress hormones viz. adrenocorticotrophic hormone (ACTH) ( $p < 0.01$ ) and corticosterone (CORT) ( $p < 0.05$ ) were also found to increase significantly in MP-RF-EMF radiation-exposed animals as compared with control. However, alteration in contextual fear memory was not significant enough. In conclusion, current study shows that chronic exposure to MP-RF-EMF radiation emitted from mobile phones may induce oxidative stress, inflammatory response, and HPA axis deregulation. However, changes in hippocampal functionality depend on the complex interplay of several opposing factors that got affected upon MP-RF-EMF exposure.

**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 Jan 24:1-40. (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". Manuscript 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  $\kappa$ B (NF- $\kappa$ B), and signal transducer and activator of transcription-3 (STAT-3) play 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) Sokolovic D, Djindjic B, Nikolic J, Bjelakovic G, Pavlovic D, Kocic G, Krstic D, Cvetkovic T, Pavlovic V. Melatonin reduces oxidative stress induced by chronic**

**exposure of microwave radiation from mobile phones in rat brain. J Radiat Res (Tokyo). 49(6):579-586, 2008.**

**PURPOSE:** The aim of the study was to evaluate the intensity of oxidative stress in the brain of animals chronically exposed to mobile phones and potential protective effects of melatonin in reducing oxidative stress and brain injury. **MATERIALS AND METHODS:** Experiments were performed on Wistar rats exposed to microwave radiation during 20, 40 and 60 days. Four groups were formed: I group (control)- animals treated by saline, intraperitoneally (i.p.) applied daily during follow up, II group (Mel)- rats treated daily with melatonin (2 mg kg(-1) body weight i.p.), III group (MWs)- microwave exposed rats, IV group (MWs + Mel)- MWs exposed rats treated with melatonin (2 mg kg(-1) body weight i.p.). The microwave radiation was produced by a mobile test phone (SAR = 0.043-0.135 W/kg). **RESULTS:** A significant increase in the brain tissue malondialdehyde (MDA) and carbonyl group concentration was registered during exposure. Decreased activity of catalase (CAT) and increased activity of xanthine oxidase (XO) remained after 40 and 60 days of exposure to mobile phones. Melatonin treatment significantly prevented the increase in the MDA content and XO activity in the brain tissue after 40 days of exposure while it was unable to prevent the decrease of CAT activity and increase of carbonyl group contents. **CONCLUSION:** We demonstrated two important findings; that mobile phones caused oxidative damage biochemically by increasing the levels of MDA, carbonyl groups, XO activity and decreasing CAT activity; and that treatment with the melatonin significantly prevented oxidative damage in the brain.

**(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.**

**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.

**(cancer) (E) Sun Y, Zong L, Gao Z, Zhu S, Tong J, Cao Y. Mitochondrial DNA damage and oxidative damage in HL-60 cells exposed to 900MHz radiofrequency fields. *Mutat Res.* 797-799:7-14, 2017.**

HL-60 cells, derived from human promyelocytic leukemia, were exposed to continuous wave 900MHz radiofrequency fields (RF) at 120 $\mu$ W/cm<sup>2</sup> 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) Tkalec M, Malaric K, Pevalek-Kozlina B. Influence of 400, 900, and 1900 MHz electromagnetic fields on Lemna minor growth and peroxidase activity. *Bioelectromagnetics.* 26(3):185-193, 2005.**

Increased use of radio and microwave frequencies requires investigations of their effects on living organisms. Duckweed (*Lemna minor* L.) has been commonly used as a model plant for environmental monitoring. In the present study, duckweed growth and peroxidase activity was evaluated after exposure in a Gigahertz Transversal Electromagnetic (GTEM) cell to electric fields of frequencies 400, 900, and 1900 MHz. The growth of plants exposed for 2 h to the 23 V/m electric field of 900 MHz significantly decreased in comparison with the control, while an electric field of the same strength but at 400 MHz did not have such effect. A modulated field at 900 MHz strongly inhibited the growth, while at 400 MHz modulation did not influence the growth significantly. At both frequencies a longer exposure mostly decreased the growth and the highest electric field (390 V/m) strongly inhibited the growth. Exposure of plants to lower field strength (10 V/m) for 14 h caused significant decrease at 400 and 1900 MHz while 900 MHz did not influence the growth. Peroxidase activity in exposed plants varied, depending on the exposure characteristics. Observed changes were mostly small, except in plants exposed for 2 h to 41 V/m at 900 MHz where a significant increase (41%) was found. Our results suggest that investigated electromagnetic fields (EMFs) might influence plant growth and, to some extent, peroxidase activity. However, the effects of EMFs strongly depended on the characteristics of the field exposure.

**(E) Tkalec M, Malarić K, Pevalek-Kozlina B. Exposure to radiofrequency radiation induces oxidative stress in duckweed *Lemna minor* L. *Sci Total Environ.* 388(1-3):78-89, 2007.**

Widespread use of radiofrequency radiation emitting devices increased the exposure to electromagnetic fields (EMFs) from 300 MHz to 300 GHz. Various biological effects of exposure to these fields have been documented so far, but very little work has been carried out on plants. The aim of the present work was to investigate the physiological responses of the plant *Lemna minor* after exposure to radiofrequency EMFs, and in particular, to clarify the possible role of oxidative stress in the observed effects. Duckweed was exposed for 2 h to EMFs of 400 and 900 MHz at field strengths of 10, 23, 41 and 120 V m<sup>-1</sup>. The effect of a longer exposure time (4 h) and modulation was also investigated. After exposure, parameters of oxidative stress, such as lipid peroxidation, H<sub>2</sub>O<sub>2</sub> content, activities and isoenzyme pattern of antioxidative enzymes as well as HSP70 expression were evaluated. At 400 MHz, lipid peroxidation and H<sub>2</sub>O<sub>2</sub> content were significantly enhanced in duckweed exposed to EMFs of 23 and 120 V m<sup>-1</sup> while other exposure treatments did not have an effect. Compared to the controls, the activities of antioxidative enzymes showed different behaviour: catalase (CAT) activity increased after most exposure treatments while pyrogallol (PPX) and ascorbate peroxidase (APX) activities were not changed. Exceptions were reduced PPX and APX activity after longer exposure at 23 V m<sup>-1</sup> and increased PPX activity after exposures at 10 and 120 V m<sup>-1</sup>. By contrast, at 900 MHz almost all exposure treatments significantly increased level of lipid peroxidation and H<sub>2</sub>O<sub>2</sub> content but mostly decreased PPX activity and did not affect CAT activity. Exceptions were exposures to a modulated field and to the field of 120 V m<sup>-1</sup> which increased PPX and CAT activity. At this frequency APX activity was significantly decreased after exposure at 10 V m<sup>-1</sup> and longer exposure at 23 V m<sup>-1</sup> but it increased after a shorter exposure at 23 V m<sup>-1</sup>. At both frequencies no differences in isoenzyme patterns of antioxidative enzymes or HSP70 level were found between control and exposed plants. Our results showed that non-thermal exposure to investigated radiofrequency fields induced oxidative stress in duckweed as well as unspecific stress responses, especially of antioxidative enzymes. However, the observed effects markedly depended on the field frequencies applied as well as on other exposure parameters (strength, modulation and exposure time). Enhanced lipid peroxidation and H<sub>2</sub>O<sub>2</sub> content accompanied by diminished antioxidative enzymes activity caused by exposure to investigated EMFs, especially at 900 MHz, indicate that oxidative stress could partly be due to changed activities of antioxidative enzymes.

