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Chromosome damage in human cells induced by UMTS mobile telephony radiation

Chromosome damage in human cells induced by UMTS mobile telephony radiation. By: Panagopoulos, D J. Published in: General Physiology and Biophysics. 2019; 29: 346–354. https://doi.org/10.4149/gpb_2019032

During the past few decades, increasing concern has been raised regarding the potential adverse effects of human-made electromagnetic fields (EMFs), more specifically radio-frequency radiation, on human health. Radio-frequency electromagnetic fields (RF-EMF), as well as extremely low frequency (ELF) electromagnetic fields, have been classified as potentially carcinogenic to humans by the International Agency for Research on Cancer (IARC). Modern wireless communication devices combine radio-frequency carrier waves with extremely low frequency pulsing and modulation in order to increase the amount and speed of transmitted data. The most common source of radio-frequency electromagnetic fields in communication is the third generation (3G) Universal Mobile Telecommunications System (UMTS). Billions of mobile phone users are exposed to 3G radiation worldwide. A number of studies, including two large-scale long-term studies by the US National Toxicology Program (NTP) and the Italian Ramazzini Institute, demonstrate adverse effects of 2G and 3G radiation on tumor rates. The author of the study presented here evaluates potential genotoxic effects of UMTS (3G) radiation emitted by a commercially available mobile phone handset on human blood cells (lymphocytes). In science, peripheral blood lymphocytes are a well-known model for the assessment of genotoxicity of environmental agents such as radiation, smoking, pharmaceuticals, etc. A so-called "G2 assay" is used for this assessment. During the G2 phase of the cell cycle, chromosomal damage caused by the studied environmental agents becomes visible under the microscope. During the G2 or premitotic phase, chromosomes condense, thereby making them microscopically visible. This allows for the observation of breaks and gaps in chromosomes that suggest DNA damage.

Science Authors: Dipl. Biol. Isabel Wilke (IW), Roman Heeren (RH), B.Sc.

Study design:

Blood samples were collected from 6 healthy donors aged between 28 and 42 years. The individuals were men and women with moderate mobile phones use (less than about 30 minutes of conversation per day). Blood samples were cultured in 30 ml vials. For each individual, 800 cells (400 exposed and 400 sham-exposed) were examined for chromosomal damage. Chromosomal damage was visualized and quantified as chromatid breaks (terminal deletions) and chromatid gaps (achromatic lesions). Sample exposure was performed by a commercially available mobile phone handset during "talk" mode for 15 minutes at a distance of 1 cm to the wall of the culture vial. The average radio-frequency radiation exposure level measured 92 ± 27 μW/cm2. Extremely low frequency modulation occurred at 100 Hz with 12 ± 4.2 V/m and 0.09 ± 0.04 μT and at 1500 Hz with 8 ± 4.6 V/m and 0.006 ± 0.002 μT. According to the manufacturer, the SAR value of the handset for the human head is 0.66 W/kg. The measured exposure levels are representative of UMTS mobile phone EMFs during talk mode and well within the current ICNIRP limits.

Results:

A single call with the 3G mobile phone handset increased the number of chromosomal changes by 100–275% compared to the sham-exposed controls. The individual blood samples differed in their sensitivity to the exposure. The exposure caused mainly chromatid gaps, but also chromatid breaks in lower percentages. Both gaps and breaks (as well as the total number of changes) were significantly increased compared to the sham-exposed controls.

