THE USE OF SR-FTIR MICROSPECTROSCOPY FOR THE PRELIMINARY BIOCHEMICAL STUDY OF THE RAT HIPPOCAMPAL FORMATION TISSUE IN CASE OF PILOCARPINE INDUCED EPILEPSY AND NEUROPROTECTION WITH FK-506

J. Dudała¹, K. Janeczko², Z. Setkowicz², D. Eichert³, J. Chwiej¹

dudala@novell.ftj.agh.edu.pl

¹AGH-University of Science and Technology, Faculty of Physics and Applied Computer Science, Krakow, Poland, ²Jagiellonian University, Institute of Zoology, Department of Neuroanatomy, Krakow, Poland, ³ELETTRA, Trieste, Italy

The main aim of the work was the biochemical analysis of the hippocampal formation tissue in case of epileptic rats treated with neuroprotective agent FK-506. Three groups of animals were compared: rats with pilocarpine induced seizures treated and non-treated with tacrolimus as well as naive controls.

Synchrotron radiation Fourier transform infrared (SR-FTIR) microspectroscopy was used for the analysis of the distribution of proteins, lipids as well as changes in protein secondary structure and saturation level of lipids. The measurements were carried out at SISSI beamline of ELETTRA. A Bruker IFS 66v/S interferometer coupled to Bruker Hyperion 2000 microscope was used. The tissue samples were analyzed in transmission mode with a beam defined by small aperture and spatial resolution steps of $10~\mu m$ which allowed us to probe the selected cross-line of the sample at cellular resolution. The obtained results enabled to compare the distributions of proteins and lipids in the three hippocampal cellular layers, i.e. in molecular, multiform and granular layers.

For epileptic animals both treated and non-treated with FK-506 the tendency for increase of amide II/amide ratio was observed however only for multiform layer these changes were statistically significant. Similar relation was noticed in case of the ratio of the absorbance at around 1631 and 1658 cm⁻¹. The mentioned results may suggest conformational changes of proteins in the direction of β -sheet secondary structure.

Additionally, statistically significant increase in the lipid massive and decrease of the ratio of absorbance at around 2921 and 2958 cm⁻¹ were observed for epileptic animals treated with tacrolimus comparing to the control group.