**(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. (WS) (LI)**

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 (900 MHz). Earthworms were exposed to the homogeneous RF-EMF at field levels of 10, 23, 41 and 120 V m<sup>-1</sup> for a period of 2h using a Gigahertz Transversal

Electromagnetic (GTEM) cell. At the field level of 23 V m<sup>-1</sup> the effect of longer exposure (4h) and field modulation (80% AM 1 kHz 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 900 MHz 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 900 MHz electromagnetic radiation. Nature of detected DNA lesions and oxidative stress as the mechanism of action for the induction of DNA damage are discussed.

**(E) \*Tök L, Nazıroğlu M, Doğan S, Kahya MC, Tök O. Effects of melatonin on Wi-Fi-induced oxidative stress in lens of rats. Indian J Ophthalmol. 62(1):12-15, 2014. doi: 10.4103/0301-4738.126166.**

Introduction: Melatonin has been considered a potent antioxidant that detoxifies a variety of reactive oxygen species in many pathophysiological states of eye. The present study was designed to determine the effects of Wi-Fi exposure on the lens oxidant, antioxidant redox systems, as well as the possible protective effects of melatonin on the lens injury induced by electromagnetic radiation (EMR). Materials and Methods: Thirty-two rats were used in the current study and they were randomly divided into four equal groups as follows: First and second groups were cage-control and sham-control rats. Rats in third group were exposed to Wi-Fi (2.45 GHz) for duration of 60 min/day for 30 days. As in the third group, the fourth group was treated with melatonin. The one-hour exposure to irradiation in second, third and fourth took place at noon each day. Results: Lipid peroxidation levels in the lens were slightly higher in third (Wi-Fi) group than in cage and sham control groups although their concentrations were significantly ( $P < 0.05$ ) decreased by melatonin supplementation. Glutathione peroxidase (GSH-Px) activity was significantly ( $P < 0.05$ ) lower in Wi-Fi group than in cage and sham control groups although GSH-Px ( $P < 0.01$ ) and reduced glutathione ( $P < 0.05$ ) values were significantly higher in Wi-Fi + melatonin group than in Wi-Fi group. Conclusions: There are poor oxidative toxic effects of one hour of Wi-Fi exposure on the lens in the animals. However, melatonin supplementation in the lens seems to have protective effects on the oxidant system by modulation of GSH-Px activity.

**(E) 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(1):39-47, 2010.**

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(6) 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(6) 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) Topal Z, Hanci H, Mercantepe T, Erol HS, Keleş ON, Kaya H, Mungan S, Odaci E. The effects of prenatal long-duration exposure to 900-MHz electromagnetic field on the 21-day-old newborn male rat liver. Turk J Med Sci. 45(2):291-297, 2015.**

BACKGROUND/AIM: To determine what effect a 900-MHz electromagnetic field (EMF) applied in the prenatal period would have on the liver in the postnatal period. MATERIALS AND METHODS: At the start of the study, adult pregnant rats were divided into two groups, control and experimental. The experimental group was exposed to a 900-MHz EMF for 1 h daily during days 13-21 of pregnancy. After birth, no procedure was performed on either mothers or pups. Male rat pups ( $n = 6$ ) from the control group mothers (CGMR) and male rat pups ( $n = 6$ ) from the experimental group mothers (EGMR) were sacrificed on postnatal day 21. RESULTS: Biochemical analyses showed that malondialdehyde and superoxide dismutase values increased and glutathione levels decreased in the EGMR pups. Marked hydropic degeneration in the parenchyma, particularly in pericentral regions, was observed in light microscopic examination of EGMR sections stained with hematoxylin and eosin. Examinations under transmission electron microscope revealed vacuolization in the mitochondria, expansion in the endoplasmic reticulum, and necrotic hepatocytes. CONCLUSION: The study results show that a 900-MHz EMF applied in the prenatal period caused oxidative stress and pathological alterations in the liver in the postnatal period.

**(E) Tsoy A, Saliev T, Abzhanova E, Turgambayeva A, Kaiyrykzy A, Akishev M, Saparbayev S, Umbayev B, Askarova S. The Effects of Mobile Phone Radiofrequency Electromagnetic Fields on  $\beta$ -Amyloid-Induced Oxidative Stress in Human and Rat Primary Astrocytes. Neuroscience. 408:46-57, 2019.**

Amyloid beta peptide ( $A\beta$ ) is implicated in the development of pathological reactions associated with Alzheimer's disease (AD), such as oxidative stress, neuro-inflammation and death of brain cells. Current pharmacological approaches to treat AD are not able to control the deposition of  $A\beta$  and suppression of  $A\beta$ -induced cellular response. There is a growing body of evidence that exposure to radiofrequency electromagnetic field (RF-EMF) causes a decrease of beta-amyloid deposition in the brains and provides cognitive benefits to Alzheimer's Tg mice. Herein, we investigated the effects of mobile phone radiofrequency EMF of 918 MHz

on reactive oxygen species (ROS) formation, mitochondrial membrane potential (MMP), activity of NADPH-oxidase, and phosphorylation of p38MAPK and ERK1/2 kinases in human and rat primary astrocytes in the presence of A $\beta$ <sub>42</sub> and H<sub>2</sub>O<sub>2</sub>. Our data demonstrate that EMF is able to reduce A $\beta$ <sub>42</sub>- and H<sub>2</sub>O<sub>2</sub>-induced cellular ROS, abrogate A $\beta$ <sub>42</sub>-induced production of mitochondrial ROS and the co-localization between the cytosolic (p47-phox) and membrane (gp91-phox) subunits of NADPH oxidase, while increasing MMP, and inhibiting H<sub>2</sub>O<sub>2</sub>-induced phosphorylation of p38MAPK and ERK1/2 in primary astrocytes. Yet, EMF was not able to modulate alterations in the phosphorylation state of the MAPKs triggered by A $\beta$ <sub>42</sub>. Our findings provide an insight into the mechanisms of cellular and molecular responses of astrocytes on RF-EMF exposure and indicate the therapeutic potential of RF-EMF for the treatment of Alzheimer's disease.

**(E) Tsoy A, Saliev T, Abzhanova E, Turgambayeva A, Kaiyrlykyzy A, Akishev M, Tumkaya L, Yilmaz A, Akyildiz K, Mercantepe T, Yazici ZA, Yilmaz H. Prenatal Effects of a 1,800-MHz Electromagnetic Field on Rat Livers. Cells Tissues Organs. 2019 Dec 4:1-9, 2019.**

The use of devices, including mobile phones, generating electromagnetic fields (EMF) is widespread and is progressively increasing. It has also been shown that EMF may have detrimental effects. This is the first study to investigate the postnatal biochemical and histological effects of prenatal exposure of rat livers to 1,800-MHz EMF at different time intervals in uteroplacental life. The 3 EMF groups of rats were exposed to 1,800-MHz EMF for 6, 12, or 24 h daily for 20 days. Unexposed rats served as control group. All rats were subjected to anesthesia, and on postnatal day 60, the livers were excised, and blood was collected for histological and biochemical analyses. Malondialdehyde levels were significantly higher in the exposed groups than the unexposed controls ( $p < 0.05$ ). In contrast, EMF-exposed groups had lower liver tissue glutathione levels than controls ( $p < 0.05$ ). Serum Ca<sup>2+</sup>, alanine transaminase, and aspartate aminotransferase levels were higher in EMF-exposed groups than controls ( $p < 0.05$ ). In addition, liver tissue total oxidant status levels were increased ( $p < 0.05$ ), and liver tissue total antioxidant status levels were decreased ( $p < 0.05$ ) compared to the control group. Furthermore, in the EMF groups, extensive vacuolation and degeneration of the hepatocytes in the portal area, as well as those surrounding the sinusoids, were evident. Affected hepatocytes had polygonally shaped nuclei and vacuolic cytoplasm imparting eosinophilic staining. Loss of cellular membrane integrity and invaginations, as well as picnotic nuclei, was prominent. This study has shown that intrauterine liver damage caused by 1,800-MHz EMF exposure persists into puberty in rats.

