Genetics and Molecular Biology, 27, 3, 460-466 (2004)

www.sbg.org.br

Short Communication

Cytogenetic analysis of the effects of 2.5 and 10.5 GHz microwaves on human lymphocytes

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Abstract

The biological effects of microwaves on living organisms remain highly controversial. Although some reports have suggested that microwaves may be directly or indirectly genotoxic, a direct action is unlikely because the low energy of microwave photons makes them unable to cause single-strand breaks in DNA. In this work, we examined the possible clastogenic properties of microwaves (2.5 and 10.5 GHz) on blood lymphocytes *in vitro* by monitoring the frequency of chromosomal aberrations. We also investigated whether blood cells showed increased radiosensitivity or radioresistance when pretreated with the microwaves and then irradiated with gamma radiation. There was no significant difference in the frequency of chromosomal aberrations between cells which had or had not been treated with microwaves. Control cells had a mean frequency of 0.013 aberrations per cell compared to 0.010 and 0.011 aberrations per cell in the microwave-exposed samples. Nor was there any alteration in the radiosensitivity of cells pretreated with microwaves. Gamma irradiated cells showed a mean frequency of 0.279 aberrations per cell compared to 0.343 and 0.310 aberrations per cell in samples pretreated with microwaves. However, cell mortality increased markedly after exposure to microwaves. The results suggest that microwaves do not interact directly or indirectly with chromosomes, although they may target other cell structures, such as cell membranes.

Key words: chromosomal aberrations, human lymphocytes, microwaves.

Received: August 15, 2003; Accepted: November 14, 2003.

Introduction

In recent decades, there have been considerable advances in the development of sources of non-ionizing radiation, such as microwaves. The widespread use of such energy sources and the increase in the number of devices emitting microwaves and radiofrequencies (RF), including mobile phones, has become a matter of concern for regulatory authorities and non-regulatory bodies (IEGMP, 2000).

Structural chromosomal aberrations may involve the chromosomes or chromatids. Most chemical mutagens and non-ionizing mutagenic radiations are unable to cause double-strand breaks in DNA and act mainly in the S phase of the cell cycle. Such agents are only indirectly clastogenic and produce mainly chromatid-type aberrations (OECD, 1997). In contrast, chromosome-type aberrations are induced directly by agents such as ionizing radiation that can produce double strand breaks in DNA (IAEA, 2001).

The biological effects of microwaves on living organisms are highly controversial (Maes *et al.,* 1993). A direct genotoxic action is unlikely because of the low energy of microwave photons which are unable to cause strand breaks in DNA. However, despite this general conviction that microwaves are not sufficiently energetic to be able to directly damage DNA, there is considerable evidence indicating that microwaves can be directly and indirectly clastogenic, with a significant increase in chromosome damage (Sagripanti and Swicord, 1986; Garaj-Vrhovac *et al.,* 1991, 1992; Maes *et al.* 1993; Haidler *et al.,* 1994; Sarkar *et al.,* 1994; Lai and Singh 1995, 1996; Timchenko and Ianchevskaia, 1995; Balode, 1996; Verschaeve *et al.,* 1994; Vijayalaxmi *et al.,* 1997; Phillips *et al.,* 1998; Tice *et al.,* 1999). In addition, cell phone radiation can alter proto-oncogene activity (Ivaschuk *et al.,* 1997; Goswami *et al.,* 1999). However, a similar number of studies have failed to detect obvious clastogenic effects following microwave irradiation of isolated animal cells *in vitro* (Alam *et al.,* 1978; Lloyd *et al.,* 1984, 1986; Wolff *et al.,* 1985; Meltz *et al.,* 1987, 1989, 1990; Kerbacher *et al.,* 1990; Maes *et al.,* 1997, 2001). Thus, there is still no conclusive answer as to whether exposure to microwaves is clastogenic, *i.e.*, whether they can direct or indirectly increase the frequency of chromosomal aberrations.