Conclusions:

The transformation of DNA damage into chromosomal abnormalities during cell division is a well-documented process. The chromosomal damage observed in this study is therefore likely due to DNA damage caused by mobile phone radiation. Cellular mechanisms were unable to repair this DNA damage and suggest a genotoxic/bioactive effect of radio-frequency radiation. The scientist was able to prove that thermal effects did not play a role. Previous studies on radio-frequency radiation and lymphocytes produced inconsistent results. The author contributes this, among other things, to the use of simulated fields instead of real devices as exposure sources. The high variability of modern mobile phone radiation, on the one hand, makes it very bioactive because living organisms can hardly adapt to it. Simulated fields, on the other hand, are more predictable and therefore easier to adapt to. The author recommends to drastically limit the use of mobile phones, to use wired headsets or speakers and generally to keep the largest possible distance between a mobile phone and the user's body. (RH)

Radio-frequency effect on male fertility

Long-term exposure to 4G smartphone radiofrequency electromagnetic radiation diminished male reproductive potential by directly disrupting Spock3–MMP2-BTB axis in the testes of adult rats

Long-term exposure to 4G smartphone radiofrequency electromagnetic radiation diminished male reproductive potential by directly disrupting Spock3–MMP2-BTB axis in the testes of adult rats. By: Yu G, Tang Z, Chen H, Chen Z, Wang L, Cao H, Wang G, Xing J, Shen H, Cheng Q, Li D, Wang G, Xiang Y, Guan Y, Zhu Y, Liu Z, Bai Z. Published in: Science of the Total Environment. 2020; 698. https://doi.org/10.1016/j.scitotenv.2019.133860

Mobile phones emit nonionizing radiation in the microwave range when in use. This radiation differs from other types of radio-frequency electromagnetic radiation because it affects organisms mainly by unclear, nonthermal effects rather than thermal effects. With the widespread use of mobile phones, the adverse impact of radiation on human health has become a global concern. As most men carry their mobile phones in their pant pockets immediately next to their testicles, scientists suspect that this habit may negatively affect the fertility of modern men. A number of studies report impaired male fertility as a result of mobile phone radiation. The present paper investigates the effect of a 4G mobile phone (4th generation of mobile communication technology; LTE) on the male reproductive system in rats. In addition to the observed effects, the research group also examined a possible biological mechanism of how mobile phone radiation affects male fertility.

Study design:

To investigate the reproductive effect of mobile phone radiation, a total of 135 rats were randomly divided into three exposure periods (50 days, 100 day and 150 days). Three experimental groups of 15 animals each were assigned to each of these periods. The rats of the normal (Nor) group remained in their cages without any intervention. The control (Con) group was sham-exposed, while the SRF group received smartphone radio-frequency (SRF) exposure. A customized unit was designed to ensure the exposure was restricted to the testicular area. A commercial 4G smartphone served as the radiation source. The smartphone under the rat's scrotum was kept in talk mode for 6 hours a day. It received an external call for 1 minute with 10 minutes intervals. During these 10 minutes, the phone was kept in talk mode. Experimental animals were exposed for 2 hours followed by a 30-minute break. This procedure was repeated 3 times a day. The frequency band of the

mobile phone was 2575–2635 MHz (TD-LTE). The strength of the electric field, power density and SAR value of the exposed area were 37.93 V/m, 22.74 W/m2 and 1.05 W/kg, respectively.

After the researchers had seen evidence of an effect of mobile phone radiation on male fertility, they started a second experiment that focused on the underlying mechanism. For this purpose, 7 experimental groups were assigned. The exposure duration was 150 days. The Nor group (n= 10) remained in their cages without any intervention. The Con group (n= 10) received saline injections into the testicles and was sham-exposed. The LV-SP3 group (n= 13) was also sham-exposed; however, they received intratesticular injections that downregulate the formation of a specific protein, namely Spock3. This protein is responsible for, among other things, the formation of correct cell-cell contacts and plays a role in the formation of the blood-testis barrier (similar to the blood-brain barrier). The blood-testis barrier is a very important cell-cell contact and is crucial for normal sperm production.

„The results of this study suggest that 4G mobile phone radiation has an adverse effect on testicular integrity as well as sperm quality."

