COST EMF - MED (Action BM1309):
European network for innovative uses of EMFs in biomedical applications

STSM Report:

Novel approach to bio-electromagnetic effects measurements and quantification of metabolic activity

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Abstract:

This STSM report details evaluation of ultra-weak photon emission (UPE) from aqueous solutions of amino-acids and wild-type Saccharomyces cerevisiae strands. The presented STSM experimental results are focused on the measurement of intensity of ultra-weak photon emission from amino-acid tyrosine and from yeast cells, both exposed and unexposed to exogenous electromagnetic fields. The results of UPE from tyrosine confirm the assumption that the highest intensity is detected from the solution, wherein the highly reactive hydroxyl radical is formed, which causes oxidation of tyrosine, formation of high-energy intermediates and electron excited species. The results of UPE from yeast cells indicate a difference in the time dependence of UPE intensity between the cells exposed to exogenous EMF and unexposed cells, which could be caused by magnetic field influence on oxidative reactions during cell metabolism. However further experiments are necessary to confirm our preliminary findings.

A. Purpose of the STSM

The project objectives included the construction of experimental apparatus exploiting the mutual know-how and available technologies from both participating institutions. Upon completion of the experimental apparatus, definition of exposure standards and protocols was elaborated, along with the selection of candidate microorganisms. The main objective of the project included experiments during which ultra-weak photon emission was recorded and evaluated from various amino-acid solutions and Saccharomyces Cerevisiae strands. Once sufficient control experimental data was obtained, exogenous LF EMF was applied to exposed samples and ultra-weak photon emission was be re-evaluated and preliminary conclusions were drawn as to the influence of EMF on reactive oxygen species (ROS). Other than the experimental results, the STSM has also helped to foster mutual cooperation and knowledge transfer between the Institute of Photonics and Electronics and the University of Zilina. Future activities foresee further experiments with exogenous electromagnetic fields and yeast cells in view of UPE since the allotted time for the STSM could not completely account for the complexity of the subject at hand.

B. Work Description

Ultra-weak photon emission (although also termed autoluminescence or biophoton emission, amongst others), is present in virtually all metabolically active living systems – from simple bacteria, through plants, tissue cultures and also in humans. The mechanism of ultra-weak photon generation is sought to originate from cellular oxidative metabolism, wherein electronically excited species are formed, even in the absence of external stimuli such as light or enzymes. Pronounced mechanisms of electromagnetic field action on biological systems is through the coupling of magnetic field to unpaired spins of radical biochemical species to affect yield of biochemical reaction [1998 Timmel, 2008 Okano, 2012 Maeda]. Such endogenous oxygen radical species lead to chemiluminescent reactions which generate weak but measurable fluxes of photon emission from biological systems [Cifra 2014].

Two types of ultra-weak photon emission (UPE) are currently known – spontaneous and induced. Our area of interest within this proposal is focused on the latter, wherein we plan to measure UPE as a novel approach to biological effects monitoring of electromagnetic fields (EMFs), and to verify non-chemical means of metabolic stimulation based on application of low frequency EMFs. Other examples of documented metabolic stimulators include various biotic (viral, bacterial, fungal etc.), abiotic (temperature, mechanical damage, light stress, ionizing radiation etc.) and oxidative stresses. Furthermore, scientific studies evaluate the effects of applied exogenous EMFs directly based on colony-forming unit (CFU) counts or the growth area, or indirectly by means of evaluating the optical density. To the best of our knowledge, only few published studies have explored this possible ultra-weak photon emission – low frequency EMF link [Cheun 2007, Dotta 2011].

In view of COST action BM1309 objectives, ultra-weak photon emission has been the subject of various studies including dermatology, neuroscience and especially oncology. Cell metabolism, accompanied by ROS production, is often correlated with cellular growth rate. Therefore, it is reasonable to explore UPE measurement in conjunction with LF EMF exposure in samples where growth is extensively high, such as in yeast and cancerous tissues.