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A further question is whether microwaves can act as epigenetic factors to influence the genotoxicity of other environmental "pollutants" (Maes *et al.,* 2001). Cancer is generally considered to be initiated by alterations in DNA. However, some non-genotoxic chemicals and processes (known as epigenetic carcinogens) are unable to damage DNA and are usually not clastogenic *in vitro*, but can enhance the progress of cells towards malignancy *in vivo.* Several studies have suggested that radiofrequency radiation (RF) has an epigenetic effect *in vivo*, and can enhance the genotoxic effects of ionizing radiation or cancer-inducing substances, or potentiate other epigenetic factors (ICNIRP, 1998).

The aim of this study was to investigate the clastogenic effects of 2.5 and 10.5 GHz microwave fields, alone and in combination with ionizing radiation, on peripheral blood lymphocytes. The combination of microwaves with ionizing radiation ("synergy" test) was designed to screen for joint effects of those two types of radiation. For this, we assessed whether blood samples pretreated with microwaves would be more sensitive (or resistant) to damage by gamma radiation. In all cases, the chromosomal damage was assessed using conventional cytogenetic techniques. The chromosome aberration test is often used to identify physical or chemical agents that cause structural chromosomal aberrations in cultured mammalian cells (OECD, 1997).

Materials and Methods

The microwave sources used were: (a) a 2450 MHz microwave thermal oven (model MARS 5, CEM Corporation) with a power output of up to 1200 W and controls for regulating power and temperature by ventilation, and (b) a 10.5 GHz, 15 mW, linearly polarized, non-thermal microwave source (model WA-9314B, PASCO). The gamma radiation source was cobalt-60 (0.034 $Gy.min^{-1}$), with the absorbed dose being 1.5 Gy (4.5 mJ) per blood aliquot.

Initially, whole blood samples were exposed to the microwave sources for varying periods of time in order to determine the best exposure time for each source. Long periods of exposure resulted in a high cell mortality, seen as a high rate of cell lysis and a low number of metaphases after culturing. Based on these preliminary experiments, exposure times of 40 s at 3 W for the 2.5 GHz oven, and 5 min for the 10.5 GHz device were used. To prevent overheating in the 2.5 GHz oven, the temperature of the blood samples was kept below 36 °C, (starting temperature was 28 °C and reached 33 °C after 40 s of exposure). In the case of the 10.5 GHz device, initial tests showed that there was no increase in the temperature of water samples, even after hours of exposure to this device.

The energies transmitted to each blood sample were calculated to be 75,310 and 230 mJ for the 2.5 and 10.5 GHz sources, respectively. When expressed as the specific energy absorption rate (SAR), these energies corresponded to 626.67 $W.kg^{-1}$ and 0.25 $W.kg^{-1}$, respectively. The SAR expresses the energy absorbed and is a function of the power absorbed in the sample (in Watts) per kg of sample mass.

A 10 mL blood sample was collected into a heparinized vacutainer and immediately divided into six blood aliquots. An equal volume of culture medium (1.5 mL) without phytohemaglutinin (PHA) was added to each blood aliquot before the treatment (irradiation with microwaves and/or gamma radiation). One 3 mL aliquot served as the untreated control, another served as the 1.5 Gy gamma-irradiated control, and the remaining aliquots were treated with microwaves, with or without subsequent 1.5 Gy gamma irradiation. All aliquots were held at 37 °C during the irradiations and incubations. A 2 h interval was allowed between the treatment with microwaves and exposure to gamma radiation. All control samples were handled in the same way as the exposed ones, but without exposure to microwaves or radiation.

Lymphocytes from all blood samples were cultured under identical conditions using standard methods (IAEA, 2001), with modifications. Briefly, 10 mL of Ham's F-10 medium (Cultilab, Campinas, SP, Brazil) supplemented with 25% fetal calf serum (Cultilab) and 0.5 mL of phytohemagglutinin M (Gibco - Invitrogen Corporation, Carlsbad, CA, USA) was used. The cells were incubated for 48 h and 0.04 mg of colchicine (Sigma Chemical Co., St. Louis, MO, USA) was added 3 h before harvesting. After treatment with hypotonic saline solution (0.075 M KCl) for 15 min, the lymphocytes were fixed in methanol:acetic acid (3:1, v/v) and transferred to clean microscope slides followed by staining with 3% Giemsa.

