2P247 In-situ 光照射固体 NMR によるバクテリオロドプシン Y185F
変異体に捕捉された O-中間体の評価
Characterization of O-like intermediate trapped in Y185F mutant in Bacteriorhodopsin by in-situ photo-irradiation solid-state NMR

Kyosuke Oshima,1 Arius Shigeta,1 Yoshiteru Makino,1 Izuru Kawamura,1 Takashi Okitsu2, Akimori Wada2, Satoru Tuzi1, Akira Naito1 (Grad. Sch. Eng., Yokohama Natl. Univ.,2Kobe Pharm. Univ.,1Univ. Hyogo)

Bacteriorhodopsin (bR) in a purple membrane of H. salinarum shows a light-driven proton pump activity under photo-irradiation. In the photocycle, it is difficult to trap O intermediate and hence its detailed structure has not revealed yet. Here, we demonstrate that the O-like intermediate in Y185F mutant can be stationary trapped by in-situ photo-irradiation solid-state NMR. In the dark adapted state, 13C NMR spectrum of [15,20-13C]Ret-[1-13C]Tyr-Y185F-bR indicates that the ratio of 13-cis and All-trans retinals is about 4:1. Under photo-irradiation with green light, both N and O intermediates were distinctly observed in the 13C NMR spectrum. Subsequently, the N-intermediate transformed to the O-intermediate under the dark condition.

2P248 光依存転写因子オーレオクロム 1 の反応ダイナミクス
Reaction Dynamics of Light Dependent Transcription Factor Aurochrome-I

Yuki Akiyama1, Yusuke Nakasone1, Osamu Hisatomi2, Yoichi Nakatani2, Masahide Terazima1 (Graduate School of Science, Kyoto University,1Graduate School of Science, Osaka University)

Aurochrome-I is a blue light dependent transcriptional factor which has two domains; photoreceptive LOV domain and DNA binding bZIP domain. Aurochrome-I exists as dimer in vitro by forming disulfide-linkages and binds to DNA both in the light and dark states. In vivo, however, it has recently been supposed that the Aurochrome-I can exist as monomer and light dependent dimerization should be relevant for the DNA binding. In order to clarify the reaction of monomeric Aurochrome-I, we constructed C162S/C182S mutant and studied its reaction dynamics by the transient grating method. Upon photoexcitation, diffusion coefficient (D) decreased significantly by the dimerization at the rate constant k= 2.5 s⁻¹. In the presence of DNA, light dependent DNA binding was observed as a further decrease of D.

2P249 (6-4)光回復酵素による 2 光子 DNA 修復の分子メカニズム
Molecular mechanism of the two photon DNA repair by the (6-4) photolyase


UV in sunlight causes formation of crosslinks between two adjacent pyrimidine bases in DNA, namely cyclobutane pyrimidine dimers (CPDs) and pyrimidin(6-4)pyrimidine photoproducts (6-4)PPs. Since these lesions lead to mutagenesis and cell death, organisms have developed UV-protecting systems that can remove the UV lesions and restore intact nucleobases. The (6-4) photolyase is a unique flavoenzyme that can repair the (6-4)PP by utilizing blue light. We have recently reported that the repair of the (6-4)PP by the (6-4) photolyase requires two photons. In this study, we investigated the role of the amino acid residues located proximal to the lesion and performed biochemical and spectroscopic studies. Their molecular role in the two-photon DNA repair will be discussed.

2P250 Theoretical study of the electron transfer reaction by DNA photolyase


The DNA photolyase repairs thymine dimers found in UV-induced DNA lesions by electron transfer reaction. In this study, the molecular mechanism involved in the efficient electron transfer of DNA photolyase was investigated by the analysis of electron tunneling pathways from FADH- to CPD using ab initio and fragment molecular orbital calculations. In particular, we focused on the roles of amino acid residues and crystallographic water molecules in the active site.

According to previous studies, the calculation of electron transfer pathways, considering excited state of FADH- and three amino acids (Gln283, Asn249 and Met353), have not been performed. We computed the electron transfer pathways from FADH- to CPD using ab initio and fragment molecular orbital method.

2P251 光化学系 II 倍合体と層状水酸化物からなるバイオ–無機ハ
イブリッド電極
Bio-inorganic hybrid water oxidation electrodes of Photosystem II and layered double hydroxides


We report bio-inorganic hybrid electrodes consisting of Photosystem II (PSII) and layered double hydroxides (LDHs) for visible light-driven water oxidation. PSII is a photosynthetic protein, which catalyzes the oxidation of water to molecular oxygen under visible light irradiation. LDHs are composed of cationic double hydroxide nanosheets and anions placed in the interlayers of the cationic nanosheets. We synthesized PSII-LDH electrodes and recorded their photocurrent in a buffered aqueous solution at pH 6.5 under visible light irradiation. In our system efficient interfacial electron transfer occurred from PSII to LDH, allowing us to experimentally determine turnover numbers of PSII in vitro for the first time.

2P252 光化学系 II における TyrZ - D1/His190 の距離と PCET の
関係
Proton-coupled electron transfer and hydrogen-bond distance of TyrZ - D1/His190 in Photosystem II

Miwa Sugiu1, Shogo Ogami2, Fabrice Rappaport3, Alain Boussac1 (1PROS, Ehime Univ./JST-PRESTO,2Dep. Chem., Ehime Univ.,3IBPC)

Here we show that the geometry of the TyrZ phenol and its environment, likely the Tyr-O---H---Nε-His bonding, are modified in PsbA2-PSII when compared to PsbA(1/3)-PSII of Thermosynechococcus elongatus. These results point to the dynamics of the proton coupled electron transfer processes associated with the oxidation of TyrZ being affected. From sequence comparison we propose that the Cys144Pro and Pro173Met substitutions in PsbA2-PSII versus PsbA(1/3)-PSII, respectively located in the interlayers of the cationic nanosheets. We synthesized PSII-LDH complexes and recorded their photocurrent in a buffered aqueous solution at pH 6.5 under visible light irradiation. In our system efficient interfacial electron transfer occurred from PSII to LDH, allowing us to experimentally determine turnover numbers of PSII in vitro for the first time.