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Genetic Effects of Nonionizing Electromagnetic Fields

Henry Lai
Bioelectromagnetics Research Laboratory
Department of Bioengineering
University of Washington
Seattle, WA
USA

Nonionizing electromagnetic fields (EMF) have photon energy less than 10eV, a level not sufficient to produce ions by ejection of orbital electrons from atoms. The biological effects of two types of nonionizing electromagnetic fields are being studies intensely: extremely-low-frequency (ELF) electromagnetic field and radiofrequency radiation. Extremely-low-frequency EMF covers the frequency range of 3 Hz to 3 KHz. The most intensely studied frequency is the power frequency of 50/60 Hz. Electric appliances and power lines emit 50/60 Hz EMF. Radiofrequency radiation (RFR) covers a frequency range between 10 KHz to 300 GHz. Different frequencies of RFR are used in different applications. For example, the frequency range of 5.4 to 16 KHz is used in AM radio transmission, while 76 to 108 MHz is used for FM radio. Mobile phone technology uses frequencies between 800 MHz and 3 GHz. And RFR of 2450 MHz is used in microwave cooking.

Genetic effects of ELF-EMF and RFR have been reported in various studies [e.g., Garaj-Vrhovac et al., 1991; Maes et al., 1993; Sarkar et al., 1994; Simko et al., 1998; Zotti-Martelli et al., 2000]. However, since the energy of nonionizing EMF is not sufficient to break chemical bonds directly, the effects have to be caused by indirect mechanisms. In this brief paper, I have described the research we carried out in our laboratory on genetic effects of nonionizing EMF. We studied mainly effects of ELF- EMF and RFR on DNA strand breaks in brain cells of rats exposed in vivo. Details of the exposure systems used in our studies have been described by Guy et al. [1979] and Lai et al. [1993]. In bioelecromagnetics research, it is very important that the exposure system be well characterized particularly with regard to energy absorption and field uniformity.

The microgel electrophoresis assay (comet assay) [cf. Singh, 1996] was used to measure single and double strand DNA breaks in brain cells of the rat. The assay can be used to evaluate DNA strand breaks in a single cell and can detect one break per 2×10^{10} daltons of DNA, which is more sensitive than other available methods of strand break detection. The assay involves making microgel with isolated cells dispersed in low-melting temperature agarose on a microscopic slide. Cells are then lysed with high salt and detergent, and then treated with enzymes to remove RNA and proteins, so that only DNA remains. The slide is then subjected to electrophoresis and the extent of DNA fragment migration from the nucleus is used an index of DNA breaks. If the electrophoresis is done at highly alkaline pH (>13), the paired strands of DNA separate prior to electrophoresis and single strand breaks will be detected. Under neutral pH

conditions, the DNA strands remain joined and any fragment migrated out must have resulted from double strand breaks. In isolated human lymphocytes, the assay can detect single and double strand DNA breaks caused by 5-10 cGy and 10-15 cGy of x-rays, respectively.

We investigated the effects of 60-Hz magnetic field exposure on DNA in brain cells of the rat [Lai and Singh, 1997a]. We observed an increase in DNA single strand breaks after 2 hrs of exposure to a magnetic field at an intensity of 0.1, 0.25, or 0.5 millitesla (mT) (0.1 mT = 1 gauss), whereas an increase in double strand breaks was observed at 0.25 and 0.5 mT, but not at 0.1 mT. The effect is proportional to the intensity of the magnetic field. Similarly, exposure to RFR (2450 MHz, at a whole body specific absorption rate (SAR) of 0.6 and 1.2 W/kg) for 2 hrs caused an increase in both single and double strand breaks in DNA of brain cells in the rat [Lai and Singh, 1995, 1996]. Another interesting finding from our research is that time and intensity can interchange in exerting effects of magnetic fields. By increasing the duration of exposure to 24 hrs, increases in single and double strand DNA breaks could be observed in brain cells of rats exposed to a 60-Hz magnetic fields at an intensity of 0.01 mT, whereas a 2-hr exposure at the same intensity had no significant effect.

From the microgel electrophoresis assay, exposure to a 60-Hz magnetic field at 0.25 mT for 2 hrs or to 2450-MHz RFR at an average SAR of 1.2 W/kg for 2 hrs produces a similar DNA migration in brain cells as that caused by 25 cGy of X-rays, i.e., an average of 250 strand breaks per cell. However, it is not likely that the three entities cause DNA breaks by similar mechanism and produce the same types of DNA damage.

