Summaries and assessments of selected studies

In the period from November 2014 to January 2015, 113 new publications have been identified, and eight of these were discussed in depth by BERENIS. Based on the selection criteria, five of these publications were selected as the most relevant ones. Their summaries and assessments are provided below.

1) Experimental studies

Extremely low frequency magnetic fields and reproduction of mice (Kim *et al.* 2014)

Kim *et al.* (2014) conducted an *in vivo* exposition study to investigate the impact of extremely low frequency magnetic fields (60 Hz) on the testes of adolescent male mice. Continuous exposure for eight weeks with field strength of up to 200 µT did not have any effect on the increase of body or testicular masses. This indicates that the magnetic field has no negative effects on the growth and development of the organs. However, a remarkable finding of this study is the increase of the amount of apoptotic cells per seminiferous tubule after eight weeks of exposure. This dose-dependent increase was observed already at 2 µT field strength and became significant at 20 µT. The authors also showed that the number of apoptotic cells increases with longer duration of exposure, when exposed to a field with 100 µT. This negatively affects the number of sperm in the epididymis, which could lead to a decrease in fertility in the long term. The authors note that it is primarily spermatogonia, i.e. germ cell populations with mitotic cell division, and not spermatocytes and maturing sperm which die. According to the authors, this indicates an increased sensitivity of germ cells for magnetic field effects. Regrettably, this statement hasn't been backed up by any experimental data. Other weaknesses of the study are the missing blinding, as well as insufficient descriptions of the sample sizes and the analytical protocol for determining the number of apoptotic cells. This makes it rather difficult to evaluate the health relevance of these otherwise interesting observations, and calls for independent replication and expansion of the study without those technical weaknesses.

Extremely low frequency magnetic fields and behaviour of fruit flies (Fedele *et al.* 2014)

Fruit flies can detect the earth's magnetic field and use it for navigation and orientation. Analyses of the behaviour of genetically modified and wild-type fruit flies *Drosophila melanogaster* with exposure to magnetic fields (MF) indicate that the blue-light sensitive photoreceptor protein cryptochrome may control the magnetic sense. There is some evidence that cryptochrome activity involves a radical pairing mechanism that might be sensitive to MF. The *in vivo* study by Fedele *et al.* (2014) shows consistent effects with regard to circadian rhythm and activity levels of the flies after MF exposure (3 and 50 Hz / 300 µT and 1 mT), which differ from previously reported data. The MF exposure in the experimental setup provided for a homogenous magnetic field, with field changes of less than 1% even at high activity levels of the flies. Both exposure chamber and sham chamber were shielded against outside fields. MF exposure of the genetically unmodified flies resulted in a reduction of the circadian period under blue light, and an increase in activity levels at both frequencies and field strengths. With MF exposure under green light, the authors observed an increase of the circadian period. Genetically induced modifications of the C-terminus of the cryptochrome resulted in an attenuation of the MF-induced circadian period changes, while genetic

modifications in the N-terminus of the cryptochrome indicate that this part of the receptor is responsible for the hyperactivity after MF exposure of the flies. Interestingly, mammalian cryptochromes were observed to respond to MF when placed into genetically modified flies, whereas in mammalian neurons, this effect was absent. Therefore, the MF effect is determined by the intracellular environment, suggesting that other (yet unknown) molecules interact with cryptochrome.

Extremely low frequency magnetic fields and cells of the connective tissue (Chen *et al.* 2014)

Chen *et al.* (2014) investigated *in vitro* possible effects of extremely low frequency magnetic fields (50 Hz) on autophagy in specific cells (fibroblasts) in the embryonal connective tissue of mice. Autophagy means "self-eating" and protects the cell from functional disorders by disintegrating defect or even damaging cell components or pathogens. They are isolated in specialised cell organelles, degraded and finally returned into the cell metabolism. Overly stressed or deficient autophagy can lead to diseases. It has been shown that mutations in genes responsible for regulating autophagy cause neurodegenerative diseases such as for example Alzheimer's disease, amyotrophic lateral sclerosis (ALS) and Parkinson's disease with exceptional frequency. In cancer, autophagy has a dual role, acting both as a tumour suppressor by getting rid of damaged proteins and cell components, and as a mechanism of cancer cell survival that can promote the growth of established tumours. The embryonic fibroblasts were exposed to magnetic fields (50 Hz, 2 mT) for 0.5, 2, 6, 12 and 24 hours. A significant increase in the autophagy marker LC3-II was observed after 6 hours of exposure. Two different methods were applied to evaluate autophagy, and positive controls were used. The number of autophagy-induced vacuoles was increased as detected by transmission electron microscopy. Neither the signaling pathway mTOR – which plays a role in autophagy – nor programmed cell death (apoptosis) were activated by this exposure. In this study, an elevation of reactive oxygen species (which are known to induce autophagy) was found after 2 and 6 hours but not after 12 and 24 hours of magnetic field exposure. Further studies are necessary to identify the signalling pathways involved, and in order to clarify whether the absence of the observed effect after longer magnetic field exposure can be explained by a metabolic adaptation.

