Self-assembled Monolayer of Light-harvesting 1 and Reaction Center (LH1-RC) Complexes Isolated from *Rhodospirillum rubrum* on an Amino-Terminated ITO Electrode

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LH1-RC complexes isolated from *Rhodospirillum rubrum* were successfully assembled onto an ITO electrode modified with aminopropylsilane. Efficient energy transfer and photocurrent responses of the complexes were observed upon illumination at 880 nm.

Highly efficient conversion from solar light to chemical energy occurs in the intracytoplasmic membranes of photosynthetic bacteria. The primary events of the light energy conversion involve the absorption of photons by light-harvesting (LH) complexes and subsequent energy transfer to the reaction center (RC) where transmembrane vectorial electron transfer takes place.[1]

Our understanding of charge separation and energy transfer in these complexes has enabled the first steps to be taken towards generating artificial systems that convert light energy into usable electrical current. Previous attempts to produce an artificial, energy-converting electrode system immobilized either the LH1 complexes[2] or RC[3] on the electrodes. Until now, there has been no attempt to immobilize the intact 'core' complex, consisting of both the LH1 and the RC components together, onto an electrode. Recently, a structure of the LH1-RC 'core' complex has been obtained by X-ray crystallography to 4.8 Å and reveals that the LH1 complex surrounds the contours of the RC to confer on the 'core' complex an overall oval rather than circular shape.[4]

We have recently developed a procedure to create a self-assembled monolayer (SAM) of reconstituted LH1 complex on a transparent indium tin oxide (ITO) electrode modified with aminopropylsilane (APS) by using electrostatic interactions or hydrogen bonding between the electrode surface and the anionic LH1 polypeptides at pH 8.0.[2] The near infra-red (NIR) absorption spectrum showed that the LH1 complex is stable when immobilized onto the electrode. Our current work extends this approach to the native LH1-RC complexes.

LH1-RC 'core' complexes isolated from *Rhodospirillum rubrum* (*R. rubrum*) were successfully assembled on an ITO electrode modified with APS (APS-ITO). Efficient energy transfer and photocurrent responses could be observed upon illumination at 880 nm.

The core complexes were isolated from *R. rubrum* and purified as described previously.[5] The OD_{880} of the isolated core complex was adjusted to 0.3 cm^{-1}. APS-ITO or bare ITO was immersed in the core complex solution, Tris-HCl, pH 8.0 for 6 h at 4 °C and then rinsed with 20 mM Tris-HCl (pH 8.0) buffer. Photocurrents were measured at −0.2 V (vs Ag/AgCl) in a homemade cell that contained three electrodes; (1) a core-modified electrode as the working electrode, (2) an Ag/AgCl (saturated KCl) as the reference electrode, and (3) a platinum flake as the counter electrode. The working electrode was illuminated with a halogen lamp unit, AT-100HG, through a monochromator, SPG-120S (Shimadzu). The solution consisted of 0.1 M phosphate buffer (pH 7.0), containing 0.1 M $NaClO_4$ and 5 mM methyl viologen.

Figure 1 shows the NIR absorption spectrum of the purified *R. rubrum* core complex in 20 mM Tris-HCl buffer (pH 8.0) (dotted line). The NIR spectrum of the core complex showed the absorption maximum at 880 nm with two smaller peaks at 800 and 760 nm. The former is attributable to the mixture of bacteriochlorophyll *a* (BChl*a*) in the LH1 complex (880 nm) and BChl*a* 'special pair' dimer (870 nm) Qy transition and the latter two peaks to the BChl*a* called "accessory" (800 nm) and bacteriopheophytin (760 nm) in the RC, respectively.[1] The NIR absorption spectrum of the core complex on the electrode (solid line) indicates that the complex on the electrode was almost native. The peak at 780 nm on the electrode was more pronounced than that in buffer, obscuring the peak at 800 nm, and can be attributed to some monomeric BChl*a* released from the LH1-RC complex during incorporation onto the electrode surface.

In the previous study it was apparent that the assembled RC by itself on the electrode was relatively labile,[3] whereas in this study the complete assembled core complex proved to be quite stable. The stability of the RC together with the LH1 complex

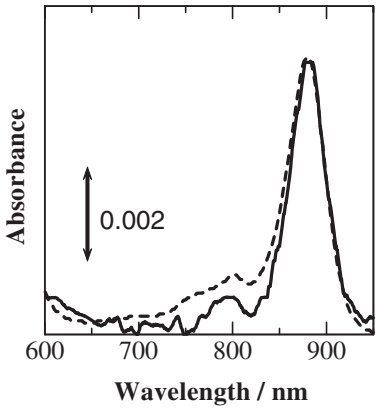

Figure 1. NIR absorption spectra of the LH1-RC core complexes in 20 mM Tris-HCl buffer (pH 8.0) (dotted line) and on the APS-ITO electrode (solid line).