**(E) Tsybulin O, Sidorik E, Kyrylenko S, Henshel D, Yakymenko I. GSM 900 MHz microwave radiation affects embryo development of Japanese quails. Electromagn Biol Med. 2012 Mar;31(1):75-86.**

A wide range of non thermal biological effects of microwave radiation (MW) was revealed during the last decades. A number of reports showed evident hazardous effects of MW on

embryo development in chicken. In this study, we aimed at elucidating the effects of MW emitted by a commercial model of GSM 900 MHz cell phone on embryo development in quails (*Coturnix coturnix japonica*) during both short and prolonged exposure. For that, fresh fertilized eggs were irradiated during the first 38 h or 14 days of incubation by a cell phone in "connecting" mode activated continuously through a computer system. Maximum intensity of incident radiation on the egg's surface was 0.2  $\mu\text{W}/\text{cm}^2$ . The irradiation led to a significant ( $p < 0.001$ ) increase in numbers of differentiated somites in 38-hour exposed embryos and to a significant ( $p < 0.05$ ) increase in total survival of embryos from exposed eggs after 14 days exposure. We hypothesized that observed facilitating effect was due to enhancement of metabolism in exposed embryos provoked via peroxidation mechanisms. Indeed, a level of thiobarbituric acid (TBA) reactive substances was significantly ( $p < 0.05-0.001$ ) higher in brains and livers of hatchlings from exposed embryos. Thus, observed effects of radiation from commercial GSM 900 MHz cell phone on developing quail embryos signify a possibility for non-thermal impact of MW on embryogenesis. We suggest that the facilitating effect of low doses of irradiation on embryo development can be explained by a hormesis effect induced by reactive oxygen species (ROS). Future studies need to be done to clarify this assumption.

**(E) Türedi S, Hancı H, Topal Z, Unal D, Mercantepe T, Bozkurt I, Kaya H, Odacı E. The effects of prenatal exposure to a 900-MHz electromagnetic field on the 21-day-old male rat heart. Electromagn Biol Med. 2014 Aug 28:1-8. [Epub ahead of print]**

Abstract The growing spread of mobile phone use is raising concerns about the effect on human health of the electromagnetic field (EMF) these devices emit. The purpose of this study was to investigate the effects on rat pup heart tissue of prenatal exposure to a 900 megahertz (MHz) EMF. For this purpose, pregnant rats were divided into experimental and control groups. Experimental group rats were exposed to a 900 MHz EMF (1 h/d) on days 13-21 of pregnancy. Measurements were performed with rats inside the exposure box in order to determine the distribution of EMF intensity. Our measurements showed that pregnant experimental group rats were exposed to a mean electrical field intensity of 13.77 V/m inside the box (0.50 W/m<sup>2</sup>). This study continued with male rat pups obtained from both groups. Pups were sacrificed on postnatal day 21, and the heart tissues were extracted. Malondialdehyde, superoxide dismutase and catalase values were significantly higher in the experimental group rats, while glutathione values were lower. Light microscopy revealed irregularities in heart muscle fibers and apoptotic changes in the experimental group. Electron microscopy revealed crista loss and swelling in the mitochondria, degeneration in myofibrils and structural impairments in Z bands. Our study results suggest that exposure to EMF in the prenatal period causes oxidative stress and histopathological changes in male rat pup heart tissue.

**(E) Türedi S, Kerimoğlu G, Mercantepe T, Odacı E. Biochemical and pathological changes in the male rat kidney and bladder following exposure to continuous 900-MHz electromagnetic field on postnatal days 22-59. Int J Radiat Biol. 93(9):990-999, 2017.**

**PURPOSE:** To investigate the effect on male rat kidney and bladder tissues of exposure to 900-megahertz (MHz) electromagnetic field (EMF) applied on postnatal days 22-59, inclusive. **MATERIALS AND METHODS:** Twenty-four male Sprague Dawley rats, aged 21 days, were used. These were divided equally into one of three groups, control (CG), sham (SG) or EMF (EMFG). CG was not exposed to any procedure. SG rats were kept inside a cage, without being exposed to the effect of EMF, for 1 h a day on postnatal days 22-59, inclusive. EMFG rats were exposed to continuous 900-MHz EMF for 1 h a day under the same conditions as those for the SG rats. Rats were sacrificed on postnatal day 60, and the kidney and bladder tissues were removed. Tissues were stained with hematoxylin and eosin (H&E) and Masson trichrome for histomorphological evaluation. The TUNEL method was used to assess apoptosis. Transmission electron microscopy (TEM) was also used for the kidney tissue. Oxidant/antioxidant parameters were studied in terms of biochemical values. **RESULTS:** The findings showed that tissue malondialdehyde increased in EMFG compared to CG and SG in both kidney ( $p = 0.004$  and  $p = 0.004$ , respectively) and bladder tissue ( $p = 0.004$ ,  $p = 0.006$ , respectively), while catalase and glutathione levels decreased compared to CG ( $p = 0.004$ ;  $p = 0.004$ , respectively) and SG ( $p = 0.004$ ;  $p = 0.004$ , respectively). In the EMF group, pathologies such as dilatation and vacuolization in the distal and proximal tubules, degeneration in glomeruli and an increase in cells tending to apoptosis were observed in kidney tissue. In bladder tissue, degeneration in the transitional epithelium and stromal irregularity and an increase in cells tending to apoptosis were observed in EMFG. Additionally, EMFG samples exhibited glomerular capillary degeneration with capillary basement membranes under TEM. **CONCLUSIONS:** We conclude that continuous exposure to the effect of 900-MHz EMF for 1 h a day on postnatal days 22-59, inclusive, causes an increase in oxidative stress and various pathological changes in male rat kidney and bladder tissues.

**(E) Türker Y, Nazıroğlu M, Gümral N, Celik O, Saygın M, Cömlekçi S, Flores-Arce M. Selenium and L-carnitine reduce oxidative stress in the heart of rat induced by 2.45-GHz radiation from wireless devices. Biol Trace Elem Res. 143(3):1640-1650, 2011.**

The aim of this study was to investigate the possible protective role of selenium and L-carnitine on oxidative stress induced by 2.45-GHz radiation in heart of rat. For this purpose, 30 male Wistar Albino rats were equally divided into five groups namely controls, sham controls, radiation-exposed rats, radiation-exposed rats treated with intraperitoneal injections of sodium selenite at a dose of 1.5 mg/kg/day, and radiation-exposed rats treated with intraperitoneal injections of L-carnitine at a dose of 1.5 mg/kg/day. Except for the controls and sham controls, the animals were exposed to 2.45-GHz radiation during 60 min/day for 28 days. The lipid peroxidation (LP) levels were higher in the radiation-exposed groups than in the control and sham control groups. The lipid peroxidation level in the irradiated animals treated with selenium and L-carnitine was lower than in those that were only exposed to 2.45-GHz radiation. The concentrations of vitamins A, C, and E were lower in the irradiated-only group relative to control and sham control groups, but their concentrations were increased in the groups treated with selenium- and L-carnitine. The activity of glutathione peroxidase was

higher in the selenium-treated group than in the animals that were irradiated but received no treatment. The erythrocyte-reduced glutathione and  $\beta$ -carotene concentrations did not change in any of the groups. In conclusion, 2.45-GHz electromagnetic radiation caused oxidative stress in the heart of rats. There is an apparent protective effect of selenium and L-carnitine by inhibition of free radical formation and support of the antioxidant redox system.