Results:

First, the scientists verified that mobile phone radiation emitted by a commercial 4G smartphone did not increase the body temperature of the experimental animals. Thus, thermal effects can be excluded. In addition, serum cortisol levels (stress marker) were not increased in the Con group compared to the Nor group, demonstrating that the experimental procedure did not increase mental stress in the rats, which may have falsified results. In the first part of the experiment (investigation of radiation effects), the researchers were able to demonstrate a significant reduction in sperm quality (concentration, motility, survival, morphology) after 150 days of exposure.

Subsequently, a mating experiment was conducted to examine the male fertility potential. Although no differences in fertility as such were observed (successful mating, duration of pregnancy, litter size, viability, sex ratio), the pup weight was significantly reduced in the long-term exposure group (150 days). Afterwards testicular morphology was analyzed. After 150 days of exposure, an increase in the interferences with sperm formation, a significant loss of germ cells and a reduced height of the epithelium were observed. These results indicate that long-term exposure induced morphological damage in the testes of the experimental animals. Moreover, long-term exposure increased the rate of oxidative stress as well as apoptosis (programmed cell death) in the testes. In the second part of the experiment, the scientists aimed to elucidate the molecular biological basis of the observed phenomena. They discovered an artificial upregulation of the protein Spock3 after 150 days of exposure. Microscopic analysis of cell-cell contacts revealed an impairment of the blood-testis barrier, probably due to the excessive production of Spock3. The data was confirmed by a so-called "knock-down" experiment. The researchers were able to restore testicular damage as well as sperm quality by inhibiting Spock3 formation in exposed animals.

Conclusions:

The results of this study suggest that 4G mobile phone radiation has an adverse effect on testicular integrity as well as sperm quality. Most likely neither thermal effects nor increased mental stress levels are responsible for these observations. The scientists identified a specific protein, which is upregulated after long-term exposure. This leads them to hypothesize the mechanism by which mobile phone radiation might affect male fertility at a molecular level. They suspect an interference with the delicate balance of testicular epithelial tissue due to an increase in Spock3 formation. Components of the blood-testis barrier are disturbed, which in turn has an adverse effect on spermatogenesis. As a result, apoptosis of secondary germ cells gradually increases. This could lead to reduced male fertility. The research group recommends that men keep smartphones away from their testicles. (RH)

DNA damage by mobile phone radiation

Evaluation of genetically harmful radiation from mobile phones in male and female rats and mice after subchronic field exposure

Evaluation of the genotoxicity of cell phone radiofrequency radiation in male and female rats and mice following subchronic exposure. By: Smith-Roe SL, Wyde ME, Stout MD, Winters JW, Hobbs CA, Shepard KG, Green AS, Kissling GE, Shockley KR, Tice RR, Bucher JR, Kristine L. Witt KL. Published in: Environmental and Molecular Mutagenesis. 2019. https://doi.org/10.1002/em.22343

In the last two decades, mobile phone use has become almost ubiquitous worldwide. According to the International Telecommunication Union, about 7.68 billion mobile phone contracts with about 5.12 billion customers existed worldwide in 2017. There is concern that radio-frequency radiation emitted by mobile phones is capable of adversely affecting human health. Some epidemiological studies suggest that mobile phone use could increase the risk of certain types of brain cancer. However, conclusions from these studies may be premature since the data only covers the past two decades and this exposure period may be too short to assess cancer-related effects. The current state of scientific knowledge regarding possible genotoxic effects of mobile phone radiation is not consistent. The contradictory findings to date may partly be due to immense technical challenges involved in studying radio-frequency radiation. For the present study, rats and mice from the cancer study of the US National Toxicology Program (NTP) (see ElektrosmogReport 7/2016 and 3/2018) were used.

