2P393**Theoretical study on color-tuning mechanism of visual pigments**○Kazuhiro Fujimoto¹, Shigehiko Hayashi², Jun-ya Hasegawa¹, Hiroshi Nakatsuji¹¹Dept. of Synthetic Chemistry and Biological Chemistry, Kyoto Univ., ²Dept. of Chemistry, Kyoto Univ.

Absorption peak position of retinal proteins significantly depends on the protein environment (opsin) around the retinal chromophore, which is called "opsin-shift". In a previous study, we have performed the SAC-CI and QM/MM calculations on several retinal chromophores in rhodopsin, bacteriorhodopsin, and sensory rhodopsin II, and succeeded in reproducing the experimental absorption energies. Protein environment causes blue-shift in the absorption spectrum. Our analysis showed that this blue-shift is due to the electrostatic (ES) potential produced mainly by the neighboring glutamate and aspartate. In this study, we extended our investigation to the human blue and compared the color-tuning mechanism with those in bovine rhodopsin. To analyze the color-tuning mechanism, the absorption energies were decomposed by three parts; distortion effect in the retinal chromophore, ES effect from the opsin environment, and hydrogen-bonding effect between the retinal chromophore and the counter-ion groups. The results show that the difference in absorption energies is mainly due to the ES effect. To further understand the mechanism in detail, we studied the ES energy contributions from each amino acid separately. It showed that the polar residues near the chromophore play vital roles in absorption energies difference.

2P395**The important role of AMP-Mg in promoting the decomposition of dioxetanone interacted with some amino acid residues in firefly luciferase**○Hironori Sakai¹, Naohisa Wada¹, Masami Kusunoki²¹Dept. of Life Sciences, Toyo Univ., ²Dept. of Physics, Meiji Univ.

In firefly bioluminescence, dioxetanone (DOX) is formed when substrate luciferin is oxygenated together with ATP and Mg²⁺ in the active site of enzyme luciferase (Lase). On the hydroxyl group of DOX releasing its H⁺, DOX decomposes efficiently into CO₂ and electronically excited oxyluciferin (Oxylⁿ). Since deprotonation of DOX is regarded as a key step of firefly bioluminescence, we focused which amino acid residue can induce this reaction in the active site of Lase. Recently, the crystal structure of the Japanese firefly Lase combined with AMP and Oxylⁿ was determined, where anionic Glu313 coexists with cationic Arg220 in the proximity of the hydroxyl group of Oxylⁿ. Furthermore, AMP-Mg-His around the carbonyl group of Oxylⁿ might be positively charged through hydrogen bond between AMP and His247. Therefore, our model of calculations is proposed as follows: in the decomposition of DOX, its polarization by cationic AMP-Mg-His triggers H⁺ transfer from DOX to Glu313, thus the excitation step of the reaction can easily follow the deprotonation of DOX. We examined this model theoretically by performing geometry optimization and calculating free energy of the system. As the result, the possibility of hydroxyl H⁺ of DOX transferring to Glu313 estimated by comparing free energy of the neutral DOX with anionic Glu313 to that of the anionic DOX with neutral Glu313 leads to the interesting conclusion that the proton transfer from DOX to Glu313 can occur in spite of the low pI value of glutamic acid (c.a. 3.2) in the aqueous solutions.

2P394**On the Yellow-Green Bioluminescence in North American Firefly : SAC-CI Study**

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Color-tuning mechanism in firefly bioluminescence is actively discussed for many years, because of its efficiency for DNA reporter assays. The yellow-green bioluminescence of North American firefly had been believed as fluorescence from enol-form of firefly oxyluciferin by analogy with chemiluminescence. However, recent experimental study proposed that the yellow-green light is obtained from analogue of oxyluciferin restricted to keto-form in bioluminescence. We investigated fluorescence energies both chemi- and bioluminescence by SAC-CI ab initio theoretical calculations. Our results give consistent explanations for both chemi- and bioluminescence, and show that the keto-form of oxyluciferin can emit the yellow-green light in bioluminescence. From detailed analysis, this can be achieved by the electrostatic interactions with the surrounding residues of luciferase activesite.

2P396**Quantum mechanical study of the proton transfer via a peptide bond in the novel pathway of proton translocation of cytochrome c oxidase**

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We have validated the mechanism of the proton transfer via the peptide bond in the H pathway of cytochrome c oxidase using the B3LYP/6-31++G(d,p)//HF/3-21G calculations with the polarizable continuum model. The proton transfer via the peptide bond was classified into two reactions: the proton transfer forming an enol peptide bond and the keto-enol tautomerism of the peptide bond. The proton transfer forming an enol peptide bond could require the protonation of the propionate group of heme *a* to decrease pK_a of the propionate group and weaken the electrostatic interaction between proton and the propionate group. In the reaction, a proton is added to the carbonyl oxygen of the peptide bond, forming imidic acid [-C(OH)=N⁺H-] followed by an enol form of the peptide [-C(OH)=N-]. The enol peptide bond could be converted to the keto one by a proton wiring mechanism using the putative water molecules and the amide group of the neighboring peptide bond (Ser441-Asp442) with the activation free energy of 21.8 kcal/mol. Superposition of the computed transition structure on the X-ray crystallographic structure indicates that a water molecule, a neighboring amide group (Ser441-Asp442), Ser205, and protonated Asp51 could be proton mediators for the proton wiring keto-enol tautomerism.