The chromosome aberration test was done using blood samples from four healthy volunteers, both sexes, different ages and not under the use of medications (age and sex in parentheses): donor 1 (44 y, F), donor 2 (28 y, M), donor 3 (23 y, F) and donor 4 (36 y, F). After exposure of the blood aliquots to the different treatments, phytohemagglutinin-stimulated (48 h) lymphocyte cultures were started to obtain chromosomal preparations. The samples were scored blind, except during the initial experiments to estimate the appropriate exposure times, during which the viability of the cultures was evaluated.

In all of the experiments, the maximum possible number of cells per sample was scored using a Nikon Labophot light microscope. Based on the guidelines for the *in vitro* mammalian chromosome aberration test issued by the OECD (1997), at least 200 well-spread metaphases per sample were scored for structural chromosome- and chromatid-type aberrations. The frequency of polyploid cells was also examined since an increase in polyploidy may indicate that a chemical or physical agent has the potential to induce numerical aberrations.

The results were expressed as the aberration yield (Y $\pm$ S.E.), with the standard errors calculated using a Poisson

distribution. The distribution of aberrations among the scored cells was tested for conformity to the Poisson distribution. Values of *U* higher than 1.96 indicated that the distribution was overdispersed (IAEA, 2001). All statistical comparisons were done using the Kruskal-Wallis *H*-test, within 95% confidence limits.

Results

Table 1 shows the chromosome- and chromatid-type aberrations seen among lymphocytes from blood samples exposed to the microwave fields. There was no significant difference between control cells and those exposed to microwave fields. Control cells had a mean frequency of 0.013 aberrations per cell compared to 0.010 and 0.011 aberrations per cell in the microwave-exposed samples. Statistical comparison of these results using the Kruskal-Wallis *H*-test revealed no significant differences within 95% confidence limits. The distribution of the aberrations among cells is shown in Table 2. Subject 1 had a somewhat higher than normal and overdispersed frequency of aberrations, probably because of previous partial-body irradiation. This donor was therefore not used in subsequent experiments (Table 3).

Table 3 shows the results of the "synergy" test. Cells were exposed to microwaves and subsequently to 1.5 Gy of ^{60}Co gamma radiation. There was no significant difference between microwave-treated or non-treated cells. Gamma irradiated cells showed a mean frequency of 0.279 aberrations per cell compared to 0.343 and 0.310 aberrations per cell in samples pretreated with microwaves. Statistical comparison of these results using the Kruskal-Wallis *H*-test revealed no significant differences within 95% confidence limits. The distribution of the aberrations among the cells scored is shown in Table 4. Acentric fragments tended to be slightly overdispersed, as normal (IAEA, 2001).

Table 5 summarizes the results of the numerical aberrations observed according to the different treatments. Again, there were no significant differences among the various groups, according to the Kruskal-Wallis *H*-test, within 95% confidence limits.

Discussion

Following exposure to microwaves from both sources, there was a high rate of cell mortality that increased with the amount of energy transferred to the cells. This mortality was reflected in the high degree of cell lysis and the low number of metaphases after culturing. For blood samples treated in the 2450 MHz oven, the cell lysis was attributed to thermal effects ("cooking"). This phenomenon was also observed by Lloyd *et al.* (1984) in similar experiments. To prevent hyperthemia in the present experiments, the temperature of the blood samples was kept below 36 °C (the starting temperature was 28 °C and reached 33 °C after 40 s of exposure). However, in the case of the 10.5 GHz device, no thermal effects were observed since there was no increase in the temperature of the water samples, even after hours of exposure to this device. Thus, the high level of cell lysis and mortality seen following exposure to the 10.5 GHz device was attributable to other non-thermal processes.

Table 1 - Number and frequencies of structural chromosomal aberrations in human lymphocytes exposed to microwave fields.