It must be pointed out that the 0.1-0.5 mT magnetic field intensities used in our study are much higher than the levels most people encounter in daily life. However, they are still within the limits contained in current magnetic field exposure guidelines and can be encountered in occupational situations. For example, the International Nonionizing Radiation Committee of the International Radiation Protection Association guidelines for maximum levels of magnetic field exposure in occupational situations are 0.5 mT for workday exposure and 5 mT for short-term exposure, whereas for the general public it is 0.1 mT for 24 hrs per day exposure and 1 mT for exposure for a few hrs per day. Regarding RFR exposure, one can get an SAR of 6-8 W/kg per gm of tissue in certain parts of the head when using a mobile phone.

In further research, we found that treatment of rats before exposure with free radical scavengers blocked the effects of EMF (ELF-EMF and RFR) on DNA [Lai and Singh, 1997b,c]. This suggests that EMF enhances free radical activity in cells, which in turn lead to DNA damage. We also found that EMF exposure caused DNA-protein and DNA-DNA crosslinks [Singh and Lai, 1998] and increased apoptosis and necrosis in brain cells of the rat. Furthermore, we found that pretreating rats with an iron-chelator could block the effects of EMF exposure on DNA.

In addition to our experiments, using the microgel electrophoresis assay, Ahuja et al. [1997, 1999], Phillips et al. [1997], and Svedenstal et al. [1999a,b] have also reported an increase in DNA strand breaks in cells after magnetic field exposure. Interestingly, Svedenstal et al [1999a] observed an increase in DNA strand breaks in brain cells of mice after 32 days of exposure to magnetic fields at a low intensity of 7.5 microtesla. Changes in DNA in cells exposed to RFR, as detected by the microgel electrophoresis assay, have also been reported by Phillips et al. [1998] and Verschaeve et al. [1994].

From the results of the above research, we hypothesize that EMF initiates an iron-mediated process (Fenton reaction) that increases hydroxy free radical formation in cells, leading to DNA strand breaks and cell death. Cells with high rates of iron intake, e.g., proliferating cells, cells infected by DNA virus, and cells with high metabolic rates such as brain cells, would be more susceptible to the effects of EMF. For proliferating cells, the most vulnerable time should be during the G_1/S phases of the cell cycle, when transferrin receptors are expressed and iron influx is high. Hydroxy radicals are generated from hydrogen peroxide via the Fenton reaction in the presence of iron. Cells with high metabolic rate generate high amount of hydrogen peroxide via the mitochondrial electron transport pathway and thus are more vulnerable to EMF. On the other hand, possible harmful effect of EMF exposure could also depend on the capability of cells to store iron in ferritin. For example, liver cells would be less susceptible to EMF, even though they have high iron influx, because they contain high amount of ferritin.

Cancer cells are known to have a higher concentration of transferrin receptors on their cell surface and uptake a large amount of iron. In a series of experiments, effects of exposure to a 60-Hz magnetic field on cancer cells were investigated. Molt-4 cells, a type of human lymphoblastoid cells, were exposed to a 60-Hz magnetic fields (0.25 mT) for 2 hrs in a medium supplemented with holotransferrin, a protein that transports iron into cells. A significant decrease in cell count was observed after exposure when compared to that of non-exposed samples. The effect lasted for at least 22 hrs after exposure. Magnetic field alone (without holotransferrin) was ineffective. In addition, similar magnetic field/holotransferrin treatment had only a slight effect on normal human lymphocytes. These data indicate that when intracellular iron concentration is increased, cancer cells become more susceptible to an alternating magnetic field, resulting in cell death or cell cycle arrest. Thus, low frequency alternating magnetic fields may be useful for cancer treatment. In studies by the late Charles Hannan and his associates, the growth rate of implanted tumors in mice was significantly decreased by exposure to a pulsed magnetic field (1 hr per day at an average intensity of 0.5 mT) [Hannan et al., 1994]. The field also enhanced the potency of the anti-tumor compound daunorubicin on implanted multi-drug resistant tumor in mice in vivo [Liang et al., 1997]. More recently, Santi Tofani and his associates [2001] in Italy reported an increase in cell death morphologically consistent with apoptosis in two transformed cell lines (WiDr human colon adenocarcinoma and human breast adenocarcinoma) exposed to magnetic fields of more than 1 mT. No toxic morphological changes were observed in non-transformed cells (MRC-5 embryonal lung fibroblast) after the same exposure. In addition, nude mice bearing WiDr tumors subcutaneously treated with daily exposure of magnetic fields showed a significant tumor growth inhibition (up to 50%).

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