**(E) Ulubay M, Yahyazadeh A, Deniz OG, Kıvrak EG, Altunkaynak BZ, Erdem G, Kaplan S. Effects of prenatal 900 MHz electromagnetic field exposures on the histology of rat kidney. Int J Radiat Biol. 91(1):35-41, 2015.**

Purpose: To research the harmful effects of prenatal exposure of 900 megahertz (MHz) electromagnetic field (EMF) on kidneys of four-week-old male rats and to determine protective effects of melatonin (MEL) and omega-3 ( $\omega$ -3). Materials and methods: Twenty-one Wistar albino rats were randomly placed into seven groups as follows: control (Cont), Sham, MEL,  $\omega$ -3, EMF, EMF+MEL and EMF+ $\omega$ -3. After mating, three groups (EMF, EMF+MEL, EMF+  $\omega$ -3) were exposed to an EMF. In the fourth week subsequent to parturition, six rats were randomly chosen from each group. Mean volume of kidneys and renal cortices, the total number of glomeruli and basic histological structure of kidney were evaluated by stereological and light microscopical methods, respectively. Results: Stereological results determined the mean volume of the kidneys and cortices were significantly increased in EMF-exposed groups compared to the Cont group. However, EMF-unexposed groups were not significantly modified compared to the Cont group. Additionally, the total number of glomeruli was significantly higher in EMF-unexposed groups compared to the Cont group. Alternatively, the number of glomeruli in EMF-exposed groups was decreased compared to the Cont group. Conclusions: Prenatal exposure of rat kidneys to 900 MHz EMF resulted in increased total kidney volume and decreased the numbers of glomeruli. Moreover, MEL and  $\omega$ -3 prevented adverse effects of EMF on the kidneys.

**(E) Upadhyaya C, Upadhyaya T, Patel I. Attributes of non-ionizing radiation of 1800 MHz frequency on plant health and antioxidant content of Tomato (Solanum Lycopersicum) plants. J Rad Res Appl Sci. 15(1):54-68. 2022.**

The Globe is marching towards the development of the telecommunication field which leads to increment of non-ionizing radiation in the environment which affects all living beings including plants grown nearby to communication base stations. The present research was focused on physiological and biochemical alterations of tomato plants exposed to high-frequency radiation. The overall plant health was analyzed by physiological changes viz., plant height, size of leaves, length to the root system, and rate of germination upon exposure of radiation and shown significant reduction ( $p < 0.05$ ) compared to control. Consecutively, radiation also negatively affects the photosynthetic pigment content of leaves which has shown significant reduction. Yet another confirmation of stress on exposed plant tissue was reported by obtaining higher H<sub>2</sub>O<sub>2</sub> content within exposed plant leaf than the control. The morphological alterations viz. curling of leaves, discoloration, and size reduction became more prominent with an increase in the exposure time. The significant outcomes denoted according to 95% confidence limit. There was a significant decline in total phenolic content (37.06%), flavonoid content (71.38%), Vitamin C content (72.45%), and DPPH (59.32%) as

well as total antioxidant assays (71.89%) which revealed significant deteriorative effects on such waves on secondary metabolites and the antioxidant potential of tomato plants. The lycopene content was continuously increasing up to 73.13% upon radiation of 120 h and such raise was the direct indication of harmful effect on fruit skin and release of lycopene due to softening of the fruit tissue. Thus, the presented findings illustrated the negative effects of such waves on the quality of tomato plants. The limited insight of metabolic pathways involved in plant responses to such non-ionizing radiation makes such investigation worth in agricultural application. Additionally, mobile communication agencies should be informed and installation of base stations for mobile communication towers should be prohibited at agricultural lands.

**(E) Varghese R, Majumdar A, Kumar G, Shukla A. Rats exposed to 2.45GHz of non-ionizing radiation exhibit behavioral changes with increased brain expression of apoptotic caspase 3. Pathophysiology. 25:19-30, 2018.**

In recent years there has been a tremendous increase in use of Wi-Fi devices along with mobile phones, globally. Wi-Fi devices make use of 2.4 GHz frequency. The present study evaluated the impact of 2.45 GHz radiation exposure for 4h/day for 45 days on behavioral and oxidative stress parameters in female Sprague Dawley rats. Behavioral tests of anxiety, learning and memory were started from day 38. Oxidative stress parameters were estimated in brain homogenates after sacrificing the rats on day 45. In morris water maze, elevated plus maze and light dark box test, the 2.45 GHz radiation exposed rats elicited memory decline and anxiety behavior. Exposure decreased activities of super oxide dismutase, catalase and reduced glutathione levels whereas increased levels of brain lipid peroxidation was encountered in the radiation exposed rats, showing compromised anti-oxidant defense. Expression of caspase 3 gene in brain samples were quantified which unraveled notable increase in the apoptotic marker caspase 3 in 2.45 GHz radiation exposed group as compared to sham exposed group. No significant changes were observed in histopathological examinations and brain levels of TNF- $\alpha$ . Analysis of dendritic arborization of neurons showcased reduction in number of dendritic branching and intersections which corresponds to alteration in dendritic structure of neurons, affecting neuronal signaling. The study clearly indicates that exposure of rats to microwave radiation of 2.45GHz leads to detrimental changes in brain leading to lowering of learning and memory and expression of anxiety behavior in rats along with fall in brain antioxidant enzyme systems.

**(E) Verma S, Keshri GK, Karmakar S, Mani KV, Chauhan S, Yadav A, Sharma M, Gupta A. Effects of Microwave 10 GHz Radiation Exposure in the Skin of Rats: An Insight on Molecular Responses. Radiat Res. 196:404-416, 2021.**

Microwave (MW) radiation poses the risk of potential hazards on human health. The present study investigated the effects of MW 10 GHz exposure for 3 h/day for 30 days at power densities of  $5.23 \pm 0.25$  and  $10.01 \pm 0.15$  mW/cm<sup>2</sup> in the skin of rats. The animals exposed to 10 mW/cm<sup>2</sup> (corresponded to twice the ICNIRP-2020 occupational reference level of MW exposure for humans) exhibited significant biophysical, biochemical, molecular and histological alterations compared to sham-irradiated animals. Infrared thermography revealed an increase in average skin surface temperature by 1.8°C and standard deviation of 0.3°C after 30 days of 10 mW/cm<sup>2</sup> MW exposure compared to the sham-irradiated animals. MW

exposure also led to oxidative stress (ROS, 4-HNE, LPO, AOPP), inflammatory responses (NFkB, iNOS/NOS2, COX-2) and metabolic alterations [hexokinase (HK), lactate dehydrogenase (LDH), citrate synthase (CS) and glucose-6-phosphate dehydrogenase (G6PD)] in 10 mW/cm<sup>2</sup> irradiated rat skin. A significant alteration in expression of markers associated with cell survival (Akt/PKB) and HSP27/p38MAPK-related stress-response signaling cascade was observed in 10 mW/cm<sup>2</sup> irradiated rat skin compared to sham-irradiated rat skin. However, MW-irradiated groups did not show apoptosis, evident by unchanged caspase-3 levels. Histopathological analysis revealed a mild cytoarchitectural alteration in epidermal layer and slight aggregation of leukocytes in 10 mW/cm<sup>2</sup> irradiated rat skin. Altogether, the present findings demonstrated that 10 GHz exposure in continuous-wave mode at 10 mW/cm<sup>2</sup> (3 h/day, 30 days) led to significant alterations in molecular markers associated with adaptive stress-response in rat skin. Furthermore, systematic scientific studies on more prevalent pulsed-mode of MW-radiation exposure for prolonged duration are warranted.

**(E) Vilić M, Gajger I, Tucak P, Štambuk A, Šrut M, Klobučar G. Effects of short-term exposure to mobile phone radiofrequency (900 MHz) on the oxidative response and genotoxicity in honey bee larvae. J. Api. Res.56:430-438 (2017)**

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<sup>-1</sup> for 2 h. At a field level of 23 V m<sup>-1</sup> 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<sup>-1</sup> 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<sup>-1</sup> 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.

**(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.**

**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 2022 Feb 1;294:118646.**

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<sup>2</sup>, 1 W/m<sup>2</sup> and 10 W/m<sup>2</sup>. 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.

