Use of C. Elegans as a model organism for sensing the effects of ELF-EMFs

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Abstract. For the past two decades, there have been concerns and controversy about the effects on human health of the increased exposure to extremely-low-frequency (ELF) electromagnetic fields (EMFs) resulting from electrification, in both residential and industrial settings. Several epidemiological studies have implicated ELF-EMFs averaging 0.4 µTesla (T) or more in increased risk of cancer, especially childhood leukaemia [1,2]; there have also been many reports demonstrating effects of power-frequency EMFs on cells [outlined in 1,3]. Unfortunately, however, the precise mechanisms by which ELF-EMFs exert biological effects have proven difficult to define and results of various studies have often been hard to reproduce [1]. We believe that C. elegans offers an exciting opportunity to elucidate the effects of power-frequency EMFs on cell signalling pathways within the whole organism and are therefore investigating the effects of ELF-EMF exposure on MAPK signalling in intact worms and fertilized embryos. Through taking a targeted approach to studying the effects of ELF-EMF’s on MAPK signalling in C. elegans we aim to gather data that is physiologically relevant. Presently, this research is at a preliminary stage of preparation, and more detailed results on the exposure of Caenorhabditis elegans to ELF-EMF radiation will be presented at the conference itself.

1. Background and Discussion

For the past two decades, there have been concerns and controversy about the effects on human health of the increased exposure to extremely-low-frequency (ELF) electromagnetic fields (EMFs) resulting from electrification, in both residential and industrial settings. Several epidemiological studies have implicated ELF-EMFs averaging 0.4 µTesla (T) or more in increased risk of cancer, especially childhood leukaemia [1,2]; there have also been many reports demonstrating effects of power-frequency EMFs on cells [outlined in 1,3]. Unfortunately, however, the precise mechanisms by which ELF-EMFs exert biological effects have proven difficult to define and results of various studies have often been hard to reproduce [1].

It is generally accepted that 50/60 Hz EMFs do not transfer sufficient energy to cells to directly damage DNA leading to genotoxic effects; thus much work has focused on the effects of such fields on cellular processes including signal transduction and the production of free radicals [3,4]. In the context of signal transduction, results suggest that ELF-EMFs can affect cellular activity particularly via actions exerted on membrane proteins such as ion channels and receptors, and intracellular proteins that include signalling enzymes [1,5]. Any modulation of signalling components, through the physical interaction of ELF-EMFs with cells, will likely result in propagation and amplification of
intracellular effects ultimately leading to modification of cell behaviour. Alterations in the cytosolic concentration of second messengers such as Ca$^{2+}$ and inositol 1,4,5-triphosphate (IP3), which can influence the activation of signalling enzymes such as protein kinase C (PKC), have also been shown to result from exposure to ELF-EMFs [6,7].

Amongst the many signalling enzymes present within cells, the family of Mitogen-Activated Protein Kinases (MAPKs) have been the focus of extensive research. MAPKs, which are activated by various mitogens and biotic and physical stresses, play a central role in diverse biological processes including growth and proliferation, development, stress responses, and immunity [8]. Three evolutionarily conserved MAPK signalling modules exist, the Extracellular signal-Regulated Kinase (ERK), Stress-Activated Protein Kinase/c-Jun N-terminal Kinase (SAPK/JNK) and the p38 MAPK pathway. Each pathway is activated through a series of phosphorylation events mediated by upstream protein kinases. Recently, a few reports have demonstrated that MAPK pathways are activated in cultured mammalian cells that have been subjected to power-frequency EMFs. Exposure of lung cells to 400 and 800 µT 50 Hz EMF caused duration and intensity-dependent activation of SAPK/JNK with maximal activation occurring after 15 min [9]. In addition, 60 Hz 100 µT EMF resulted in a significant ERK activation, possibly via a PKC-mediated mechanism, in human leukaemia and breast cancer cells, and rat fibroblasts, with maximal activation occurring after 10-20 min exposure [10]. Moreover, some of these cells have been found to possess increased ERK activity following ELF-EMF exposure [11]. Interestingly, MAPKs have also been shown to be activated in invertebrate cells (immunocytes) when exposed to ELF-EMFs [12]. In this study 50 Hz 400 and 600 µT magnetic fields activated p38 MAPK leading to increased expression of heat shock proteins (HSP) 70 and 90, whereas 50 Hz 300 µT EMF was without effect [12]. These isolated reports suggest that MAPK pathways might play an important role in transducing the effects of EMFs on cells; however, this phenomenon is yet to be tested robustly, particularly in the context of cells in intact organisms.

The small free-living, transparent, nematode worm Caenorhabditis elegans (Fig. 1) is a tractable multi-cellular experimental model organism that offers excellent opportunities for studying cellular processes within the intact animal. Unlike studies with cell cultures, communication between cells is maintained using this model. Caenorhabditis elegans has a short life cycle (3.5 days at 20°C), adult hermaphrodite worms are approximately 1 mm long and contain only 959 somatic cells. Importantly, many cell types in C. elegans are shared with humans; these include neuronal, gut, excretory and muscle cells. Crucially, humans also have in common with this worm many genes and cellular mechanisms. Consequently work with C. elegans has had a major impact on understanding fundamental biological processes important to humans such as development, apoptosis, ageing and immunity [see for examples 14-16]. Although C. elegans is a versatile model organism, it has only been applied to a few studies on the cellular effects of EMFs, and largely this work has concerned the...
effect of radio-frequency electromagnetic radiation on the HSP response in transgenic worms [17,18]. Thus, engineered *C. elegans* containing reporter genes under the control of HSP promoters displayed increased gene expression after prolonged exposure to microwave radiation (750 MHz and 0.5 W for 16 h) [18]. A further report focusing on the effects of power-frequency EMF exposure (with or without mild heat-shock) showed that 50 Hz EMF also induced reporter gene expression [19]. These important studies highlight the value of *C. elegans* to EMF research. It is now important to understand the effects of ELF-EMFs on whole, wild-type (rather than transgenic) worms and, crucially, to extend work beyond the HSP response.

Commensurate with its status as a model organism, *C. elegans* possesses a number of cell signalling modules that are homologous to those present in humans, including the highly evolutionarily conserved MAPK pathways. In common with humans, MAPKs have been shown to be activated in *C. elegans* in response to biotic and physical stress [see for examples 20-22]. We believe that *C. elegans* offers an exciting opportunity to elucidate the effects of power-frequency EMFs on cell signalling pathways within the whole organism and are therefore investigating the effects of ELF-EMF exposure on MAPK signalling in intact worms and fertilized embryos.

Through taking a targeted approach to studying the effects of ELF-EMF’s on MAPK signalling in *C. elegans* we aim to gather data that is physiologically relevant. Presently, this research is at a preliminary stage of preparation, and more detailed results on the exposure of *Caenorhabditis elegans* to ELF-EMF radiation will be presented at the conference itself.

**References**

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