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Biomarkers in volunteers exposed to mobile phone radiation

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HIGHLIGHTS

- This single blinded randomized counterbalanced study tested whether short term exposure to an 890-MHz phone-like signal affects the integrity of brain-shielding barriers.
- The study had multiple exposure conditions and included 24 healthy subjects aged 18–30 years.
- Biomarkers analyzed in blood serum before and after the three different exposure conditions were used to evaluate potential effects.
- The study failed to show any clinically or statistically significant effect of short term microwave exposure on the serum levels of S100β, TTR and β-trace protein with a follow up limited to two hours.

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ABSTRACT

For some time it has been investigated whether low-intensity non-thermal microwave radiation from mobile phones adversely affects the mammalian blood–brain barrier (BBB). All such studies except one have been either in vitro or experimental animal studies. The one carried out on humans showed a statistically significant increase in serum transthyretin (TTR) 60 min after finishing of a 30-min microwave exposure session. The aim of the present study was to follow up on the finding of the previous one using a better study design. Using biomarkers analyzed in blood serum before and after the exposure this single blinded randomized counterbalanced study, including 24 healthy subjects aged 18–30 years that all underwent three exposure conditions (SAR_{10G} = 2 W/kg, SAR_{10G} = 0.2 W/kg, sham), tested whether microwaves from an 890-MHz phone-like signal give acute effects on the integrity of brain-shielding barriers. Over time, statistically significant variations were found for two of the three biomarkers (TTR; β-trace protein); however, no such difference was found between the different exposure conditions nor was there any interaction between exposure condition and time of blood sampling. In conclusion this study failed to show any acute clinically or statistically significant effect of short term microwave exposure on the serum levels of S100β, TTR and β-trace protein with a follow up limited to two hours. The study was hampered by the fact that all study persons were regular wireless phone users and thus not *naïve* as to microwave exposure.

1. Introduction

There is concern about adverse health effects of radiofrequency electromagnetic fields (RF EMF) emanating from wireless phones, including mobile as well as cordless desktop phones. A main topic of such adverse effects is increased permeability of the blood–brain barrier (BBB). This hydrophobic barrier formed by the vascular endothelial cells with adjoining pericytes and extracellular matrix is a dynamic and reactive structure responding to physiological and environmental influences. Acting as both a transport and metabolic interface between the blood and the brain the BBB serves to maintain a highly stable extracellular environment for optimal central nervous system (CNS) function and to protect the brain from potentially toxic macromolecules circulating in the peripheral blood stream. Alterations of the BBB that exceed normal physiological variation are therefore considered potentially harmful as they may disrupt homeostasis and lay open the CNS to assault from extraneous substances (Marchi et al., 2004). In contrast to that, research is on-going to open up BBB to allow drugs to penetrate into the brain for treatment of e.g., brain

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tumors (Alyautdin et al., 2014; Li et al., 2013). During use of a mobile or cordless desktop phone the brain is exposed to relatively high levels of RF EMF due mainly to the close proximity of the handset on the head. Therefore the question of whether wireless phone radiation adversely affects BBB integrity is important not least since in most countries a large part of the population is frequently exposed.

1.1. In vitro and experimental animal studies

There are numerous in vitro and experimental animal studies published on the effects of RF EMF on BBB integrity (Nittby et al., 2008; Orendacova et al., 2007; Stam, 2010). These have shown that exposure intensities that significantly increase brain temperature can reversibly increase BBB permeability, however, the balance of evidence does not support an effect of non-thermal exposure at mobile phone frequencies. Studies on effects solely of exposure that does not cause heating are scarce and, therefore, do not yet permit valid conclusions. The fact that results have been contradictory is not unexpected since there were large methodological differences between the studies. This being so, some studies have shown increased permeability (Eberhardt et al., 2008; Fritze et al., 1997; Neubauer et al., 1990; Nittby et al., 2009; Salford et al., 1994, 2003; Schirmacher et al., 2000; Tang et al., 2015; Töre et al., 2001) whereas others reported no effect (de Gannes et al., 2009; Finnie et al., 2009, 2002, 2001; Franke et al., 2005a,b; Grafstrom et al., 2008; Kuribayashi et al., 2005; Masuda et al., 2007a,b; Tsurita et al., 2000). The matter is complicated further by the lack of a general mechanism by which mobile phone radiation at non-thermal exposure levels might adversely affect BBB function, though alterations of physicochemical characteristics of membranes for example have been suggested (Pall, 2013; Shivers et al., 1987). Salford et al. (2012) reported that exposure with whole-body average power densities below 10 mW/kg gives rise to a more pronounced albumin leakage than higher power densities, all at non-thermal levels. On page 45 of the BioInitiative Report, Salford et al. write that " . . . SAR levels below 1 or 0.1 mW/kg in the human brain were reported to cause a pathological leakage of the BBB and to neuronal damage." (Salford et al., 2012).