**(E) Wen X, Rui X, Caiyun F, Chunyu Y, Haiyan C, Yi C. 900 MHz Radiofrequency Field Induces Mitochondrial Unfolded Protein Response in Mouse Bone Marrow Stem Cells. Frontiers in Public Health. 9:1265. 2021. doi: 10.3389/fpubh.2021.724239**

Objective: To examine whether exposure of mouse bone marrow stromal cells (BMSC) to 900 MHz radiofrequency fields used in mobile communication devices can induce mitochondrial unfolded protein response (UPR) Methods: BMSCs were exposed to continuous wave 900 MHz radiofrequency fields (RF) at 120  $\mu\text{W}/\text{cm}^2$  power intensity for 4 h/d for 5 consecutive days. Cells in sham group (SH) were cultured in RF exposure system, but without RF radiation. The positive control cells were irradiated with 6 Gy X-ray at a dose rate of 1.103 Gy/min (XR). To inhibit the upstream molecular JNK2 of UPR, cells in siRNA + RF, and siRNA + XR group were also pretreated with 100 nM siRNA-JNK2 for 48 h before RF/XR exposure. Thirty minutes, 4 h, and 24 h post-RF/XR exposure, cells were collected, the level of ROS was measured with flow cytometry, the expression levels of UPR-related proteins were detected using western blot analysis. Results: Compared with Sham group, the level of ROS in RF and XR group was significantly increased 30 min and 4 h post-RF/XR exposure ( $P < 0.05$ ), however, the RF/XR-induced increase of ROS level reversed 24 h post-RF/XR exposure. Compared with Sham group, the expression levels of HSP10/HSP60/ClpP proteins in cells of RF and XR group increased significantly 30 min and 4 h post-RF/XR exposure ( $P < 0.05$ ), however, the RF/XR-induced increase of HSP10/HSP60/ClpP protein levels reversed 24 h post-RF exposure. After interfering with siRNA-JNK2, the RF/XR exposures could not induce the increase of HSP10/HSP60/ClpP protein levels any more. Conclusions: The exposure of 900 MHz RF at 120  $\mu\text{W}/\text{cm}^2$  power flux density could increase ROS level and activate a transient UPR in BMSC cells. Mitochondrial homeostasis in term of protein folding ability is restored 24 h post-RF exposure. Exposure to RF in our experimental condition did not cause permanent and severe mitochondrial dysfunctions. However, the detailed underlying molecular mechanism of RF-induced UPR remains to be further studied.

**(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]. [Article in Chinese] Zhejiang Da Xue Xue Bao Yi Xue Ban. 37(1):34-38, 2008.**

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 2  $\mu\text{T}$  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) Xie W, Xu R, Fan C, Yang C, Chen H, Cao Y. 900 MHz Radiofrequency Field Induces Mitochondrial Unfolded Protein Response in Mouse Bone Marrow Stem Cells. Front Public Health 9:724239, 2021.**

**Objective:** To examine whether exposure of mouse bone marrow stromal cells (BMSC) to 900 MHz radiofrequency fields used in mobile communication devices can induce mitochondrial unfolded protein response (UPR<sup>mt</sup>). **Methods:** BMSCs were exposed to continuous wave 900 MHz radiofrequency fields (RF) at 120  $\mu\text{W}/\text{cm}^2$  power intensity for 4 h/d for 5 consecutive days. Cells in sham group (SH) were cultured in RF exposure system, but without RF radiation. The positive control cells were irradiated with 6 Gy X-ray at a dose rate of 1.103 Gy/min (XR). To inhibit the upstream molecular JNK2 of UPR<sup>mt</sup>, cells in siRNA + RF, and siRNA + XR group were also pretreated with 100 nM siRNA-JNK2 for 48 h before RF/XR exposure. Thirty minutes, 4 h, and 24 h post-RF/XR exposure, cells were collected, the level of ROS was measured with flow cytometry, the expression levels of UPR<sup>mt</sup>-related proteins were detected using western blot analysis. **Results:** Compared with Sham group, the level of ROS in RF and XR group was significantly increased 30 min and 4 h post-RF/XR exposure ( $P < 0.05$ ), however, the RF/XR-induced increase of ROS level reversed 24 h post-RF/XR exposure. Compared with Sham group, the expression levels of HSP10/HSP60/ClpP proteins in cells of RF and XR group increased significantly 30 min and 4 h post-RF/XR exposure ( $P < 0.05$ ), however, the RF/XR-induced increase of HSP10/HSP60/ClpP protein levels reversed 24 h post-RF exposure. After interfering with siRNA-JNK2, the RF/XR exposures could not induce the increase of HSP10/HSP60/ClpP protein levels any more. **Conclusions:** The exposure of 900 MHz RF at 120  $\mu\text{W}/\text{cm}^2$  power flux density could increase ROS level and activate a transient UPR<sup>mt</sup> in BMSC cells. Mitochondrial homeostasis in term of protein folding ability is restored 24 h post-RF exposure. Exposure to RF in our experimental condition did not cause permanent and severe mitochondrial dysfunctions. However, the detailed underlying molecular mechanism of RF-induced UPR<sup>mt</sup> remains to be further studied.

**(E) Xu S, Zhou Z, Zhang L, Yu Z, Zhang W, Wang Y, Wang X, Li M, Chen Y, Chen C, He M, Zhang G, Zhong M. Exposure to 1800 MHz radiofrequency radiation induces oxidative damage to mitochondrial DNA in primary cultured neurons. Brain Res. 1311:189-196, 2010.**

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) Yahyazadeh A, Altunkaynak BZ. Investigation of the neuroprotective effects of thymoquinone on rat spinal cord exposed to 900 MHz electromagnetic field. J Chem Neuroanat. 2019 Oct;100:101657.**

Exposure to electromagnetic field in long-term use of cell phones has increased concerns about serious health problems. Our aim was to survey the possible effects of electromagnetic field radiation (60 min/day for 28 days) on the spinal cords of 12 weeks old rats. Further, we investigated whether the administration of thymoquinone (10 mg/kg/day) would protect the spinal cord tissue against the adverse effects of electromagnetic field or not. Twenty-four adult male Wistar albino rats were assigned randomly into four groups: control, electromagnetic field, thymoquinone and electromagnetic field + thymoquinone. The cervical spinal cords of all rats was evaluated using the stereological, biochemical and histological methods. The number of motor neurons were reduced in the electromagnetic field group compared to the control group ( $p < 0.05$ ). Superoxide dismutase level was higher in the electromagnetic field group compared to the control group ( $p < 0.05$ ). In the electromagnetic field + thymoquinone group, we found an increase in the number of motor neurons and decrease in superoxide dismutase activity compared to the electromagnetic field group ( $p < 0.05$ ). Our histological findings also exhibit the remarkable architectural alterations. We speculated that electromagnetic field radiation induced the morphological and biochemical damage to the spinal cord tissue of rats. Administration of antioxidant, thymoquinone, also ameliorated such complications caused by electromagnetic field.

**(E) Yahyazadeh A, Altunkaynak BZ, Kaplan S. Biochemical, immunohistochemical and morphometrical investigation of the effect of thymoquinone on the rat testis following exposure to a 900-MHz electromagnetic field. Acta Histochem. 2020 Jan;122(1):151467.**

Long-term use of cell phones emitting electromagnetic fields (EMFs) have raised concerns regarding public health in recent year. We aimed to investigate the possible effects of 900 MHz EMF exposure (60 min/day for 28 days) on the rat testis. Another objective was to determine whether the deleterious effect of EMF radiation would be reduced by the administration of thymoquinone (TQ) (10 mg/kg/day). Twenty-four male adult Wistar albino rats were randomly selected, then assigned into four groups as follow Control, EMF, TQ and EMF + TQ. Testicular samples were analyzed using histological, stereological, biochemical and immunohistochemical techniques. Total numbers of primary spermatocytes and spermatids as well as Leydig cells were significantly decreased in the EMF group compared to the Control group ( $p < 0.05$ ). In the EMF + TQ group, the total number of primary spermatocytes was significantly increased compared to the EMF group ( $p < 0.05$ ). Superoxide dismutase (SOD) activity was significantly increased in the EMF group compared to the Control group ( $p < 0.05$ ). Also, serum testosterone levels and wet weight of testes were significantly decreased in the EMF group compared to the Control group ( $p < 0.05$ ). Our findings suggested that exposure to a 900 MHz EMF had adverse effects on rat testicular tissue and that the administration of TQ partially mitigated testicular oxidative damages caused by EMF radiation.