Study design:

In the NTP study, Sprague–Dawley rats and B6C3F1/N mice of both sexes were exposed to whole-body radio-frequency radiation. The exposure with GSM (2G) and CDMA (3G) was performed in custom-designed facilities, which had been specifically developed for the cancer study. The exposure of rats was initiated in the uterus (day 5 after fertilization), while the exposure of mice began on day 35 after birth. The exposure was performed over 18 hours a day at 10-minute intervals, resulting in a total exposure of 9 hours and 10 minutes per day. The experimental animals were divided into 4 exposure groups with different radiation intensities, resulting in 0, 1.5, 3 or 6 W/kg (rats) and 0. 2.5, 5 or 10 W/kg (mice), respectively. For the rats, an exposure frequency of 900 MHz was chosen and for the mice 1900 MHz. After a total exposure of 19 weeks (rats) and 14 weeks (mice), 5 animals per sex from each exposure group were removed from the ongoing NTP cancer study and the effect of subchronic exposure to radio-frequency electromagnetic radiation was analyzed. Two methods were used, the alkaline comet test and the micronucleus test. The comet test provides information on DNA damage, while the micronucleus test reveals chromosomal damage. Numerous tissues were analyzed: The comet test was performed on cells from 3 brain regions (frontal cortex, hippocampus, cerebellum), liver cells and leucocytes from peripheral blood. The micronucleus test was conducted on immature and mature erythrocytes of the peripheral blood.

Results:

In rats, the comet test revealed significant, dose-dependent DNA damage only in the hippocampal region of male rats after 3G exposure. In all other assays (tissue, 2G exposure, female animals), only inconclusive or no DNA damage was observed. In mice, dose-dependent DNA damage was detected in the frontal cortex of male animals with both types of radio-frequency radiation exposure (2G and 3G). The leucocytes of female mice exhibited increased and dose-dependent DNA damage as well. The remaining assays showed no or inconclusive DNA damage. In total, 8 out of 40 sample sets showed clear or ambiguous DNA damage, while the remaining samples had no evidence of an adverse effect of mobile phone radiation on DNA damage. The micronucleus test provided no indications of chromosomal damage whatsoever.

Conclusions:

Although the results of the micronucleus test were negative, the comet test revealed DNA damage in several tissues of rats and mice after exposure to CDMA and GSM radiation. This indicates that mobile phone radiation can cause DNA damage. DNA damage was mainly observed in brain tissues of male rats and mice. According to the scientists, a larger number of animals could have contributed to improving the detectability of DNA damage. A further limitation of the study is the lack of a histopathological analysis, which could, for example, determine inflammatory or cytotoxic processes. The results suggest that exposure to mobile phone radiation could cause measureable genotoxic effects. However, the authors emphasize that the groups with the highest exposure (6 W/kg in rats and 10 W/kg in mice) were exposed at higher levels than humans normally are. Thus, the question of whether radio-frequency electromagnetic radiation can cause adverse health effects in humans has not yet been clarified; further studies are in progress. (RH)

Review on radio-frequency radiation

The contribution of in vivo mammalian studies to the understanding of the adverse effects of radiofrequency radiation on human health

The contribution of in vivo mammalian studies to the knowledge of adverse effects of radiofrequency radiation on human health. By: Vornoli A, Falcioni L, Mandrioli D, Bua L, Belpoggi F. Published in: International Journal of Environmental Research and Public Health. 2019; 16: 3379. https://doi.org/10.3390/ijerph16183379

As the use of mobile phones has become an integral part of daily life for the vast majority of the population, the last two decades have witnessed an unprecedented exposure to radiofrequency radiation. Consequently, a growing public interest in the potential health risks associated with mobile phone use and the exposure to base stations arises. Radio-frequency electromagnetic radiation (RFR) provides enough energy to move or vibrate atoms of a molecule, but not enough to remove electrons from atoms or molecules. Therefore, RFR is classified as nonionizing radiation. Important physical properties of nonionizing radiation are frequency, intensity and specific absorption rate (SAR), which is defined as the energy absorption per unit weight of biological tissue. RFR is capable of heating tissue with sufficient intensity, comparable to a microwave oven. The adverse effects of RFR on biological systems are often divided into thermal and nonthermal effects. While thermal effects are often plausible and easy to explain (tissue stress), humans are usually exposed to RFR exposure levels below the thermal threshold level. Although some scientists challenge the capability of RFR to have nonthermal effects, there is an increasing number of studies which report specific biological effects. The present review covers the current state of knowledge about carcinogenic and reproductive hazards of RFR, as demonstrated by experimental "in vivo" (in experimental animals) studies.