Treatment/ subject	Cells scored	Chromosome aberration			Chromatid damage		Overall frequency of aberrations
		Dicentrics	Centric rings	Acentrics	Gaps	Breaks	
2.5 GHz	1402						0.010 ± 0.003
1	900	5 (0.006 ± 0.003)	-	7 (0.008 ± 0.003)	-	1 (0.001 ± 0.001)	
2	502	-	-	1 (0.002 ± 0.002)	-	-	
10.5 GHz	1562						0.011 ± 0.003
1	526	6 (0.011 ± 0.005)	-	7 (0.013 ± 0.005)	4 (0.008 ± 0.004)	-	
2	595	-	-	-	1 (0.002 ± 0.002)	-	
4	441	-	-	-	-	-	
Controls	1286						0.013 ± 0.003
1	471	4 (0.008 ± 0.004)	2 (0.004 ± 0.003)	6 (0.013 ± 0.005)	-	-	
2	547	-	1 (0.002 ± 0.002)	-	1 (0.002 ± 0.002)	1 (0.002 ± 0.002)	
4	268	-	-	1 (0.004 ± 0.004)	-	1 (0.004 ± 0.004)	

Table 2 - Distribution of the chromosome-type structural aberrations among the scored cells indicated in Table 1 and according to the treatment given.

Treatment/ subject	Aberrations	Number of cells with X chromosomal aberrations				*U* value
		0	1	2	3	
2.5 GHz 1	Dicentrics	896	3	1	-	9.39[a]
	Centric rings	900	-	-	-	-
	Acentrics	893	7	-	-	-0.15
2.5 GHz 2	Dicentrics	502	-	-	-	-
	Centric rings	502	-	-	-	-
	Acentrics	501	1	-	-	-
10.5 GHz 1	Dicentrics	520	6	-	-	-0.17
	Centric rings	526	-	-	-	-
	Acentrics	521	3	2	-	9.82[a]
10.5 GHz 2	Dicentrics	595	-		-	-
	Centric rings	595	-	-	-	-
	Acentrics	595	-	-	-	-
10.5 GHz 4	Dicentrics	441	-	-	-	-
	Centric rings	441	-	-	-	-
	Acentrics	441	-	-	-	-
Control 1	Dicentrics	468	2	1	-	8.76[a]
	Centric rings	469	2	-	-	-0.05
	Acentrics	467	2	2	-	11.04[a]
Control 2	Dicentrics	547	-	-	-	-
	Centric rings	546	1	-	-	-
	Acentrics	547	-	-	-	-
Control 4	Dicentrics	268	-	-	-	-
	Centric rings	268	-	-	-	-
	Acentrics	267	1	-	-	-

[a]Overdispersed.

Table 4 - Distribution of the chromosome-type structural aberrations among the scored cells in Table 3.

Treatment/ subject	Aberrations	Number of cells with X chromosomal aberrations					*U* value
		0	1	2	3	4	
2.5 GHz + γ 2	Dicentrics	250	45	5	-	-	0.02
	Centric rings	288	12	-	-	-	-0.47
	Acentrics	267	29	3	-	1	4.16[a]
2.5 GHz + γ 3	Dicentrics	313	36	4	-	-	0.81
	Centric rings	346	7	-	-	-	-0.24
	Acentrics	305	41	7	-	-	1.36
10.5 GHz + γ 2	Dicentrics	243	39	1	-	-	-1.12
	Centric rings	277	6	-	-	-	-0.23
	Acentrics	247	32	3	1	-	1.83
10.5 GHz + γ 3	Dicentrics	212	35	2	-	-	-0.57
	Centric rings	246	3	-	-	-	-0.11
	Acentrics	233	12	3	1	-	5.63[a]
γ only 2	Dicentrics	267	44	2	-	-	-0.85
	Centric rings	307	6	-	-	-	-0.22
	Acentrics	283	26	3	1	-	2.98[a]
γ only 3	Dicentrics	264	38	2	-	-	-0.50
	Centric rings	300	4	-	-	-	-0.14
	Acentrics	277	25	2	-	-	0.58

[a]Overdispersed.

Table 3 - Number and frequencies of structural chromosomal aberrations in human lymphocytes exposed to microwave fields and subsequently to 1.5 Gy of gamma radiation.