**(Review) Yakymenko I, Tsybulin O, Sidorik E, Henshel D, Kyrylenko O, Kyrylenko S. Oxidative mechanisms of biological activity of low-intensity radiofrequency radiation. Electromagn Biol Med. 35(2):186-202, 2016.**

This review aims to cover experimental data on oxidative effects of low-intensity radiofrequency radiation (RFR) in living cells. Analysis of the currently available peer-reviewed scientific literature reveals molecular effects induced by low-intensity RFR in living cells; this includes significant activation of key pathways generating reactive oxygen species (ROS), activation of peroxidation, oxidative damage of DNA and changes in the activity of antioxidant enzymes. It indicates that among 100 currently available peer-reviewed studies dealing with oxidative effects of low-intensity RFR, in general, 93 confirmed that RFR induces oxidative effects in biological systems. A wide pathogenic potential of the induced ROS and their involvement in cell signaling pathways explains a range of biological/health effects of low-intensity RFR, which include both cancer and non-cancer pathologies. In conclusion, our analysis demonstrates that low-intensity RFR is an expressive oxidative agent for living cells with a high pathogenic potential and that the oxidative stress induced by RFR exposure should be recognized as one of the primary mechanisms of the biological activity of this kind of radiation.

**(E) Yakymenko I, Burlaka A, Tsybulin I, Bricieva 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.**

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  $\mu\text{W}/\text{cm}^2$ , 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) Yang H, Zhang Y, Wu X, Gan P, Luo X, Zhong S, Zuo W. Effects of Acute Exposure to 3500 MHz (5G) Radiofrequency Electromagnetic Radiation on Anxiety-**

**Like Behavior and the Auditory Cortex in Guinea Pigs. *Bioelectromagnetics* 43(2):106-118, 2022.**

Numerous studies have shown that radiofrequency electromagnetic radiation (RF-EMR) may negatively affect human health. We detected the effect of 3500 MHz RF-EMR on anxiety-like behavior and the auditory cortex (ACx) in guinea pigs. Forty male guinea pigs were randomly divided into four groups and exposed to a continuous wave of 3500 MHz RF-EMF at an average specific absorption rate (SAR) of 0, 2, 4, or 10 W/kg for 72 h. After exposure, malondialdehyde (MDA) levels, antioxidant enzyme activity, anxiety-like behavior, hearing thresholds, cell ultrastructure, and apoptosis were detected. Our results revealed that hearing thresholds and basic indexes of animal behavior did not change significantly after exposure ( $P > 0.05$ ). However, the MDA levels of ACx were increased ( $P < 0.05$ ), and catalase (CAT), superoxide dismutase (SOD), and glutathione peroxidase (GSH-px) activities were decreased ( $P < 0.05$ ) in the exposure groups compared to the sham group. Ultrastructural changes of ACx, including swollen mitochondria and layered myelin sheaths, were observed. Cytochrome-c relocalization, caspase-9, and cleaved caspase-3 activation were detected in the exposure groups. In conclusion, these results suggest that oxidative stress is an important mechanism underlying the biological effects of RF-EMR, which can induce ultrastructural damage to the ACx and cell apoptosis through a mitochondria-dependent mechanism. Moreover, oxidative stress, apoptosis induction and ultrastructural damage increase in a SAR-dependent manner. However, RF-EMR does not increase hearing thresholds or induce anxiety.

**(E) Yang R, Chen J, Deng Z, Liu X, [Effect of vitamin E on morphological variation of retinal ganglion cells after microwave radiation]. Wei Sheng Yan Jiu 30(1):31-33, 2001. [Article in Chinese]**

To determine the morphological variation in the primary cultured pig retinal ganglion cells induced by microwave and the protection of VE can supply some experiment foundation for study of effect of microwave and its protection. Retinal ganglion cells of pig were cultured in vitro and added VE of different concentration, Each group was taken after 30 mW/cm<sup>2</sup> microwave intensity radiated for 1 h in shielded room by 2450 MHz continuous wave physiotherapy machine. Immediately after radiation, the morphological variation of cells was observed by optics microscope and transmission electronic microscope. The result showed that a portion of cells congregated, with their axon disappeared after radiation. Mitochondria and endoplasmic reticulum are detected swelling by transmission electronic microscope. The results showed that A popnosis cells can be observed. Cells of VE added groups had not obvious changes with optics microscope, but could be found that mitochondria swelling lightly and integrate mitochondria cristae by transmission electronic microscope. The results showed that microwave induced the morphological damage in primary cultured retinal ganglion cells, VE could reduced the damage of retina ganglion cells by microwave in some extent.

**(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.**

**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) Yarıktas M, Doner F, Ozguner F, Gokalp O, Dogru H, Delibas N. Nitric oxide level in the nasal and sinus mucosa after exposure to electromagnetic field. Otolaryngol Head Neck Surg. 132(5):713-716, 2005.**

**OBJECTIVE:** The purpose of this study was to examine the changes in nitric oxide (NO) level in the nasal and paranasal sinus mucosa after exposure radiofrequency electromagnetic fields (EMF). **STUDY DESIGN AND SETTING:** Thirty male Sprague-Dawley rats were randomly grouped as follows: EMF group (group I; n, 10), EMF group in which melatonin received (group II; n, 10) and the control (sham operated) group (group III; n, 10). Groups I and II were exposed to a 900 MHz. Oral melatonin was given in group II. Control rats (group III) were also placed in the tube as the exposure groups, but without exposure to EMF. At the end of 2 weeks, the rats were sacrificed, and the nasal and paranasal sinus mucosa dissected. NO was measured in nasal and paranasal mucosa. **RESULTS:** The nasal and paranasal sinus mucosa NO levels of group I were significantly higher than those of the control group (group III) ( $P < 0.05$ ). However, there was no statistically significant difference between group II and the control group (group III) regarding NO output ( $P > 0.05$ ). **CONCLUSION:** Exposure to EMF released by mobile phones (900 MHz) increase NO levels in the sinus and nasal mucosa. **SIGNIFICANCE:** Increased NO levels may act as a defense mechanism and presumably related to tissue damage. In addition, melatonin may have beneficial effect to prevent these changes in the mucosa.

**(E) Yavaş MC, Yegin K, Oruç S, Delen K, Sirav B. Analysis of thiol/disulphide homeostasis and oxidant-antioxidant status as a result of exposure to radio-frequency electromagnetic fields. *Electromagn Biol Med* 2021 Jan 17;1-8. doi:10.1080/15368378.2021.1874970. Online ahead of print.**

The aim of the current study is to investigate the effect of daily long-term radio-frequency electromagnetic field (RF-EMFs) exposure on thiol/disulphide homeostasis and oxidant-antioxidant status in the serum of rats. Fourteen male Sprague Dawley rats were divided into two equal groups (n = 7). They were grouped as group 1 (sham-control) and group 2 (2100 MHz exposed). Group 2 rats were exposed to GSM-like signals for 5 h/day, 7 days per week for 14 days. In serum of blood samples, the native, total and native/total thiol levels and antioxidant-oxidant parameters were analyzed. The study revealed that the mean serum total thiol levels of GSM exposure group was rather higher than sham-control group ( $p = 0,219$ ); the mean serum native thiol levels of GSM exposure group was rather higher than sham-control group ( $p = 0,416$ ), and the mean serum disulphide levels of GSM exposure group was rather higher than sham-control group ( $p = 0,566$ ). On the other hand, the mean serum total antioxidant status (TAS), total oxidant status (TOS) and oxidative stress index (OSI) of GSM exposure group were higher than sham-control group. There are no data in the literature, that examine the effects of RF-EMFs on thiol/disulphide homeostasis. The results of this study, indicate that RF-EMFs did not cause oxidative stress and statistically changes in thiol/disulphide homeostasis. More sample sizes and longer-term exposures are needed to confirm the results of this study.