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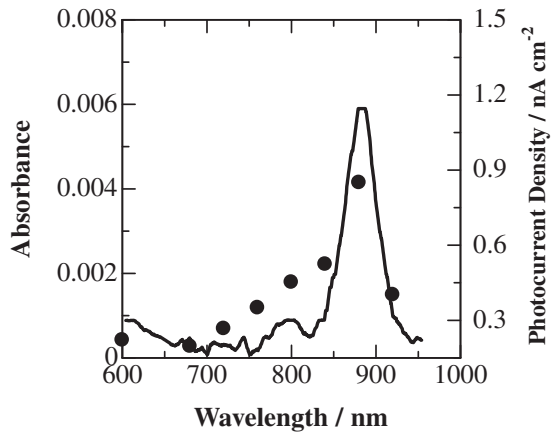

Figure 2. Photocurrent density (dots) and NIR absorption spectrum (solid line) of the LH1-RC core complexes assembled onto an APS-ITO electrode.

likely results from mutually supportive interactions between the two complexes to provide a combined rigidity that is in excess of either of the individual complexes. Interestingly, the fluorescence of BChl*a* molecules in the LH complex on the APS-ITO was strongly quenched because of the presence of RC when illuminated at 880 nm, implying that an efficient energy transfer from BChl*a* in the LH1 complex to RC in the core complex occurred on the electrode (data not shown).

Figure 2 shows the photocurrent generated from the core complex assembled onto an APS-ITO (dots). Under the experimental conditions a cathodic photocurrent was observed, implying that the electron was transferred from pigments in the core complex to methyl viologen.[6] Interestingly, an enhanced photocurrent was observed especially upon illumination at 880 nm. When the LH1 complex alone was immobilized on the electrode, the observed photocurrent was mainly generated by light absorbing at 770 nm from monomeric BChl*a*.[6] Furthermore, when the RC complex only was immobilized on the electrode, an efficient photocurrent was not observed upon illumination at 880 nm. Thus, the enhanced photocurrent observed at 880 nm in the assembled core complex can be ascribed to energy transfer from LH1 to the RC and then electron transfer from the RC to the electrode. The enhanced photocurrent results from either electron transfer from the special pair or the accessory BChl*a* in RC,[7] although more detailed experiments will be needed to elucidate the exact mechanism of this reaction. This data indicates that the core complex was well organized on the ITO and the photocurrents were driven by light that was initially absorbed by the LH components.

In conclusion, the SAM method is clearly successful in allowing assembly of functional core complexes on the electrode. This has been confirmed by NIR absorption spectroscopy, demonstrating that the photocurrent response is derived from electron transfer between the RC and the electrode and is enhanced by illumination at 880 nm.

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References

1 R. E. Blankenship, M. T. Madigan, and C. E. Bauer, in "Anoxygenic Photosynthetic Bacteria," Kluwer Academic Pubrishers, Dorderecht (1995).
2 M. Ogawa, R. Kanda, T. Dewa, K. Iida, and M. Nango, *Chem. Lett.*, **2002**, 464.
3 a) K. Matsumoto, K. Nomura, Y. Tohnai, S. Fujioka, M. Wada, and T. Erabi, *Bull. Chem. Soc. Jpn.*, **72**, 2169 (1999). b) J. Kong, Z. Lu, Y. M. Lvov, R. Z. B. Desamero, H. A. Frank, and J. F. Rusling, *J. Am. Chem. Soc.*, **120**, 7371 (1998).
4 A. W. Roszak, T. D. Howard, J. Southall, A. T. Gardiner, C. J. Law, N. Isaacs, and R. J. Cogdell, *Science*, **302**, 1969 (2003).
5 D. Fotiadis, P. Qian, A. Philippsen, P. A. Bullough, A. Engel, and C. N. Hunter, *J. Biol. Chem.*, **279**, 2063 (2004).
6 M. Nagata, Y. Yoshimura, J. Inagaki, Y. Suemori, K. Iida, T. Ohtsuka, and M. Nango, *Chem. Lett.*, **32**, 852 (2003).
7 M. E. van Brederode and R. van Grondelle, *FEBS Lett.*, **455**, 1 (1999).

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