News from Science

The following paragraphs deal with recent original scientific studies investigating effects of mobile radio frequency fields. The publications were selected by the author, Prof. Roland Glaser, based on his personal preferences.

Roland Glaser

Does modulation of radiofrequency fields matter in potential bioeffects? This much-discussed, very important question was the subject of a commentary prepared of two scientists who have demonstrated their expertise in this area in many publications: Kenneth R. Foster and Michael H. Repacholi.

The article is written from the perspective of the WHO and basically focuses on two questions: Do modulation parameters have to be considered when defining limits? What research recommendations would be appropriate to obtain clarity as quickly and safely as possible and by rationally applying research means? Both is relevant regarding the risk assessment of the WHO planned for 2006/2007.

At first it is stated that mobile phones, except perhaps in the case of induction by currents of their operating system, do not transmit low-frequency fields, despite pulsation. A demodulation of varying power pulses at 217 Hz would be possible at best caused by thermal variations within this frequency range. We know that microwave hearing is based on such an effect, which can occur at strong radar pulses, but not due to mobile radio fields.

The defenders of the opinion that modulated RF fields have a specific non-thermal effect, basically use the theory of **H. Fröhlich** on coherent oscillations, but also on the publication of S. M. Bawin et al. from 1975 (Ann. N.Y. Acad. Sci. **24**, 774). It reports a sensitive 16 Hz modulation window of 147 MHz fields which is said to lead to an increased calcium release in the brain of chicken. Although these findings could not be reproduced over the past 25 years, and although nobody knows what the physiological relevance of this unspecific effect would be, if it exists at all, this study is repeatedly cited and obviously used as principal witness of the alleged window effects of modulation frequencies. The situation for the theory of coherent oscillations is similar.

The study of **R.K. Adair** (Biophys. J. **821**, 147. 2002), which demonstrated that such resonance effects caused by the damping in water systems, are not possible, is simply negated. Summaries of more re-

cent studies investigating specific effects of modulated RF fields of the NRPB (National Radiation Protection Board) from the years 2001 and 2003 came to th conclusion that such effects have not been fully proven yet in experiment.

There is clear knowledge, however, about special thermal effects of extremely strong RF pulses from military research experiments. But these can be neglected for the time being. In spite of this, there are repeated findings of modulation specific effects of weak fields, especially in Russian literature. What to do? What should WHO recommend? An optimum compromise has to be found between the unlimited number of possible variations of carrier and modulation frequencies (and types), limited research means, and, ultimately, required replications of relevant experimental results.

Unfortunately, a theoretical basis lacks, or at least a working hypothesis which could serve as guidance for the recommendation of relevant experiments to perform. Ultimately, we should concentrate on world wide applied frequencies and modulations (resp. pulsations) for the time being, e.g. the field of the GSM system. The probability of finding such specifics, however, is small when looking at prior experiences *(Foster, K.R. and Repacholi, M.H.: Biological effects of radiofrequency fields: Does modulation matter? Radiat. Res. **162**, 219-225. 2004).*

Are pregnant women put at risk from mobile phone radiation? Could RF fields lead to malformations in the fetus? This question was the subject of study of a Japanese working group at Tohoku University. They exposed mice from the first day of pregnancy until birth and examined the generation exposed in utero at the age of 10 weeks. Tested were extracts from spleen, liver and brain examined for mutations by means of the determination of the lacZ reporter gene established at the Institute. Exposure of the pregnant animals was performed daily, 16 hours, at 2.45 GHz. At continuous field exposure, the SAR was 4.25 W/kg leading to an increase in rectal temperature by 1.38° after 16 hours. Other animals were exposed only 20 or 10 sec per minute leading to SARs of 1.4 W/kg resp. 0.71 W/kg, accordingly. These experiments could not prove any genetic change.