**(E) Yüksel M, Nazıroğlu M, Özkaya MO. Long-term exposure to electromagnetic radiation from mobile phones and Wi-Fi devices decreases plasma prolactin, progesterone, and estrogen levels but increases uterine oxidative stress in pregnant rats and their offspring. *Endocrine*. 52:352-362, 2016.**

We investigated the effects of mobile phone (900 and 1800 MHz)- and Wi-Fi (2450 MHz)-induced electromagnetic radiation (EMR) exposure on uterine oxidative stress and plasma hormone levels in pregnant rats and their offspring. Thirty-two rats and their forty newborn offspring were divided into the following four groups according to the type of EMR exposure they were subjected to: the control, 900, 1800, and 2450 MHz groups. Each experimental group was exposed to EMR for 60 min/day during the pregnancy and growth periods. The pregnant rats were allowed to stand for four generations (total 52 weeks) before, plasma and uterine samples were obtained. During the 4th, 5th, and 6th weeks of the experiment, plasma and uterine samples were also obtained from the developing rats. Although uterine lipid peroxidation increased in the EMR groups, uterine glutathione peroxidase activity (4th and 5th weeks) and plasma prolactin levels (6th week) in developing rats decreased in these groups. In the maternal rats, the plasma prolactin, estrogen, and progesterone levels decreased in the EMR groups, while the plasma total oxidant status, and body temperatures increased. There were no changes in the levels of reduced glutathione, total antioxidants, or vitamins A, C, and E in the uterine and plasma samples of maternal rats. In conclusion, although EMR exposure decreased the prolactin, estrogen, and progesterone levels in the plasma of maternal rats and their offspring, EMR-induced oxidative stress in the uteri of maternal rats increased during the

development of offspring. Mobile phone- and Wi-Fi-induced EMR may be one cause of increased oxidative injury in growing rats and decreased hormone levels in maternal rats. TRPV1 cation channels are the possible molecular pathways responsible for changes in the hormone, oxidative stress, and body temperature levels in the uterus of maternal rats following a year-long exposure to electromagnetic radiation exposure from mobile phones and Wi-Fi devices. It is likely that TRPV1-mediated  $\text{Ca}^{2+}$  entry in the uterus of pregnant rats involves accumulation of oxidative stress and opening of mitochondrial membrane pores that consequently leads to mitochondrial dysfunction, substantial swelling of the mitochondria with rupture of the outer membrane and release of oxidants such as superoxide ( $\text{O}_2^-$ ) and hydrogen peroxide ( $\text{H}_2\text{O}_2$ ). The superoxide radical is converted to  $\text{H}_2\text{O}_2$  by superoxide dismutase (SOD) enzyme. Glutathione peroxidase (GSH-Px) is an important antioxidant enzyme for removing lipid hydroperoxides and hydrogen peroxide and it catalyzes the reduction of  $\text{H}_2\text{O}_2$  to water.

**(E) Yurekli AI, Ozkan M, Kalkan T, Saybasili H, Tuncel H, Atukeren P, Gumustas K, Seker S. GSM Base Station Electromagnetic Radiation and Oxidative Stress in Rats. Electromagn Biol Med. 2006;25(3):177-188, 2006. (LI)**

The ever increasing use of cellular phones and the increasing number of associated base stations are becoming a widespread source of nonionizing electromagnetic radiation. Some biological effects are likely to occur even at low-level EM fields. In this study, a gigahertz transverse electromagnetic (GTEM) cell was used as an exposure environment for plane wave conditions of far-field free space EM field propagation at the GSM base transceiver station (BTS) frequency of 945 MHz, and effects on oxidative stress in rats were investigated. When EM fields at a power density of  $3.67 \text{ W/m}^2$  (specific absorption rate =  $11.3 \text{ mW/kg}$ ), which is well below current exposure limits, were applied, MDA (malondialdehyde) level was found to increase and GSH (reduced glutathione) concentration was found to decrease significantly ( $p < 0.0001$ ). Additionally, there was a less significant ( $p = 0.0190$ ) increase in SOD (superoxide dismutase) activity under EM exposure.

**(E) Zeni, O., Di Pietro, R., d'Ambrosio, G., Massa, R., Capri, M., Naarala, J., Juutilainen, J. and Scarfi, M. R. Formation of Reactive Oxygen Species in L929 Cells after Exposure to 900 MHz RF Radiation with and without Co-exposure to 3-Chloro-4-(dichloromethyl)-5-hydroxy-2(5H)-furanone. Radiat. Res. 167, 306-311, 2007.**

The aim of this study was to investigate the induction of reactive oxygen species in murine L929 fibrosarcoma cells exposed to radiofrequency (RF) radiation at 900 MHz, with or without co-exposure to 3-chloro-4-(dichloromethyl)-5-hydroxy-2(5H)-furanone (MX), a potent environmental carcinogen produced during chlorination of drinking water. Both continuous-wave and GSM mobile phone signals were applied for 10 or 30 min at specific absorption rates of 0.3 and  $1 \text{ W/kg}$ . Simultaneous sham exposures were performed for each exposure condition. MX treatment was performed at a subtoxic level of  $500 \text{ }\mu\text{M}$ , and the RF-field exposure was carried out during the first 10 or 30 min of the chemical treatment. The formation of reactive oxygen species was followed soon

after the exposure and at different harvesting times until 1 h after RF-field treatment. The studied provided no indication that 900 MHz RF-field exposure, either alone or in combination with MX, induced formation of reactive oxygen species under any of the experimental conditions investigated. In contrast, exposure to MX resulted in a statistically significant increase in the formation of reactive oxygen species for all the treatment durations investigated, confirming that MX is an inductor of oxidative stress in L929 cells.

**(E) Zhang X, Gao Y, Dong J, Wang S, Yao B, et al. (2014) The Compound Chinese Medicine “Kang Fu Ling” Protects against High Power Microwave-Induced Myocardial Injury. PLoS ONE 9(7): e101532. doi:10.1371/journal.pone.0101532.**

Background. The prevention and treatment of Microwave-caused cardiovascular injury remains elusive. This study investigated the cardiovascular protective effects of compound Chinese medicine “Kang Fu Ling” (KFL) against high power microwave (HPM)-induced myocardial injury and the role of the mitochondrial permeability transition pore (mPTP) opening in KFL protection. Methods. Male Wistar rats (100) were divided into 5 equal groups: no treatment, radiation only, or radiation followed by treatment with KFL at 0.75, 1.5, or 3 g/kg/day. Electrocardiography was used to Electrophysiological examination. Histological and ultrastructural changes in heart tissue and isolated mitochondria were observed by light microscope and electron microscopy. mPTP opening and mitochondrial membrane potential were detected by confocal laser scanning microscopy and fluorescence analysis. Connexin-43 (Cx-43) and endothelial nitric oxide synthase (eNOS) were detected by immunohistochemistry. The expression of voltage-dependent anion channel (VDAC) was detected by western blotting. Results. At 7 days after radiation, rats without KFL treatment showed a significantly lower heart rate ( $P<0.01$ ) than untreated controls and a J point shift. Myocyte swelling and rearrangement were evident. Mitochondria exhibited rupture, and decreased fluorescence intensity, suggesting opening of mPTP and a consequent reduction in mitochondrial membrane potential. After treatment with 1.5 g/kg/day KFL for 7 d, the heart rate increased significantly ( $P<0.01$ ), and the J point shift was reduced favorably ( $P<0.05$ ) compared to untreated, irradiated rats; myocytes and mitochondria were of normal morphology. The fluorescence intensities of dye-treated mitochondria were also increased, suggesting inhibition of mPTP opening and preservation of the mitochondrial membrane potential. The microwave-induced decrease of Cx-43 and VDAC protein expression was significantly reversed. Conclusion. Microwave radiation can cause electrophysiological, histological and ultrastructural changes in the heart. KFL at 1.5 g/kg/day had the greatest protective effect on these cardiovascular events. mPTP plays an important role in the protective effects of KFL against microwave-radiation-induced myocardial injury. (See Hu et al., 2014).