Treatment/ subject	Cells scored	Chromosomal aberration			Chromatid damage		Overall frequency of aberrations
		Dicentrics	Centric rings	Acentrics	Gaps	Breaks	
2.5 GHz + γ	653						0.343 ± 0.023
2	300	55 (0.183 ± 0.025)	12 (0.040 ± 0.016)	39 (0.130 ± 0.021)	5 (0.017 ± 0.008)	3 (0.003 ± 0.003)	
3	353	44 (0.125 ± 0.019)	7 (0.020 ± 0.008)	55 (0.156 ± 0.021)	3 (0.085 ± 0.016)	1 (0.003 ± 0.003)	
10.5 GHz + γ	532						0.310 ± 0.024
2	283	41 (0.145 ± 0.023)	6 (0.021 ± 0.009)	41 (0.145 ± 0.023)	1 (0.003 ± 0.003)	1 (0.003 ± 0.003)	
3	249	39 (0.157 ± 0.025)	3 (0.012 ± 0.007)	31 (0.124 ± 0.022)	2 (0.008 ± 0.006)	-	
γ only	617						0.279 ± 0.021
2	313	48 (0.153 ± 0.022)	6 (0.019 ± 0.008)	35 (0.112 ± 0.019)	2 (0.006 ± 0.004)	-	
3	304	42 (0.138 ± 0.021)	4 (0.013 ± 0.007)	29 (0.095 ± 0.018)	3 (0.010 ± 0.006)	3 (0.010 ± 0.006)	

Table 5 - Number and frequencies of chromosomal aberrations in human lymphocytes according to the different treatments. Samples from the different subjects were pooled for each treatment.

Treatment	Cells scored	Euploidy		
		Triploidy	Tetraploidy	Polyploidy
Controls	1286	1	5	-
		(0.001 ± 0.001)	(0.004 ± 0.002)	-
2.5 GHz	1402	5	5	-
		(0.004 ± 0.002)	(0.004 ± 0.002)	-
MW 10.5 GHz	1562	-	1	-
		-	(0.001 ± 0.001)	-
Gamma	617	2	2	-
		(0.003 γ 0.002)	(0.003 γ 0.002)	-
MW 2.5 GHz + γ	653	-	1	-
		-	(0.001 γ 0.001)	-
MW 10.5 GHz + γ	532	-	-	-
		-	-	-

There were no significant differences in the frequencies of chromosomal aberrations between microwave-treated or untreated samples, despite the fact that samples treated with microwaves received huge amounts of transferred energy that were 50-17,000 times greater than the energy transferred by 1.5 Gy of ionizing radiation. The intracellular targets for ionizing radiation are the chromosomes in the nucleus (IAEA, 2001). The results shown here suggest that microwaves do not interact directly or indirectly with chromosomes, although they may target other cell structures, such as cell membranes. This would explain the high degree of lysis seen in the microwave experiments.

Although direct genetic effects from microwave exposure were not expected to occur, indirect effects of microwaves would be more likely, because of the influence of eletromagnetic fields on the free radical system (Maes *et al.,* 1997), but this was not seen in the present chromosome aberration test. These findings agree with other reports showing that the microwave irradiation of human lymphocytes *in vitro* has no direct or indirect clastogenic effects (Lloyd *et al.,* 1984; Maes *et al.,* 1997). In addition to this lack of a direct or indirect effect of microwaves on chromosomes, pretreating cells with microwaves also failed to affect their sensitivity to ionizing radiation.

A long-standing dogma in radiation science has been that energy from radiation must be deposited in the nucleus to elicit its biological effects. In recent years, a number of epigenetic effects have been described that challenge this dogma. Epigenetic factors, although not themselves genotoxic, act synergistically to enhance the carcinogenic effects of other agents. Several studies (Szmigielski *et al*., 1982; Scarfi *et al*., 1996; Maes *et al*., 1997; Pakhomova *et al.*, 1997) have suggested that microwaves can have an epigenetic effect *in vivo*, and that they can exacerbate the genotoxicity of ionizing radiation or cancer-inducing substances, or potentiate other epigenetic factors (IEGMP, 2000). However, the evidence for an epigenetic effect of microwaves is equivocal since some studies have failed to reproduce the positive results reported by others (Ciaravino *et al*., 1987, 1991; Meltz *et al*., 1989, 1990; Cain *et al*., 1997).