However, we have to take into account that, due to the smallness of the animals, embryos were actually fully exposed to the fields, which would be not the case in humans, due to the small penetration depth of fields of these frequencies *(Ono, T.; Saito, Y.; Komura, J.; Ikehata, H.; Tarusawa, Y.; Nojima, T.; Goukon, K.; Ohba, Y.; Wang, J.Q.; Fujiwara, O., and Sato, R.: Absence of mutagenic effects of 2.45 GHz radiofrequency exposure in spleen, liver, brain, and testis of lacZ-transgenic mouse exposed in utero. Tohoku J. Exp. Med. **202**, 93-103. 2004).*

Similar to the Japanese group around Ono et al., also a multidisciplinary working group at the University of Thessaloniki (Greece) dealt with the question whether the fields of mobile radio can have **effects on the development of embryos in utero**. However, due to different body sizes and resonance frequencies, they found it necessary to apply fields with higher frequencies in experiments with rats so they might be able to extrapolate results to humans. Therefore the animals were exposed to pulsed 9.4 GHz fields (20 µs pulse length, 50 Hz pulse frequency). The cages with the freely moving test animals were placed inside closed chambers, in the middle between two Horn antennas with parabolic reflectors positioned at a 10 m distance from each other. SARs of 0.5 mW/kg were calculated for a power flux density of 5 µW/cm².

Besides 20 control animals, 25 resp. 26 pregnant female rats were exposed once from the 1st to the 3rd day, and from the 4th to 7th day post coitum. Right after birth, the young animals were sacrificed, examined macroscopically for malformations, and the kidneys were tested immunohistologically and biochemically. Malformations could not be proven in any of the newborn animals.

A semi-quantitative control of kidney slices for a particular growth factor BMP-4 (BMP = “bone morphogenic protein”) showed an increase for both exposure conditions compared to controls. The same fac-

tor could also be shown to be significantly increased by eletrophoresis. The authors do not give an answer to the question whether this change is relevant to health and neither present a theory on possible physiological interaction mechanisms resp. targets of fields.

Since a penetration depth of less than 5 mm is expected for this frequency, an indirect effect of fields caused by neurohumoral activation of the exposed brain cannot be excluded. Blinded analysis is mentioned only once in the publication, in association with electrophoretic tests. But in the case of "semi-quantitative" histological tests, double-blindness would be especially important. You may also doubt whether one can conclude effects within the frequency range of mobile radio from different resonant frequencies between humans and rats in 9.4 GHz tests *(Pyrpasopoulou, A.; Kotoula, V.; Cheva, A.; Hytirogiou, P.; Nikolokaki, E.; Magras, I.N.; Xenos, T.D.; Tsiboukis, T.D., and Karkavelas, G.: Bone morphogenetic protein expression in newborn rat kindeys after prenatal exposure to radiofrequency radiation. Bioelectromagnetics **25**, 216-227. 2004).*

A Russian working group examined **possible genetic effects of ultrashort microwave pulses (8.8 GHz, 180 ns, 50 Hz pulse sequence)**. Tested were red blood cells of the claw frog containing a nucleus. DNA fragments were identified by means of the alkaline Comet Assay. Since there are no comparable data yet on the sensitivity of this system in literature, positive controls were performed with gamma radiation (up to 200 cGy) and chemical mutagens (etyl methane sulfonate = EMS, 5 mM). There were also temperature controls at 20°, 25° and 30° C. Both the ionizing radiation (from 50 cGy) and EMS showed clear effects. Even the temperature increase led to significant changes in the Comet Assay. The changes which could be revealed in the cells caused by 40-min exposure to the MW pulses were small though and were similar to controls at accordingly increased temperatures. A mean SAR of 1.6 W/kg could be calculated from the measured temperature changes, which equals a SAR of 300 MW/kg during the 180 ns *(Chemeris, N.K; Gapeyev, A.B.; Sirota, N.P. et al.: DNA damage in frog erythrocytes after in vitro exposure to a high peak-power pulsed electromagnetic field. Mutation Res., Genetic Toxicol. Environ. Mutagen. **558**, 27-34. 2004).*

The Dutch Health Council denies the assumption that children are at a higher risk from mobile radio fields. The warning against exposure of children came from a study of the English "Independent Expert Group on Mobile Phones" (IEGMP) from the year 2000 leading to a corresponding recommendation of the UK Department of Health in 2002.