C. Results

The STSM experimental research can be divided into three periods. In the first period the ultra-weak photon emission from aqueous solutions of amino-acids was analyzed. It was shown, that the highest intensity of UPE is detected after oxidation of amino-acids by hydroxyl radical. Three aqueous solutions of amino-acids – tyrosine, phenylalanine and histidine were used several experiments. The average results of UPE from solution of tyrosine and solution of tyrosine and hydrogen peroxide can be seen in the Fig. 1. Purified water is also depicted as control sample.

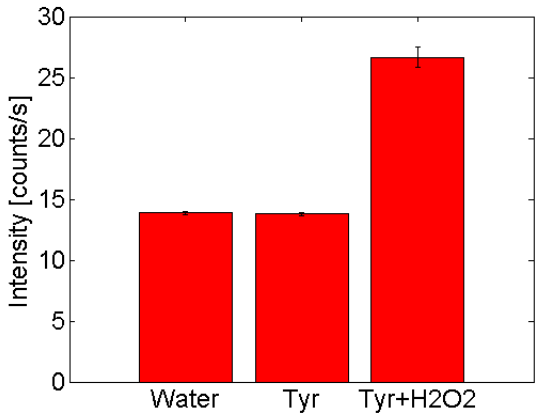

Fig.1 Ultra-weak photon emission from different aqueous solutions

The presented results show that the intensity of UPE from water and solution of tyrosine is low and not distinguishable from dark count (noise) of photomultiplier (15 counts/s). Although hydroxyl radical in the aqueous solution of tyrosine and hydrogen peroxide is not formed, molecules of tyrosine are probably oxidized by molecular oxygen or by products of auto-oxidation of hydrogen peroxide. This could lead to the formation of unstable intermediates such as dioxetane and tetroxide and follow origins of electronically excited species, whose decomposition is accompanied by emission of photons.

A significant increase of UPE is observed from the solution of tyrosine, hydrogen peroxide and ferrous sulphate heptahydrate (Fig. 2).

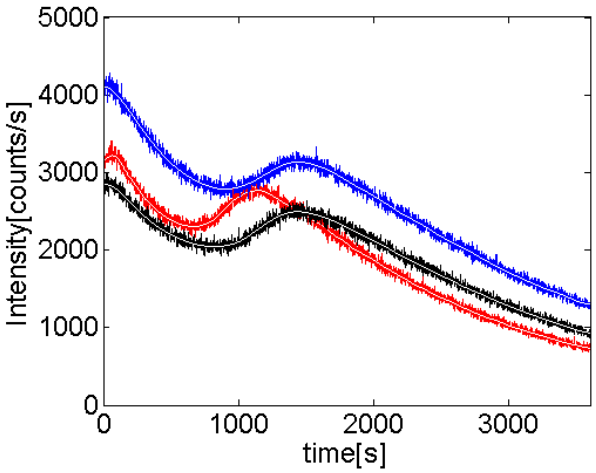

Fig.2 Three repetitions of measurement of ultra-weak photon emission from solution of tyrosine, hydrogen peroxide and ferrous sulphate dehydrate

The reaction of hydrogen peroxide with iron ions, called Fenton reaction, creates a highly reactive hydroxyl radical. The said radical probably initiates oxidation of tyrosine, leading to the formation of dioxetane and tetroxide and followed origin of electronically excited species and emission of photons.

The second period of experimental research was focused on measurements of UPE from yeast cells. Multiple experiments were conducted to evaluate the growth curve and also the concentration of carbon dioxide, that is produced during the metabolism of cells.