Study design:

The authors analyzed the results of peer-reviewed studies conducted on laboratory animals (in vivo). With regard to carcinogenicity studies (potentially carcinogenic properties), the authors followed the guidelines of the OECD (Organisation for Economic Co-operation and Development) and NTP (National Toxicology Program). These guidelines state, among other things, that (1) each exposure and respective control group should contain at least 50 animals of each sex, (2) at least three doses should be investigated and the exposure period as well as the duration of the study should last at least 24 months. Only studies that met these criteria were covered in this review. Concerning the fertility studies, the authors followed the OECD and NTP study guidelines as well.

Results:

The carcinogenicity studies were reviewed first. They were divided according to the model organism used: rats, mice and "others." The latter specifically refers to transgenic/tumor-prone animal strains that address the effects of radio-frequency electromagnetic radiation in a specific tumor context. In total, the authors reviewed 6 studies in rats, 4 in mice and 4 in tumor-prone mice. The studies were conducted between 1982 and 2018. Fifty percent of the respective studies (3 rats, 2 mice, 2 tumor-prone mice) discovered increased tumor rates after RFR exposure. Subsequently, the authors addressed studies that focus on possible adverse effects on male fertility. In this case, the study period was between 2003 and 2019. Fourteen out of twenty (70%) studies on male rat fertility confirmed significant detrimental effects of RFR. Studies in mice and rabbits also confirmed significant impairments in male fertility with 4 out of 4 (100%) and 2 out of 2 (100%), respectively. Studies on female fertility (between 1983 and 2013) confirmed a deterioration after RFR exposure in rats in 5 out of 11 cases (45%). A similar result was observed in female fertility studies on mice, with 2 out of 5 (40%) reporting impairments after exposure.

Conclusions:

These results of studies on the male reproductive system indicate that radio-frequency radiation can adversely affect male fertility. Researchers frequently observe a decrease in sperm viability, quality and motility. Oxidative stress also occurs more abundantly, which mainly damages testicular tubules, sperm progenitor cells and Leydig cells. According to the authors, the data on the female reproductive system, however, varies greatly, the studies are very inconsistently conducted and report mainly different specific findings. As a result, it is difficult to draw conclusions from the current state of scientific knowledge on the female reproductive system. The authors criticize the study design of cancer studies, which discovered no effects of RFR on tumorigenesis. Among the deficiencies are short daily exposure periods, low exposure levels and keeping experimental animals under crowded conditions. In addition, the radiation doses were unjustifiable and characterized by poor dosimetry that did not consider animal growth. Even for studies reporting increased tumor rates, the authors make suggestions on how to improve them. The experiments of the NTP and Ramazzini Institute use simulated signals produced by signal generators instead of real mobile phones or base station antennas. Real-world signals are much more unpredictable, versatile and therefore more bioactive. It is assumed that the protective mechanisms of living organisms are less protective against these extremely fast-changing environmental stressors. The data from the NTP study and the Ramazzini Institute study could underestimate the potentially detrimental effect of radio-frequency electromagnetic radiation. (RH)

2.4 GHz radiation from Wi-Fi causes poor insulin secretion and increases oxidative stress in the pancreatic islet cells of rats

Radiofrequency radiation emitted from Wi-Fi (2.4 GHz) causes impaired insulin secretion and increased oxidative stress in rat pancreatic islets. By: Masoumi A, Karbalaei N, Mortazavi SMJ, Shabani M. Published in: International Journal of Radiation Biology. 2019. https://doi.org/10.1080/09553002.2018.1490039