This led to world wide discussion and the request for public measures. But what is the basis of this assumption? The paper once claims that early exposure would lead to a longer storage of potential effects over the course of life. Apart from the fact that such storage has not yet been proven, it would be insignificant considering the relation of the respective life period with the whole lifetime. On the other side, children's tissue, especially the brain, is claimed to have an up to twofold absorption capacity of radiofrequency fields. At closer inspection, this assessment is based only on a publication of C. Gabriel from the year 2000, where respective impedance differences are listed. Although both increased conductivity of brain tissue in the first years of life and an increase in bone thickness in the first 12 years of life of children are undeniable, the numbers given in this study ("A 1 year old could absorb around double, a 5 year old around 60%, more than an adult") are not reproducible in detail. Basically, there is no scientific basis justifying the aforementioned recommendation from the year 2002, according to the Dutch Electromagnetic Fields Committee (van Rongen, E.; Roubos, E.W.; van Aernsbergen, L.M. et al.: Mobile phones and children: Is precaution warranted? Bioelectromagnetics **25**, 142-144. 2004).

There is a "Letter to the editor" referring to this publication of the Chair of the Russian National Committee for Non-Ionizing Radiation Protection, Youri Grig-

oriev, as well as a reply by Eric van Rongen (Bioelectromagnetics **25**, 322-323, 2004).

Grigoriev challenges the authors of the publication by saying their analysis was purely physical ignoring the world wide experience of physiologists, psychologists, morphologists and pediatricians. In this context, he cites a WHO statement from the year 2003: "Children have a unique vulnerability. As they grow and develop, there are 'windows of susceptability': periods when their organs and systems are particularly sensitive to the effects of certain environmental threats." Many years of experience with chemical toxins and ionizing radiation would emphasize this. The Russian National Committee for Non-Ionizing Radiation Protection therefore had recommended a limit for mobile phone use by children in September 2001. Consequently, these recommendations were included in the standards of the Ministry of Health of the Russian Federation. He criticizes the Netherlands commitee for promoting the tendency to disregard this principle according to the slogan "to each child his/her mobile phone".

In his reply, van Rongen denies this criticism. In his view, the addressed physiological and medical aspects were very well considered, as well as the cited WHO recommendation. He added that there was no scientific basis for the assumption that the sensitivity towards radiofrequency radiation could change after the second year of life. The recommendation of the Steward Committee was a general precautionary measure and not based on scientific data. "One can think of a number of arguments why children should make less use of their mobile phones than they do now, but the fear of health effects lacks any sound scientific basis."

A comprehensive epidemiological case-control study based on the Danish Cancer Registry dealt with the question if there was a **correlation between a specific cancer at acoustic nerve and the use of mobile phones**. This slowly growing cancerous tumor mainly manifests with one-sided hearing loss, accompanied by Tinnitus symptoms. It mainly concerns persons older than fifty years, and occurs three times as often in women as in men. Nevertheless, it is a relatively rare disease concerning an average of 1 to 20 persons per year in a population of 1 million. The study comprised 141 patients who were diagnosed with this disease between 1st September 2000 and 31st August 2002. They were compared to a slightly larger number of control persons, selected by statistical methods from the Danish Central Registry. Age and gender of the population was approximately equal to that in the circle of diseared persons. By written correspondence, trained staff collected information on phone use (in particular: length, frequency, type of device) and other relevant conditions. Moreover, comprehensive socio-economic data were obtained.