**(E) Zhu W, Cui Y, Feng X, Li Y, Zhang W, Xu J, Wang H, Lv S. The apoptotic effect and the plausible mechanism of microwave radiation on rat myocardial cells. Can J Physiol Pharmacol. 94(8):849-857, 2016.**

.Microwaves may exert adverse biological effects on the cardiovascular system at the integrated system and cellular levels. However, the mechanism underlying such effects remains poorly understood. Here, we report a previously uncharacterized mechanism through

which microwaves damage myocardial cells. Rats were treated with 2450 MHz microwave radiation at 50, 100, 150, or 200 mW/cm<sup>2</sup> for 6 min. Microwave treatment significantly enhanced the levels of various enzymes in serum. In addition, it increased the malondialdehyde content while decreasing the levels of antioxidative stress enzymes, activities of enzyme complexes I-IV, and ATP in myocardial tissues. Notably, irradiated myocardial cells exhibited structural damage and underwent apoptosis. Furthermore, Western blot analysis revealed significant changes in expression levels of proteins involved in oxidative stress regulation and apoptotic signaling pathways, indicating that microwave irradiation could induce myocardial cell apoptosis by interfering with oxidative stress and cardiac energy metabolism. Our findings provide useful insights into the mechanism of microwave-induced damage to the cardiovascular system.

**(E) Zielinski J, Ducray AD, Moeller AM, Murbach M, Kuster N, Mevissen M. Effects of pulse-modulated radiofrequency magnetic field (RF-EMF) exposure on apoptosis, autophagy, oxidative stress and electron chain transport function in human neuroblastoma and murine microglial cells. Toxicol In Vitro 2020 Oct;68:104963.**

The use of body-worn wireless devices with different communication protocols and rapidly changing exposure scenarios is still multiplying and the need to identify possible health effects of radiofrequency electromagnetic field (RF-EMF) exposure with extremely low-frequency (ELF) modulation envelopes. In this study, effects of ELF-modulated 935 MHz RF-EMF on apoptosis, autophagy, oxidative stress and electron exchange in N9 microglial and SH-SY5Y neuroblastoma cells were investigated. Cells were exposed at 4 W/kg or sham-exposed for 2 and 24 h. RF-EMF exposure of both cell types did not alter apoptosis, the number of living cells nor the apoptosis-inducing factor (AIF), irrespective of the exposure duration. RF-EMF exposure for 24, but not for 2 h, increased protein levels of the autophagy marker ATG5, whereas LC3B-I and II and pERK were not altered in both cell types and exposure times investigated. A transient increase in glutathione (GSH), but not hydrogen peroxide and cytochrome c oxidase was found only in SH-SY5Y cells, indicating that short-time RF-EMF at SAR levels accepted by today's safety guidelines might cause autophagy and oxidative stress with the effect being dependent on cell type and exposure duration. Further studies are needed to evaluate possible underlying mechanisms involved in pulse-modulated RF-EMF exposure.

**(E) Zmyslony M, Politanski P, Rajkowska E, Szymczak W, Jajte J. Acute exposure to 930 MHz CW electromagnetic radiation in vitro affects reactive oxygen species level in rat lymphocytes treated by iron ions. Bioelectromagnetics. 25(5):324-328, 2004.**

The aim of this study was to test the hypothesis that the 930 MHz continuous wave (CW) electromagnetic field, which is the carrier of signals emitted by cellular phones, affects the reactive oxygen species (ROS) level in living cells. Rat lymphocytes were used in the experiments. A portion of the lymphocytes was treated with iron ions to induce oxidative processes. Exposures to electromagnetic radiation (power density 5 W/m<sup>2</sup>, theoretical calculated SAR = 1.5 W/kg) were performed within a GTEM cell. Intracellular ROS were measured by the fluorescent probe dichlorofluorescein diacetate (DCF-DA). The results show

that acute (5 and 15 min) exposure does not affect the number of produced ROS. If, however, FeCl<sub>2</sub> with final concentration 10 microg/ml was added to the lymphocyte suspensions to stimulate ROS production, after both durations of exposure, the magnitude of fluorescence (ROS level during the experiment) was significantly greater in the exposed lymphocytes. The character of the changes in the number of free radicals observed in our experiments was qualitatively compatible with the theoretical prediction from the model of electromagnetic radiation effect on radical pairs.

**(E) Zong C, Ji Y, He Q, Zhu S, Qin F, Tong J, Cao Y. Adaptive Response in Mice Exposed to 900 MHz Radiofrequency Fields: Bleomycin-induced DNA and Oxidative Damage/Repair. *Int J Radiat Biol.* 91(3):270-276, 2015.**

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  $\mu\text{W}/\text{cm}^2$  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 hours later. In each animal: (i) the primary DNA damage and BLM-induced damage as well as its repair kinetics were determined in blood leukocytes; (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 is remaining after each 30, 60, 90, 120 and 150 minutes repair time, (c) 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) Zosangzuali M, Lalremruati M, Lalmuansangi C, Nghakliana F, Pachau L, P, Siama Z Effects of radiofrequency electromagnetic radiation emitted from a mobile phone base station on the redox homeostasis in different organs of Swiss albino mice. *Electromagn Biol Med* 40:393-407, 2021.**

This study was designed to investigate the possible effects of exposure to mobile phone base station (MPBS) emits 1800-MHz RF-EMR on some oxidative stress parameters in the brain, heart, kidney and liver of Swiss albino mice under exposures below thermal levels. Mice were randomly assigned to three experimental groups which were exposed to RF-EMR for 6 hr/day, 12 hr/day and 24 hr/day for 45 consecutive days, respectively, and a control group. The glutathione (GSH) levels and activities of glutathione-s-transferase (GST) and superoxide dismutase (SOD) were significantly reduced in mice brain after exposure to RF-EMR for 12 hr and 24 hr per day. Exposure of mice to RF-EMR for 12 hr and 24 hr per day also led to a significant increase in malondialdehyde (an index of lipid peroxidation) levels in mice brain. On the contrary, exposures used in this study did not induce any significant change in various oxidative stress-related parameters in the heart, kidney and liver of mice. Our findings showed no significant variations in the activities of aspartate amino-transferase (AST), alanine amino-

transferase (ALT), and on the level of creatinine (CRE) in the exposed mice. This study also revealed a decrease in RBC count with an increase in WBC count in mice subjected to 12 hr/day and 24 hr/day exposures. Exposure to RF-EMR from MPBS may cause adverse effects in mice brain by inducing oxidative stress arising from the generation of reactive oxygen species (ROS) as indicated by enhanced lipid peroxidation, and reduced levels and activities of antioxidants.

**(E) Zothansiyama, 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.**

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) 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. 2015 May 29;12(1):105. [Epub ahead of print]**

**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 dichlorofluorescein-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<sub>2</sub>O<sub>2</sub> 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.