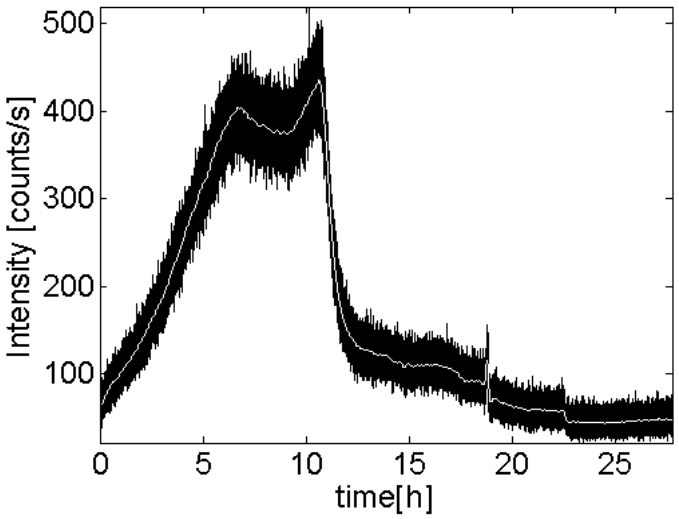

Fig.3 Ultra-weak photon emission from yeast Saccharomyces cerevisiae (wild-type)

The third period of experimental work was concentrated on evaluation of non-thermal electromagnetic effects on metabolic activity of yeast cells using UPE detection. It is proposed that magnetic field can affect the radical reactions during cell metabolism and therefore also photon emission. Preliminary results can be seen in Fig. 4.

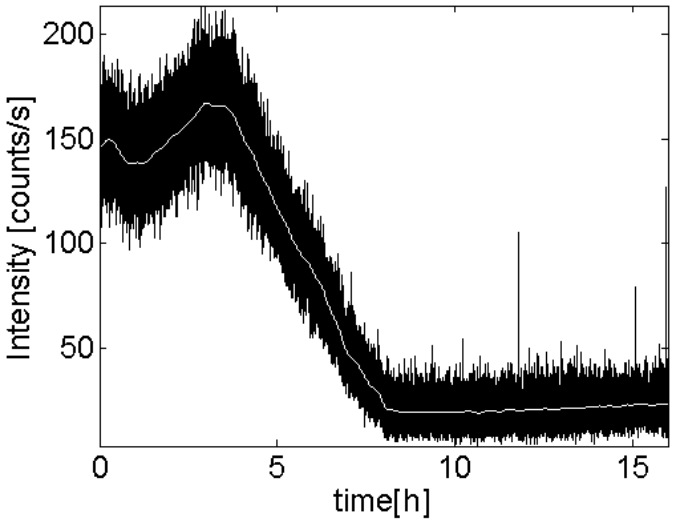

Fig.4 Ultra-weak photon emission from Saccharomyces cerevisiae (wild-type) exposed to exogenous electromagnetic field

The results of UPE from cells Saccharomyces cerevisiae exposed to exogenous magnetic field (magnetic flux density 0.35 mT, frequency 1 kHz) seem to indicate differences compared to UPE from unexposed cells. The UPE from unexposed cells exhibits different time curve when compared to experiments without exogenous EMF. Specifically, the fast decrease of intensity after reaching its maximum (Fig. 3), whereas the decrease of UPE from cells exposed to magnetic field is significantly slower (Fig. 4). This difference could be likely caused by alterations in oxidative reactions during cell metabolism and perhaps we could speculate the degree of influence of the exogenous electromagnetic field on the lifetime and concentration of reactive oxygen species in solution. Further experiments would be beneficial to observe the growth curve of cells and correlate the said growth curve with the electromagnetic field effects on UPE. The results represent our first attempt to

evaluate the magnetic field influence on UPE from living cells, however it is necessary to carry out further experiments to state any conclusion(s).

D. Future collaboration with host institution

Further collaboration is prospectively planned between both institutions within the Socrates/Erasmus programme and by mutual staff and student exchange.

E. Expected Publications

Transcom 2015 - 11th European conference of young researchers and scientists

Communications – Scientific letters of the University of Zilina

Confirmation by the host institution of the successful execution of the STSM:

We confirm that Dr. Ján Barabáš has performed the research work as described above.

Contact Person of Host Institution	Name of researcher
Dr. Michal Cifra	**Dr. Ján Barabáš**
Signature 	Signature