In short: Neither correlations between the occurrence, the size or right/left distribution of the cancerous growth and mobile phone use could by found. The odds ratio of the occurrence of this disease was 0.90. We could even conclude that phoning protects against cancer (!), but the 95% margin of this value is between 0.51 and 1.57. According to the limited number of cases treated in this study, the pobability of proving a potential positive correlation between mobile phone use and the occurrence of this type of cancer can be calculated to 75%. The study strikes the eye by the carefully done evaluation and its size *(Christensen, H.C.; Schüz, J.; Kosteljanetz, M.: Poulsen, H.S.; Thomsen, J., and Johansen, C.: Cellular telephone use and risk of acoustic neuroma. Am. J. Epidem.* ***159****, 277-283. 2004).*

Can UV radiaton effects be enhanced by non-thermal field influences from mobile phones? Some years ago, the group around Markkanen reported that 50 Hz fields with a flux density of 0.12 mT are sufficient to enhance UV-induced effects in yeast cells (Bioelectromagnetics **22**, 345-350. 2001).

This time they dealt with continuous and amplitude modulated (217 Hz pulsed) fields with SARs of 0.4 (872 MHz) and 3 W/kg (872 MHz). Aside from a wild-type stem of yeasts *(saccharomyces cerevisae),* a particularly thermosensitive mutant was used, which

spontaneously induces programmed cell death (apoptosis) at 37° C. Two fluorescence markers were applied to prove apoptosis (marking of inverted phosphatidyl serine in the membrane) and uncontrolled cell death (necrosis – cell penetration by propidium iodide). The irradiation of cells with UV light (2 hours, 250 J/m², 280-320 nm) in each case produced an increase in the number of apoptotic cells. At subsequent one hour exposure, this effect could be enhanced only in the case of pulsed fields of both intensities and in the heat-sensitive mutant. The authors see that this as a specific "non-thermal" effect especially of pulsed fields.

However, a few questions remain unresolved: Why did they use a thermosensitive stem in order to prove "non-thermal" effects? What is behind the highly different values of UV impact in different experiments (mean values vary between approx. 6 and 10%). The difference between pulsed and continuous fields in ill. 3 results only from the fact that the compared value (UV) in the first case was considerably smaller than in the second case (did they cells know what was in store for them?). Also, it is doubtful whether water cooling below the exposed petri dish really excludes any local heating of samples. Respective measurements were not given. Unfortunately, these aspects are not discussed in the paper *(Markkanen, A.; Penttinen, P.; Naarala, J.; Pelkonen, J.; Sihvonen, A.P., and Juutilainen, J.: Apoptosis induced by ultraviolet radiation is enhanced by amplitude modulated radiofrequency radiation in mutant yeast cells. Bioelectromagnetics **25**, 127-133. 2004)*.

Can mobile radio fields cause eye lens clouding?

This several times examined issue was reexamined by a Chinese working group. They prepared epithelial cells from the rabbit lens, cultivated them and exposed them after two passages in the course of 8 hours to unpulsed 2.45 GHz fields at power flux densities of 0.1 to 2 mW/cm². From 0.5 mW/cm², significant changes in cell proliferation and protein expression could be determined. The biochemical and cytological methods used in the study were thoroughly described, but, unfortunately, details on exposure conditions are lacking. SARs were not given either – perhaps due to fact that they were difficult to determine. At least some samples were exposed inside wave plates, plastic plates with 96 cylindrical indentations. Although the part on methodology mentions temperature control at 25° and a temperature rise caused by irradiation by less than 0.6°, the authors concede in discussion that local temperatures in the samples could not be measured exactly during and after exposure, and that effects consequently could also be explained thermally.

Regrettably, only publications that hace found radiofrequency effects on cells are cited in the paragrahs "Introduction" and "Discussion". Studies with contrary results were ignored. They also omitted the fact that Steward De Haan et al. 1980 (IEEE MTT-S Int. Symp. 341) and later on Lu et al. (Bioelectromagnetics **21**, 439. 2000), as well as Elder (Bioelectromagnetics 148, 2003), had shown that the lens epithelium of the eye can be damaged only by distinct heating far above permitted limits *(Yao, K.; Wang, K.J.; Sun, Z.H.; Tan, J.; Xu, W.; Zhu, L.J., and Lu, D.Q.: Low power microwave radiation inhibits the proliferation of rabbit lens epithelial cells by upregulating P27 (kip-1) expression. Molecular Vision **10**, 138-143. 2004)*.