EFFECT OF MOBILE TELEPHONE RADIATION ON HUMAN PERIPHERAL BLOOD LYMPHOCYTES *IN VITRO*

~ Protection by a metallo-crystalline device ~

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SUMMARY

Samples of human peripheral blood lymphocytes (0.5ml each) maintained in an appropriate nutrient culture in a sealed 2ml phial (into which was inserted a conductive gold/platinum wire) were exposed for 12 hours to the 900MHz microwave (MW) radiations from a Motorola ETACS Classic mobile telephone on standby at 2cm distance. The telephone was active but not in transmission mode. Simultaneously an 0.5ml sample from the same culture was kept in a similar 2ml sealed phial in a double skinned mu-metal container at the same room temperature (c.17°C) and for the same period. A device claimed to protect against MW radiation (***Spiral of Tranquility***) was also placed 10cm from the telephone with a sealed 2ml phial containing an 0.5ml sample from the same lymphocyte culture.

After 12 hrs overnight exposure (2200hrs to 1000hrs) all three samples were removed to the same mu-metal shielded container and left for a further 3 hours before testing for viability by trypan blue exclusion. The samples were coded so that the microscopist was unaware of which sample was being counted.

Viability was assessed by visual counting. Two tests were made for each sample, with at least 200 cells counted in each test. All three sample cultures were then kept in the same mu-metal container for a further 24 hours, re-coded and recounted. This revealed that the lymphocytes kept only in the mu-metal container during the study retained their % viability at around 50%, whereas viability of those exposed to the mobile phone radiations had fallen even further to a new low 13% viability level. The "protected" cell samples' viability lay between the two at 20.3%, and displayed significantly better viability than the fully exposed sample.

We conclude that (uncharacterised) MW radiations from a mobile phone on stand-by can significantly adversely affect the viability of lymphocytes *in vitro*, and that "protection" claimed by a commercially available shielding device (***Spiral of Tranquility***) is supported by the results presented here. Replication with a larger sample is needed to confirm this result.

1. INTRODUCTION

The issue of possible adverse health effects from the now widespread consumer use of mobile telephony handsets has been an increasing focus of scientific discussion in recent years. Epidemiological evidence is scanty, but cell and animal studies of MW exposure bioeffects have been reported for some decades (especially at 2450MHz), though not often at the frequencies associated with mobile phones (e.g. c.900 and 1800MHz).

Microwaves are generally defined as electromagnetic waves of length between 1 metre and 1mm in air (between 300MHz and 300GHz in frequency). MW effects on the haemopoitec (blood forming) system in animals were being reported as early as 1967. Baranski found induction of peripheral blood lymphocytes as a result of long term low-level radiation together with structural changes in their nuclei. Czerski, in 1975, exposed rabbits to 2950MHz MW at 3mW/cm^2 for 2 hrs daily and found significant decrease in red cell productions when the radiation was pulsed at 1200z for 74 hrs, but only after 158 hrs with continuous wave (CW) exposure. Exposure of mice at 2950MHz and 0.5mW/cm^2 for up to 12 weeks revealed depression of antibody-producing cells but a recovery with time. Non-thermal adverse effects of MW radiation (e.g. on CTLL-1 cytotoxic T-lymphocytes) were later reported by Lyle, Schecter et al. (1983) with a significant inhibition of cytotoxicity after 4 hrs exposure at 45OMHz. Radiations from most mobile phones fall between the frequencies applied in these studies.

Research in China during the 1980s began reporting that MW radiation at levels as low as 0.1-1mW/cm^2 increased the numbers of acid phosphates positive rat lymphocytes (Wang & Low, 1984) and had deleterious effects on sperm and testicular tissue chromosomes (Chiang & Low, 1983). In 1988, Szmigielski, Bielec, et al., reviewed the growing literature and reported their own work on rabbit-granulocytes, where an increased number of dead cells was observed (by the increase in nigrosine staining) following CW exposure at 3000MHz at 105mW/cm^2, and an enhanced liberalisation of lysosomal hydrolases (a symptom of sublethal cell damage). They also reported the first large scale study of MW neoplastic abnormalities in the Polish army, showing significantly increased odds with increasing low level chronic exposure to MW radiation. Other studies since then have reported a wide variety of adverse effects, including single and double strand DNA breaks 2450MHz pulsed at 500/s *in vivo* in rat brain cells (Lai & Singh, 1995, 1996).

In 1996, the literature reporting studies of health effects of radio frequency and microwave (RF/MW) radiation from wireless communication technology were reviewed by Lin, who concluded that despite controversy, the results of studies at low SAR level (0.016-2.6 W/kg) *"suggest that repeated and/or high power pulsed microwave radiation may be capable of causing damage that is not related to average temperature elevation"*.

The mechanism of interaction is however not well understood, with a number of competing hypotheses. In an effort towards this goal we investigated effects on lymphocyte viability as measured by trypan blue exclusion at two periods after a single 12 hr exposure to a constant envelope analogue ETACS frequency mobile phone (c.890MWz). Exposure of the cells at c. 2 cm distance is compared with a sample of the same culture protected within a mu-metal enclosure, and also with a similar sample exposed in the presence of a "protective" device (***Spiral of Tranquility***).

2. METHOD AND MATERIALS

From (25ml) whole human peripheral blood freshly drawn via *vena cubitale* and differentially centrifuged at 800g for 20 minutes lymphocytes were extracted from the collected white layer, adding red cell lysing buffer (Sigma Chemicals) and BSS. This solution was centrifuged at 100g for 7 minutes and the lymphocytes collected as a pellet and re-suspended in RPMI-1640, antibiotics, antimycotics and heat-inactivated serum from the original donor, to make about 3ml of final culture. Homogeneity was ascertained by light microscopy.