In the pancreas, more precisely in the beta cells of the islets of Langerhans, insulin is produced and secreted. Insulin plays an essential role in metabolism and blood sugar balance. Functional disorders disturb the blood sugar balance and contribute to diabetes. Some studies demonstrate a detrimental effect of electromagnetic fields on morphology and insulin secretion. Currently, little is known about the effect of Wi-Fi radiation on insulin secretion and the insulin level of islet cells. One of the aims of this study was to address the effect of Wi-Fi radiation on insulin secretion. The pancreas is sensitive to oxidative stress since it has a weak antioxidant defense system. Therefore, it is of particular relevance as to whether Wi-Fi radiation (2.45 GHz) generates oxidative stress in the pancreas of rats. Numerous physiological processes such as those of mitochondria can produce reactive oxygen species (ROS). ROS production is regulated by enzymatic and nonenzymatic antioxidants. A disturbance of this oxidative balance is capable of causing oxidative damage to biomolecules such as nucleic acids, fats and proteins. Consequently, the influence of Wi-Fi radiation on antioxidant enzyme activities as well as lipid peroxidation in the pancreas of rats was investigated as well.

Study design:

Male rats (12 weeks of age) were randomly divided into 3 groups of 8 animals each: control group, sham-exposed group and Wi-Fi-exposed group. Both exposure groups were either exposed or sham-exposed for 4 hours a day for 45 days. The Wi-Fi router operated with 0.1 W; cages were placed 30 cm away. Animal weight was determined weekly. At the end of the exposure period, the weight of the pancreas was recorded as well.

Furthermore, plasma glucose and plasma insulin levels were analyzed and a glucose tolerance test was performed. To evaluate insulin secretion, islet cells were exposed to 2.8 and 16.7 mmol/l of glucose for 60 minutes. Afterwards, the insulin content in the culture medium was measured to determine insulin resistance. On day 45 of the experiment, the pancreas was removed and islet cells were isolated to evaluate insulin secretion and content. Moreover, antioxidant status in the pancreatic tissue was analyzed. For this analysis, relevant parameters were GSH, SOD, catalase and GPx activities as well as lipid peroxidation.

Results:

Data shows that the weight gain of Wi-Fi-exposed animals was significantly lower compared to the two control groups (sham-exposed and control group). No significant variations in pancreatic weight were observed. The exposed group showed increased glucose concentrations (high blood glucose) during glucose tolerance test. The insulin plasma levels as well as the glucose-stimulated insulin secretion of islet cells were significantly reduced in rats exposed to Wi-Fi radiation.

In the beginning of the glucose tolerance test (null value), no significant differences in glucose levels, insulin levels and insulin content of islet cells could be observed. The Wi-Fi radiation led to significantly increased plasma glucose levels, i.e. glucose tolerance was significantly lower than in the two control groups. Moreover, plasma insulin levels were significantly reduced during glucose tolerance test. The analysis of oxidative stress markers in the pancreatic tissue showed highly significantly increased lipid peroxidation in the exposed group, while antioxidant activities of GSH, SOD and GPx were significantly reduced. Catalase activity did not differ significantly between exposure and control groups.

Conclusions:

This study shows that 2.45 GHz Wi-Fi radiation causes lower weight gain, impaired glucose tolerance and insulin secretion as well as high blood glucose in rats. It also results in increased oxidative stress in the pancreas and islet cells (increase in ROS and lipid peroxidation paired with reduced antioxidant activity). Rats exposed to 2.45 GHz radiation exhibited lower glucose tolerance leading to significantly increased plasma glucose and lower plasma insulin levels. Further experiments revealed an impaired insulin secretion of islet cells at high glucose stimulation levels. It is known that pancreatic insulin secretion is crucial for glucose homeostasis. Therefore, an increase in blood glucose induced by 2.45 GHz exposure could be associated with a diminished insulin secretion of islet cells. Possible mechanisms for elevated levels of blood glucose due to Wi-Fi radiation may include conformational changes of insulin, insulin receptors and glucose transport proteins. (IW & RH)

