

5th INTERNATIONAL CHOLNOKY SYMPOSIUM

ABSTRACT BOOK



25TH APRIL 2019
DEAN'S CONFERENCE ROOM,
UNIVERSITY OF PÉCS MEDICAL SCHOOL,
12 SZIGETI STREET, PÉCS



Examination of biologically relevant electron transfer in photolyases with ultrafast laser spectroscopy

Jonatan Pasitka, Dr. Andras Lukacs, Dr. Grama Laszlo, Fekete Zsuzsanna, Stephen R. Meech

University of Pécs

Introduction: In the last few decades an enormous technological expansion could have been seen in the field of lasers. This resulted in the application of basic biophysical methods in the every day medicine, methods like super-resolution microscopy, CARS microscopy, Raman-spectroscopy and even ultrafast laserspectroscopy.

Materials and Methods: With the help of ultrafast laser spectroscopy the recording of intramolecular procedures became possible. Several, biologically relevant processes are known that take place within the femtosecond scale. Such an example is photoisomerisation. We have been focusing on the ultrafast laser spectroscopic features of these latter two proteins. They are phylogenetically connected, however photolyase is a DNA repair enzyme while cryptochrome is the member of the group of proteins composing the circadian clock. The explanation for such different functions so far from each other lays in the understanding of the procedures happening intramolecularly on the scale of picoseconds. We made two photolyase mutants, in which we altered the N378 AA, of which supposed that it was to change the redox potential of FAD in the enzyme, thus changing its functionality to resemble to cryptochromes. First, we had to construct the mutant proteins, for which we used a pPROEX plasmid, overlap extension mutagenesis PCR technique, TOP10 and BL21 E.coli strains.

Results and Discussion: With our measurements we could prove our expectations being correct, which means that the redox state of the FAD altered due to the given mutations to a state that is characteristic to cryptochromes.