Abstract. The question whether exposure to RF radiation poses a non-thermal risk factor for cancer has awakened much interest among researchers and in the general population, especially due to the use of mobile phones. In an attempt to answer this entangling question we have examined whether in-vitro exposure of human peripheral blood lymphocytes (PBL) to CW 800MHz (RF) electromagnetic fields, associated with second generation cellular phones, leads to an increase in the level of aneuploidy - the presence of an improper number of chromosomes, a phenomenon that is associated with carcinogenesis. As the ICNIRP guidelines are based on the assumption that the RF induced biological consequences are due to thermal effects, we have taken precautions to perform the experiments under conditions where the temperature of the exposed samples did not exceed the range 36.5-37.5°C.

We exposed PBL to RF at 800 MHz in specially designed exposure set-up, consisting of a custom-made waveguide resonator, at specific absorption rates (SAR) of 5.3 and 7.4 W per kg. The numerical analysis of the distribution of the SAR levels was complemented by experimental measurement of the initial heating rate by optical fiber based thermometry.

All cultures were harvested, using standard cytogenetic procedures, 72 hours after culture set up. In 10 experiments we exposed the samples for the complete duration of 72 hours of culturing. In five experiments we exposed the samples for 24 hours, on three different days during the overall duration of 72 hours. While series one (I) was exposed for the first 24 hours of culturing; series two (II) was exposed during the middle of the culturing period and series three (III) was exposed for the final 24 hours of culturing. The incubator housing the experimental set-up was maintained in a humid atmosphere of 5% CO₂, set to 33.5°C.

The level of aneuploidy was determined in interphase nuclei by applying fluorescence in situ hybridization (FISH). We performed two color FISH using two probe combinations: We used Vysis Inc (USA) probes recognizing the centromers of either chromosomes 11 and 17 or chromosomes 1 and 10 with the orange and green fluorophores, respectively. The level of aneuploidy is given as the proportion of cells with less then the expected two signals (monosomy) or more then the expected two (multisomy). All ten samples were analyzed with the probes for chromosomes 11 and 17. In addition, five of the samples exposed for 72 hours, randomly chosen, were also analyzed with the probes for chromosomes 1 and 10. We used four additional volunteers in order to ascertain that the increased aneuploidy levels are not due to thermal effects, by setting up concurrent cultures at four temperatures between 33.5°C and 37°C.

Images of the hybridized nuclei were acquired using an automated image analysis and scoring system (Metafer 4, Metacyte, Germany). To the obtained galleries we performed manual correction, analyzing about 1500 nuclei per sample.
The results are summarized in Figure 1. The 72 hour exposure led to increased aneuploidy of chromosome 1 and to a nearly significant increase of chromosomes 10, 11 and 17. Exposure for 24 hours during different days led to the most pronounced increase for the last exposure day. Exposure for 24 hours led to a more pronounced aneuploidy when compared to the exposure for 72 hours. When comparing aneuploidy of chromosomes 11 and 17 following exposure for 24 hours with those of 72 hours reveals that the shorter exposure led to a higher level of aneuploidy. We could demonstrate that these effects are not due to temperature elevation since the aneuploidy levels of chromosomes 11 and 17 were not affected by temperature increase between 33.5°C and 40°C. Thus the effect described as caused by the RF is of a non-thermal nature.

Our findings indicate that the exposure to CW 800 MHz at 5.3 and 7.4W per kg increases chromosomal instability. We assume that changes occur due to a compromised mitotic checkpoint that allows the existence and thriving of aneuploid cells. These findings should be taken into consideration in the future evaluation of guidelines for exposure.

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