In conclusion, the results described here do not support the hypothesis that microwaves enhance the direct effect of gamma radiation or cause cells to respond differently to ionizing radiation *in vitro*. It is possible that some of the epigenetic responses to microwaves *in vivo* could be the result of thermal effects (IEGMP, 2000), as concluded by Pakhomova *et al.* (1997), who found that high frequency microwaves (61 GHz) enhanced DNA recombination, but not mutagenesis, in yeast cells exposed to ultraviolet radiation. Our findings indicate that further investigations are needed to examine the influence of microwaves *in vivo* and *in vitro*.

Acknowledgements

The authors thank colleagues at the IME and IRD/CNEN for their help and suggestions, and also Dr. David Lloyd (NRPB, UK) for useful comments.

References

Alam MT, Barthakur N, Lambert NG and Kasatiya SS (1978) Cytological effects of microwave radiation in Chinese hamster cells *in vitro*. Can J Genet Cytol 20:23-28.

Balode Z (1996) Assessment of radio-frequency electromagnetic radiation by the micronucleus test in bovine peripheral erythrocytes. Sci Total Environ 180:81-86.

Cain CD,Thomas DL and Adey WR (1997) Focus formation of C3H/10$T_{1/2}$ cells and exposure to a 836.55 MHz modulated radiofrequency field. Bioelectromagnetics 18:237-243.

Ciaravino V, Meltz ML and Erwin DN (1987) Effects of radiofrequency radiation and simultaneous exposure with mytomicin C on the frequency of sister chromatid exchanges in Chinese hamster ovary cells. Environ Mutagen 9:393-398.

Ciaravino V, Meltz ML and Erwin DN (1991) Absence of a synergistic effect between moderate-power-radiofrequency electromagnetic radiation and adriamycin on cell-cycle progression and sister-chromatid exchange. Bioelectromagnetics 12:289-294.

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

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

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

Goswami PC, Albee LD, Parsian AJ, Baty JD, Moros EG, Pickard WF, Roti Roti JL and Hunt CR (1999) Proto-oncogene mRNA levels and activities of multiple transcription factors in C3H 10T 1/2 murine embryonic fibroblasts exposed to 835.62 and 847.74 MHz cellular telephone communication frequency radiation. Radiat Res 151:300-309.

Haidler T, Knasmueller S, Kundi M and Haidler M (1994) Clastogenic effects of radiofrequency radiations on chromosomes of Tradescantia. Mutat Res 324:65-71.

IAEA (2001) International Atomic Energy Agency. Cytogenetic Analysis for Radiation Dose Assessment: A Manual. Technical Report Series 405, Vienna, 127 pp.

ICNIRP (1998) International Committee on Non Ionising Radiation Protection. Guidelines for limiting exposure to time-varying electric, magnetic, and electromagnetic fields (up to 300 GHz). Health Phys 74:494-522.

IEGMP (2000) Independent Expert Group on Mobile Phones. Mobile Phones and Health. NRPB publication, Chilton, UK, 159 pp.

Ivaschuk OI, Jones RA, Ishida-Jones T, Haggren Q, Adey WR and Phillips JL (1997) Exposure of nerve growth factor-treated PC12 rat pheochromocytoma cells to a modulated radiofrequency field at 836.55 MHz: Effects on c-jun and c-fos expression. Bioelectromagnetics 18:223-229.

Kerbacher JJ, Meltz ML and Erwin DN (1990) Influence of radiofrequency radiation on chromosome aberrations in CHO cells and its interaction with DNA-damaging agents. Radiat Res 123:311-317.

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

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

Lloyd DC, Saunders RD, Finnon P and Kowalczuk CI (1984) No clastogenic effect from *in vitro* microwave irradiation of G_0 human lymphocytes. Int J Radiat Biol 46:135-141.

Lloyd DC, Saunders RD, Moquet JE and Kowalczuk CI (1986) Absence of chromosomal damage in human lymphocytes exposed to microwave radiation with hyperthermia. Bioelectromagnetics 7:235-240.

