Electrostatic field inside the photosynthetic pigment-protein complexes probed by Stark spectroscopy

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Photosynthetic pigments such as carotenoids and bacteriochlorophylls (Bchls) are under the influence of electric field generated by surrounding apo-proteins, when they are bound to pigment-protein complexes. Although the high resolution structures of the photosynthetic pigment-protein complexes have been obtained by using X-ray crystallography [1], relatively little attention has been paid to correlate the electrostatic interaction in the pigment-protein complexes and their physiological functions. Stark spectroscopy is a useful technique with which to probe the pigment-pigment and pigment-protein interactions in photosynthetic pigment-protein complexes [2]. Although the general theory behind Stark shift is relatively well developed, a better theoretical understanding of Stark spectroscopy in a biological context is required. This is needed in order to be able to correlate details of the measured spectra with a structural understanding of the pigment interactions, and how these affect the magnitude of the experimentally determined Stark shifts.

In this talk, we will present some new results of X-ray crystallography of the reaction centre (RC) from a photosynthetic bacterium *Rhodobacter* (*Rba.*) *sphaeroides* [3]. Our up-to-date high-resolution crystal structure analysis (1.95 Å resolution) visualizes the electron density of the crystalline water, detergent molecules, lipids, and some other small molecules, which are surrounding the RC. These molecules may affect the electrostatic environment of the protein itself and hence that of the pigment molecules embedded in the pigment-protein complexes.

We will also present the results of Stark spectroscopy on the pigment-protein complexes from *Rba. sphaeroides* [4,5]. Stark spectroscopy using dual-phase lock-in technique was applied to the B850 absorption band of the peripheral antenna (LH2) complex from *Rba. sphaeroides* strain G1C [4]. When a pigment is subjected to an electric field and shows a Stark shift, the quadrature-phase (out-of-phase) signal depends on the pigment's interaction with the surrounding environment. By studying the frequency and temperature dependence of the quadrature-phase signals the nature of this interaction can be investigated. Similar technique was applied to the special-pair Bchls absorption band of the RC from *Rb. sphaeroides* strain R26.1 with or without an analogue carotenoid had been reconstituted [5]. The magnitude of electric field around the special-pair Bchls induced by the presence or absence of carotenoid was quantitatively determined.

Finally, some new results of Stark spectroscopy on the purified complete and reconstituted core antenna (LH1) complexes from a purple photosynthetic bacterium *Rhosospirillum rubrum* will be presented. The effect of the detergent molecules surrounding the LH1 complex is discussed.

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