High frequency electromagnetic fields and blood brain barrier (Tang *et al.* 2015)

Some experimental studies on possible changes of the permeability of the blood brain barrier due to high frequency electromagnetic fields (RF EMF) are available. In their *in vivo* study with male Sprague Dawley rats, Tang *et al.* (2015) report changes in the permeability of the blood brain barrier and cognition after 28 days of exposure with an unmodulated 900 MHz EMF (10 W/m^2), with specific absorption rates (SAR) of 0.016 W/kg (whole body) and 2 W/kg (locally in the brain). Three groups with 36 animals each were studied: sham exposure, 14 days of exposure, and 28 days of exposure. The rats used in the experiment were an outbred strain, and thus had a higher genetic variability than rats from an inbred strain. The spatial memory performance of the animals was examined by applying the "Morris water maze". Morphological changes in the brain of the animals were analysed with ultrastructural methods in two brain regions (hippocampus and neighbouring parietal cortex). HO-1 positive neurons and possible involved signal transduction pathways such as the signal protein ERK were analysed. ERK has been identified in previous studies and it was found to be a relevant signal transduction pathway involved in RF EMF exposure. These signal transduction pathways are important for the regulation of embryogenesis, cell differentiation and cell growth as well as the programmed cell death (apoptosis). The results of this study demonstrate that rats exposed to RF EMF for 28 days show an impaired spatial memory compared to sham-exposed animals. Furthermore, RF EMF exposure induced cellular edema and the degeneration of neuronal cell

organelles. Increased emersion of albumin and HO-1 in both investigated brain regions indicated increased permeability of the blood brain barrier. For the first time, an increased expression of the enzyme MAPK phosphatase was seen, resulting in ERK dephosphorylation, which might result in an interruption of the signal transduction pathway. In summary, the results show an impaired spatial memory and an increased permeability of the blood brain barrier after RF EMF exposure of the rats for 28 days at 900 MHz. A possible mechanism involved could be the mkp-1/ERK signal transduction pathway. These results are contradictory to many studies that have not found an increased permeability of the blood brain barrier when exposed to EMF.

2) Review article

Exposure to radiofrequency electromagnetic fields in Europe (Gajšek *et al.* 2015)

On the basis of published scientific literature, an overview of radiofrequency EMF exposure (10 MHz – 6 GHz) of the European population was compiled in the framework of the EFHRAN project (European Health Risk Assessment Network on EMF Exposure). In this context, Gajšek *et al.* (2015) have published a review article in which they emphasize that the available measurement surveys have been collecting and analysing data in different ways, and are therefore not directly comparable. In total, the authors have identified and analysed nine spot and long term measurement surveys as well as one review article related to such measurements. The average exposure found in those ten surveys was between 0.08 and 1.8 V/m. Surveys using randomly selected measurement locations generally found lower values compared to surveys that had chosen measurement locations close to transmitters. Surveys related to personal exposure originated from nine different countries. Average exposure levels in those surveys was between 0.09 and 0.27 V/m. The values thus tended to be lower compared to spot and long term measurement surveys. Based on the overall data situation, the authors concluded that radio frequency electromagnetic fields in the environment are typically below 1 V/m. They estimated that 1% of the measured values in Europe are above 6 V/m, and 0.1% above 20 V/m. None of the measured data were above the regulatory limits recommended by the EU (28 to 61 V/m, depending on the frequency). This review article highlights typical exposure values in everyday life. Average exposure levels are clearly below the regulatory limits. Unfortunately, the impact of different measurement methods and study site selection criteria on the results of the measurements has not been analysed in depth. It thus remains unclear to which extent the collected data are representative, particularly with regard to spot and long term measurement surveys. Furthermore, it was not described how the proportion of measured values above 6 and 20 V/m has been assessed, and whether these refer to locations where people are present, or to any locations in the environment. The authors note that it is difficult to assess whether there are population groups with higher exposure, and if so, how much higher their exposure is.

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Additional information related to the Swiss expert group on electromagnetic fields and non-ionising radiation (BERENIS) and a list of abbreviations can be found at http://www.bafu.admin.ch/elektrosmog/01095/15189/index.html?lang=en