This culture was divided into three 2ml cylindrical containers sterilised by autoclaving at 121°C for 20 minutes, whose sealing caps were drilled to accept gold wire inserts and sealed with gel (Dow Corning). There was approximately 0.5ml solution in each container. These containers were exposed at room temperature (c.17°C) for 12 hrs:

a) 2 cm from an ETACS mobile phone antenna on stand-by but not receiving or transmitting speech;
b) on top of a ***Spiral of Tranquility*** at 10 cm distance;
c) within a double skinned mu-metal enclosure (control).

The ***Spiral of Tranquility*** is a clear acrylic block of square base of side 45mm and five other square sides of equal dimensions, being effectively a cube of side 60mm with its corners cut off. In the acrylic are vertically set two copper split rings of diameter 35mm between which are embedded eight different crystalline stones of approximate width 4mm in a pattern of one at each corner of a square, and the remaining four set midway between them. Below one of the copper split rings is tri-spiral printed motif. The scientific significance of these components is not explained by the inventor (**Michael Poynder** Ph.D.).

After exposure all three containers were rested for 3 hrs in the same mu-metal container, then trypan blue 0.4% solution (one to one, v/v) added and the samples then coded. Two samples from each were placed under coverslips and about 250 cells counted in each sample by light microscopy (Olympus BX50). Cells dyed blue were regarded as non viable. Colourless cells were regarded as viable and video micrographs taken during the counting period to establish that they were still alive. After counting the code was broken and viable/non viable numbers compared.

3. RESULTS

The results are shown numerically below.

	3 hrs post exposure			**27 hrs post exposure**		
	viable	non viable	**% viable**	viable	non viable	**% viable**
Enclosed						
count a	159	122	**56.58**	99	107	**45.83**
count b	123	100	**55.16**	120	98	**55.05**
Total	*282*	*222*	***55.95***	*219*	*205*	***51.65***
"Protected"						
count a	109	137	**44.31**	47	200	**19.03**
count b	80	134	**37.38**	56	204	**20.32**
Total	*189*	*271*	***41.09***	*103*	*404*	***20.32***
Exposed						
count a	74	135	**35.41**	42	228	**15.55**
count b	81	127	**38.94**	33	263	**11.15**
Total	*155*	*262*	***37.17***	*75*	*491*	***13.25***

TABLE 1
Cell counts of peripheral blood lymphocytes

It is safe to assume normal distribution since:

$$p = 178/1073 = 0.\ 1685 \text{ and } q = 1-p = 0.8341.$$

Since $n_T = 507$ and $n_c = 566$ it may be seen that each of $n_T p$, $n_T q$, $n_c p$ and $n_c q$ are certainly well above 5. Since the survival rate of the "protected" group is greater than that of the exposed group, the critical value with the continuity ratio becomes:

therefore is 3.026 which indicates that the difference is statistically significant ($p < 0.001$).

GRAPHS 1 & 2

Viability of lymphocytes after 12 hr exposure to ETACS mobile phone on stand-by

To see if the effect might have been due to a thermal effect, in a separate test two probes were used, one attached to the antenna and the other immersed in an equivalent solution. Readings were taken for a 12 hr period at 30 second intervals and the data captured to a datalogger (Delta T-Devices, Burwell, Cambs). These data are shown in **Graph 3**, from which it can be seen that the temperature did not vary between the probes by more than 0.1°C.

GRAPH 3
Temperature changes of mobile phone antenna and egg-white in a cuvette

4. DISCUSSION

In this study no attempt was made to measure the level of radiation emitted by the telephone nor the frequency of its pulses, though it was assumed that even at 2cm distance this level was within the guidelines presently suggested by NRPB. The likely field strength of a 0.6 Watt output mobile phone at 1 metre is 5.3V/m, but at 2cm it will be nearer to 190V/m, and in the near field conditions differ sharply from far field exposures: there is a consensus that maximum field levels occur in skin, subcutaneous tissues, and immediately subjacent brain tissue.

In any case in the near field it is the SAR (specific absorption rate) which is used to define dose, as the incremental electromagnetic power absorbed in an incremental mass contained in a volume element of given density. In most body postures the derived limits substantially overestimate the actual absorption. This is especially true when the body or parts of the body are in the close vicinity of low powered transmitters, although actual induced absorption may be only a fraction of the SAR limits. In other words the exposure limits are of no use for the safety assessment of mobile phone handsets. For a more extensive treatment see Kuster, Balzani et al., 1997.

In this study we are only concerned with the nul hypothesis that the phone has or does not have a biological effect on the cells of interest. Furthermore, no thermal effect was detected above 0.1°C eliminating the possibility that heat absorption may be the reason for the protective effect.

Having identified the protective effect, there remains the problem of explaining it in terms of accepted science, and also explaining the underlying biological mechanisms. The device as constructed contains features pointing to the possibility of signal acquisition, transduction and re-radiation via the copper coils, which are capable of resonance and self-induction, and the crystalline structures which are capable of frequency modulation, but this avenue lies outside the scope of the present study, whose contribution is confined to a demonstration that radiations at the weakest handset transmission delivery conditions still markedly affect vital organic cells.

5. CONCLUSIONS

The radiation from some TACS mobile phones even on stand-by is capable of adversely affecting human peripheral blood lymphocytes *in vitro*. This adverse effect is mitigated to a significant extent by a device called ***The Spiral of Tranquility***.

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