THE ROLE OF INTER-HELICAL INTERACTIONS IN ELECTRON-TRANSFER GATING IN PHOTOSYSTEM II

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Non-covalent interactions between transmembrane (TM) helices of membrane-protein complexes may affect subunit association and domain flexibility. Conformational changes of flexible protein domains have been suggested to be involved in electron-transfer gating in photosynthetic reaction centers (RC). We set out to study the role of non-covalent inter-helical interactions between two subunits of photosystem II (PSII) in the gating mechanism of electron-transfer between the quinones QA and QB.

The core of PSII RC is made of two protein subunits, D1 and D2. Two TM helices of these subunits are associated in their membranal region through a single hydrogen bond (H-bond) embedded within a common helix packing motif (GxxxSxxxG). A putative H-donor in this contact point, D1-Ser212, was mutated to all other amino acids in the cyanobacterium Synechocystis sp. PCC 6803.

Thirteen mutations were found to support photoautotrophic growth excluding bulky residues that are positively charged (Arg, Lys), or aromatic (Phe, Trp, Tyr and His). In the photoautotrophic mutants, the effect of the mutations on forward electron transfer rates and charge recombination was evaluated following fluorescence decay and thermoluminescence as a function of temperature. The results showed that weakly polar residues such as Ser (wild type), Thr, Ala and Cys had similar rate constants (kAB) of electron transfer over a range of temperatures (10 – 40 °C). On the other hand, strong polar and/or bulkier residues, such as Gln, Asn, Glu and Asp, had lower kAB, which increased in a temperature-dependent manner. The temperature effect on kAB varied among the mutants suggesting that protein conformations influence electron transfer rates. Moreover, good correlations were found between the activation enthalpies of forward electron transfer dH‡ and the activation energy of charge recombination (EA), and between dH‡ and mean packing values, especially when comparing residues that can form H-bonds. These findings suggest that weak hydrogen bonding and polar interactions at the TM helical interface between the D1 and D2 subunits affect local protein conformations involved in the gating of electron-transfer from QA to QB in PSII.

NATURE OF BINDING OF BOMBYKOL IN PHEROMONE BINDING PROTEIN. AN AB INITIO STUDY

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Interactions of insects with their surroundings are mostly based on chemical signals. One of the most remarkable communication systems known mediates sexual behavior of moths. Mature females ready to have offspring emit a sexual pheromone from their abdomen to attract conspecific males for mating. The 'single-pheromone molecule' tuned detection system of males is located in branches of male's antennae. On these antennae olfactory hairs, *sensila trichodea*, are located. They are filled with sensilar liquor and house specialized dendritic cells, innervated to insect brain globular structures. Here, the signal received from the cell is proceeded and further recognized as a call for copulation.

The sensilar liquor contains a high concentration (10 mM) of water-soluble pheromone-binding-protein (PBP).

An analysis of the crystal structure of Bombyx mori PBP···bombykol (pheromone) complex [1] identified nine amino acid residues involved in intermolecular hydrogen bonds, ··· interactions, C-H·· hydrogen bonds and weak interactions of purely van der Waals character. Using the model fragments as the representatives of each residue, the interaction energies of their complexes with bombykol were computed by *ab initio* calculations. The values were compared with literature and further discussed in terms of the method and basis set dependence, and the co-operative effect (influence of the neighboring groups on the interacting pair). It enabled us to explain quantitatively the nature of the binding forces in [BmPBP···bombykol] complex in terms of contribution of the individual amino acids and individual types of interaction. It was observed that 70% of the stabilization is due to interactions other than classical hydrogen bonds.

1. B. H. Sandler, L. Nikonova, W. S. Leal, J. Clardy, *Chemistry & Biology* 7 **(2000)**, 143-151. PDB code: 1DQE.