1.2. Human studies

There is to date an almost complete lack of human studies. Our group has published three on the matter (Soderqvist et al., 2009b,a, c) but because of methodological limitations neither is conclusive as to risk assessment. Additionally in this context it should be mentioned that the BBB is not the only barrier that serves to protect the brain and maintain an optimal CNS function. Albeit, a higher permeability than the BBB the blood–cerebrospinal fluid barrier (BCSFB) has an equally important role in precluding the circulation between the blood and the cerebrospinal fluid (CSF). Very little research is available on possible EMF effects specifically related to this barrier. Made up of epithelial tight junctions of the choroid plexus (CP) the BCSFB is located throughout the ventricle system of the brain and the arachnoid epithelium forming the middle layer of the meninges. Alterations in permeability (i.e., deviations from normal physiological variations) of either one of these brain-shielding barriers would be potentially harmful.

In our previous studies we analyzed blood serum content of brain permeability specific biomarkers both in association with self-reported mobile phones use (Soderqvist et al., 2009a,c) and controlled mobile phone exposure, the latter in experimental settings using an indoor base station antenna (Soderqvist et al., 2009b). In the cross-sectional study ($n = 314$) linear and logistic regression analyses of associations between serum biomarker content and self-reported long- and short-term wireless phone use adjusted for known covariates were performed. In the experimental study with human volunteers ($n = 41$) repeated blood sampling with pair wise comparisons of analyzed biomarker content before and after 30 min of controlled exposure to an 890-MHz GSM signal were performed. In neither of the two studies did we find support of an association or an effect of the exposure on the human BBB using the calcium-binding protein S100β as marker. However, using serum transthyretin (TTR), a less brain-specific marker although produced and secreted by the BCSFB at a relatively high rate, we found in the observational study that the more years of reported wireless phone use the higher the serum concentrations of TTR (Soderqvist et al., 2009c). Also in the experimental study we found small, but statistically significant elevated TTR levels 60 minutes after the end of a 30-min GSM exposure session (Soderqvist et al., 2009b).

The primary aim of this study is to follow up on the previous experimental finding using a better study design.

1.3. Biomarkers

The present study used three biomarkers to test an effect of the microwave exposure from a GSM-phone. Two markers were the same as in our previous publication, S100β and TTR. Briefly the calcium-binding protein S100β is synthesized by the astrocytic end-feet where along with other members of the S100 family it regulates cytoskeleton and cell proliferation. Normally peripheral concentrations of S100β are very low, but quickly released from the brain into the blood in response to insult leading to functional and/or morphological disruption of the BBB (Buccoliero et al., 2002; Dyck et al., 1993; Kapural et al., 2002; Marchi et al., 2003). At least 80–90% of the total S100β content is pooled within the brain (Sen and Belli, 2007). Transthyretin (prealbumin), synthesized by the liver, CP and retinal pigment epithelium, is compared to S100β a much less brain-specific marker. However, alterations in TTR content originating from the BCSFB should be detectable in serum because the CP is highly vascularized, the CSF turns over four times per 24 h (Begley and Brightman, 2003) and TTR is synthesized 13 times more rapidly in CP than in the liver (Schreiber et al., 1990).

β-Trace protein (prostaglandin D synthase) is one of the most abundant proteins in CSF (Watanabe et al., 1994). This marker is secreted from the choroid plexus, leptomeninges (pia-arachnoid membrane) and oligodendrocytes of the CNS. β-Trace protein is produced also mainly in the event of neuron and glial cell damage, which is rapidly manifested by increased concentrations also in serum (Anckarsater et al., 2007). Increased levels of β-trace protein in liquor has been linked to chronic neurodegenerative diseases and CSF leakage (Larsson et al., 2003). Normally, only small quantities of β-trace are found outside the brain. In healthy persons the concentration is 32 times lower in serum than in CSF. This is due to the small and continuous release of β-trace into CSF that by bulk flow eventually enters the blood stream where its half-life has been calculated to around 4 h (Reiber, 2003).

Another relevant aspect of β-trace protein is its role in regulation of sleep (Jordan et al., 2004; Pinzar et al., 2000) since there are studies suggesting effects on sleep associated with pulse-modulated radiofrequency fields (PRF) used in today's mobile and cordless phones. There are reports of adverse effects in observational studies but these provide little or no evidence of causal relationship (Punamaki et al., 2007; Soderqvist et al., 2008; Van den Bulck, 2007). Effects seen in experimental studies seem to be more consistent (Arnetz et al., 2007; Borbely et al., 1999; Huber et al., 2000, 2002; Hung et al., 2007; Regel et al., 2007) on the other hand, yet unclear as to their importance for health, by which mechanisms they may occur and how they relate to the associations seen in observational studies.

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