Maes A, Collier M, van Gorp U, Vandoninck S and Verschaeve L (1997) Cytogenetic effects of 935.2-MHz (GSM) microwaves alone and in combination with mitomycin C. Mutat Res 393:151-156.

Maes A, Collier M and Verschaeve L (2001) Cytogenetic effects of 900 MHz (GSM) microwaves on human lymphocytes. Bioelectromagnetics 22:91-96.

Maes A, Verschaeve L, Arroyo A, de Wagner C and Vercruyssen L (1993) *In vitro* cytogenetic effects of 2450 MHz waves on human peripheral blood lymphocytes. Bioelectromagnetics 14:495-501.

Meltz ML, Eagan P and Erwin DN (1989) Absence of mutagenic interaction between microwaves and mitomycin C in mammalian cells. Environ Mol Mutagen 13:294-299.

Meltz ML, Eagan P and Erwin DN (1990) Proflavin and microwave radiation: Absence of a mutagenic interaction. Bioelectromagnetics 11:149-154.

Meltz ML, Walker KA and Erwin DN (1987) Radiofrequency (microwave) radiation exposure of mammalian cells during UV-induced DNA repair synthesis. Radiat Res 110:255-260.

OECD (1997) Organisation for Economic Cooperation and Development. OECD Guidelines for the Testing of Chemicals, Guideline 473: *In vitro* Mammalian Chromosome Aberration Test, Paris, 10 pp.

Pakhomova ON, Pakhomov AG and Akyel Y (1997) Effect of millimeter waves on UV-induced recombination and mutagenesis in yeast. Bioelectrochem Bioenerg 43:227.

Phillips JL, Ivaschuk O, Ishida-Jones T, Jones RA, Campbell-Beachler M and Haggnen W (1998) DNA damage in molt-4 T-lymphoblastoid cells exposed to cellular telephone radiofrequency fields *in vitro*. Bioelectrochem Bioenerg 45:103-110.

Sagripanti J and Swicord ML (1986) DNA structural changes caused by microwave radiation. Int J of Radiat Biol 50:47-50.

Sarkar S, Ali S and Behari J (1994) Effect of low power microwave on the mouse genome: A direct DNA analysis. Mutat Res 320:141-147.

Scarfi MR, Lioi MB, dÁmbrosio G, Massa R, Zeni O, De pietro R and De Berardino D (1996) Genotoxic effects of mytomicin-C and microwave radiation on bovine lymphocytes. Electro-Magnetobiology 15:99-104.

Szmigielski S, Szudzinski A, Pietraszek A, Bielec M andWrembel JK (1982) Accelerated development of spontaneous and benzopyrene-induced skin cancer in mice exposed to 2450-MHz microwave-radiation. Bioelectromagnetics 3:179-185.

Tice R, Hook ± and McRee DI (1999) Chromosome aberrations from exposure to cell phone radiation. Microwave News 7-8:7.

Timchenko OI and Ianchevskaia NV (1995) The cytogenetic action of electromagnetic fields in the short-wave range. Psychopharmacol Ser 7-8:37-39.

Verschaeve L, Slaets D, Van Gorp U, Maes A and Vanderkom J (1994) *In vitro* and *in vivo* genetic effects of microwaves from mobile phone frequencies in human and rat peripheral blood lymphocytes. Proceedings of Cost 244 Meetings on Mobile Communication and Extremely Low Frequency

Field: Instrumentation and Measurements in Bioelectromagnetics Research. D. Simunic (ed), Information Ventures Inc., Plzen, Chech Republic, pp 74-83.

Vijayalaxmi BZ, Frei MR, Dusch SJ, Guel V, Meltz ML and Jauchem JR (1997) Frequency of micronuclei in the peripheral blood and bone marrow of cancer-prone mice chronically exposed to 2450 MHz radiofrequency radiation. Radiat Res 147:495-500.

Wolff S, James TL, Young GB, Margulis AR, Bodycote J and Afzal V (1985). Magnetic resonance imaging: Absence of *in vitro* cytogenetic damage. Radiology 155:163-169.

Associate Editor: Carlos F.M. Menck