Morphological changes induced by extremely low frequency electric fields

Morphological changes induced by extremely low-frequency electric fields. By: Imani M, Kazemi S, Saviz M, Farahmand L, Sadeghi B, Faraji-dana R. Published in: Bioelectromagnetics. 2019; 40: 375–390. https://doi.org/10.1002/bem.22195

In this paper, different cell types were examined for possible changes after exposure to extremely low frequency electric fields. Electric fields affect growth, regeneration, survival rate, organization and activity of cells in the laboratory (in vitro). For example, these properties are used to stimulate the healing of wounds or fractured bones. The regeneration of nervous tissue is enhanced as well. We know that amphibians are capable of regenerating damaged organs, such as lost limbs. Differentiated cells from the wound environment are dedifferentiated for reconstruction of a new limb. Differentiated cells are basically "normal" body cells with a specific function (e.g. liver, kidney or nerve cells) and mature from (undifferentiated) stem cells into these specific cells. These cell processes are probably controlled by endogenous electric fields, as experiments with mammalian bone marrow cells demonstrated. This led to the hypothesis that electric fields are capable of inducing cell dedifferentiation. Further evidence was provided in experiments with static and alternating fields, where red blood cells of amphibians changed into stem-like cells. Calcium plays a role in these morphological changes. Since morphological changes in the erythrocytes of birds (which contain cell nuclei in contrast to human erythrocytes, the editor) may be due to dedifferentiation, the researchers assumed that the reason for the discoloration of the cells is a loss of hemoglobin and that the loss of hemoglobin leads to dedifferentiation. The experiments were designed to verify this.

Study design:

First, red blood cells were examined for alterations. Cells from a female city pigeon (Columba livia domestica) were exposed to 5, 12, 25 and 40 Hz for 4 days at room temperature. The electric field at 50 Hz was 200 mV. Field strengths were 2.103 V/m at the central electrode, 0.055 V/m at the peripheral electrode and 0.404 V/m midway between the two electrodes. Two control sets consisted of sham-exposed cells (with electrodes turned off) and cells without electrodes. Human mesenchymal stem cells (i.e. stem cells of connective tissue) from umbilical cord blood and human bone marrow were used to elucidate the mechanisms. Both types of mesenchymal stem cells were exposed to 50 Hz. Alizarin red staining was used to determine the calcium concentration. During the 4-day exposure, cells were counted under the microscope and pictures as well as videos were taken. Trypan blue staining was used to identify dead cells. Temperature and pH-value in the culture media were monitored regularly.

Results:

Under the light microscope, deformations from elliptical to spherical were observed in exposed pigeon erythrocytes. Moreover, cytoplasm was more transparent, i.e. less staining, probably due to a loss of hemoglobin. Trypan blue staining was performed to determine whether these changes were caused by cell death. Microscopic analysis showed living cells. Exposed samples developed radial stripes around the cell nucleus before the beginning of cell deformation, indicating a high RNA concentration in this area. Especially at 50 Hz, significant discrepancies between exposed and control cells were evident. Therefore, these changes are frequency-dependent. Thermal effects could be excluded, as the temperature in the culture medium varied by only 0.002 °C. No shifts in pH were detected. Since the culture medium was changed after exposure, indirect alterations of macromolecules by the electric fields can be excluded and a direct interaction between electric fields and cells can be assumed. In this context, cell membranes are of particular interest.

Dedifferentiation in the two mesenchymal cells from bone marrow, which had differentiated into bone cells, could be detected. Bone cells contain high calcium concentrations. After exposure to electric fields, the quantification of calcium revealed decreasing concentrations, a sign for a less differentiated state of the cells.

Conclusions:

The investigation of the effects of selected frequencies (5, 15, 25 and 50 Hz) on living cells revealed morphological alterations, which are possibly based on mechanisms in calcium channels. The consequence might be the dedifferentiation of cells, which can be assumed for at least 2 types of human cells. Given the medical importance of these results, further research should be performed to understand the underlying mechanisms. (IW)