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Effects of co-exposure to extremely low frequency (50 Hz) magnetic fields and xenobiotics determined in vitro by the alkaline comet assay

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Abstract

In the present study, we used human peripheral blood leukocytes from 4 different donors, to investigate in vitro the possible genotoxic and/or co-genotoxic activity of extremely low frequency magnetic fields (ELF-MF) at 3 mT intensity. Two model mutagens were used to study the possible interaction between ELF-MF and xenobiotics: *N*-methyl-*N*′-nitro-*N*-nitrosoguanidine (MNNG) and 4-nitroquinoline *N*-oxide (4NQO). Primary DNA damage was evaluated by the alkaline single-cell microgel-electrophoresis ("comet") assay. Control cells (leukocytes not exposed to ELF-MF, nor treated with genotoxins) from the different blood donors showed a comparable level of basal DNA damage, whereas the contribution of individual susceptibility toward ELF-MF and the tested genotoxic compounds led to differences in the extent of DNA damage observed following exposure to the genotoxins, both in the presence and in the absence of an applied ELF-MF. A 3 mT ELF-MF alone was unable to cause direct primary DNA damage. In leukocytes exposed to ELF-MF and genotoxins, the extent of MNNG-induced DNA damage increased with exposure duration compared to sham-exposed cells. The opposite was observed in cells treated with 4NQO. In this case the extent of 4NQO-induced DNA damage was somewhat reduced in leukocytes exposed to ELF-MF compared to sham-exposed cells. Moreover, in cells exposed to ELF-MF an increased concentration of GSH was always observed, compared to sham-exposed cells. Since following GSH conjugation the genotoxic pattern of MNNG and 4NQO is quite different, an influence of ELF-MF on the activity of the enzyme involved in the synthesis of GSH leading to different activation/deactivation of the model mutagens used was hypothesized to explain the different trends observed in MNNG and 4NQO genotoxic activity in the presence of an applied ELF-MF. The possibility that ELF-MF might interfere with the genotoxic activity of xenobiotics has important implications, since

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human populations are likely to be exposed to a variety of genotoxic agents concomitantly with exposure to this type of physical agent.

Keywords: Extremely low frequency electromagnetic fields; ELF-EMF; Primary DNA damage; Alkaline comet assay; Genotoxicity; Co-genotoxicity

1. Introduction

The generation, transmission, distribution and use of electric energy is associated with the production of electric and magnetic fields (EMF), which oscillate 50 to 60 times per second (power-line frequency: 50 or 60 Hz, in Europe and North America, respectively). This range falls within the extremely low frequency (ELF) region of the electromagnetic spectrum (frequencies from 3 to 3000 Hz) (Poole and Ozonoff, 1996). As a consequence of the very wide wavelengths (approximately 6000 and 5000 Km at 50 and 60 Hz, respectively) in the ELF range, electric and magnetic fields (EF and MF, respectively) propagate effectively uncoupled. Moreover, in consideration of the size of human body and the common distance from ELF-EMF sources, the exposure always corresponds to "near-field" conditions (less than one wavelength) (Polk and Postow, 1995). As the MF component of ELF fields readily enters the body – unlike the EF component which is efficiently shielded – it is more likely that any biological effect is due to secondary currents induced in the body by the MF.

The first suggestion of adverse health effects associated with exposure to ELF-MF derived from studies performed in the former USSR in the early 1960s. Physical examinations of workers in high-voltage switchyards revealed symptoms of nervous system dysfunction (e.g. vegetative dystonia, neurasthenic/hypertonic symptoms) or cardiovascular disorders (e.g. arrhythmia, increased heart rate and arterial pressure) during and shortly after ELF-MF exposure (Asanova and Rakov, 1966).

The possibility of an association between ELF-MF exposure and increased cancer incidence (e.g. childhood acute leukaemia, cancer of the nervous system and lymphomas) was first proposed in the late 1970s. The results of a case–control study on childhood cancer carried out in Denver, USA, indicated an excess risk of nervous system tumors (OR 2.36 — 95% CI 1.03–5.41) and leukaemia (OR 2.98 — 95% CI 1.78–4.98) among children living in homes categorized as with "high current configuration" on the basis of proximity to power lines and/or transformers ("wire coding") (Wertheimer and Leeper, 1979). This report was followed by a considerable number of epidemiological studies investigating the possible association between residential or occupational exposure to ELF-MF and various cancer diseases (e.g. leukaemia, cancer of the central nervous system and lymphoma).

Reviews of epidemiological studies focusing on the potential role of 50/60 Hz MF in the aetiology of cancer suggested that the epidemiological evidence to support the association between exposure to ELF-MF and the risk of childhood leukaemia is less consistent than what was observed in the mid 1980s, with an OR of about 1.5 (US National Research Council, 1997; Portier and Wolfe, 1998; Repacholi and Greenebaum, 1999).

It is generally accepted that ELF-MF are unable to transfer energy to cells in sufficient amounts to damage DNA directly and thus are considered to be non-genotoxic. However, it is possible that certain cellular processes altered by exposure to ELF-MF, such as free radicals production and/or activity (Brocklehurst and McLauchlan, 1996), might indirectly affect the structure of DNA. In this context, several research groups sought to determine whether a link existed between 50/60 Hz ELF-MF generated by high-voltage power lines or electrical appliances and mutagenesis, and to determine the possible mechanism of cancer risk. Comprehensive reviews regarding in vivo and in vitro laboratory studies on ELF-MF (Juutilainen and Lang, 1997; McCann et al., 1998; Moulder, 1998) pointed out the conflicting results reported, with genotoxic endpoints such as chromosome aberrations (CA), micronuclei (MN), sister chromatid exchange (SCE), and DNA strand-breakage at exposure levels ranging from 1 μT to 10 mT.