SYNTHESIS OF A COVALENTLY LINKED BACTERIOPYROPHEOPHORBIDE—PYROPHEOPHORBIDE HYBRID DIMER[1]

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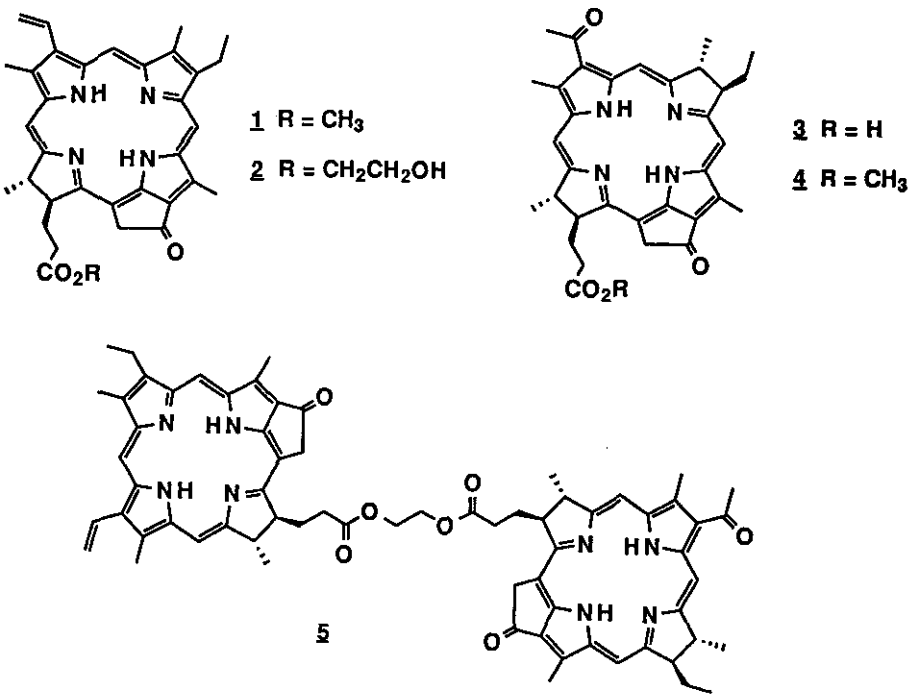
Abstract — A covalently-linked bacteriopyropheophorbide *a*—pyropheophorbide *a* hybrid dimer was prepared. In this molecule, singlet energy transfer takes place from the pyropheophorbide to the bacteriopyropheophorbide subunit.

The synthesis of covalently linked chlorophyll dimers has attracted considerable attention in relation to the duplication of optical and redox properties similar to those of the special pair in the photosynthetic reaction center.[2,3] To date, however, synthesized models have been rather limited to homodimers of pyropheophorbides and bacteriopyropheophorbides in which possible intramolecular singlet energy transfer is usually difficult to detect since it regenerates the same excited state. Here, we report the synthesis of bacteriopyropheophorbide *a*—pyropheophorbide *a* (BP—P) heterodimer (5) as the first example of a bacteriochlorin—chlorin hybrid dimer. In this molecule, the BP and the P subunits have different excited energies, 1.65 and 1.86 eV,[4] respectively, and thus one can expect one-directional intramolecular energy transfer from the latter to the former.

Methyl pyropheophorbide *a* (1) was conveniently transformed by the acid-catalyzed ester exchange reaction into 2-hydroxyethyl pyropheophorbide *a* (2) which was then coupled with bacteriopyropheophorbide (3) to give the dimer (5). We found the combination of 2-chloro-1-methylpyridinium iodide and 4-dimethylaminopyridine as condensation catalysts gave 5 in 75%. The spectroscopic data obtained for 5 are fully consistent with the assigned structure; FAB mass showed a peak at m/z 1112 ($M^+ + 2$), and 400 MHz ^{1}H nmr spectrum gives a set of the signals of the BP and B subunits in addition to a singlet peak appearing at 4.12 ppm due to the protons of the ethylenedioxy bridge. No appreciable upfield shifts were detected, suggesting a rather extended conformation in $CDCl_3$.



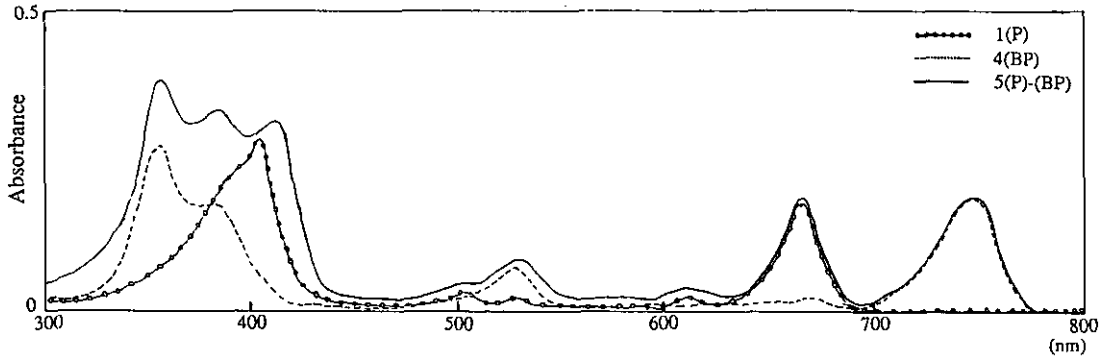
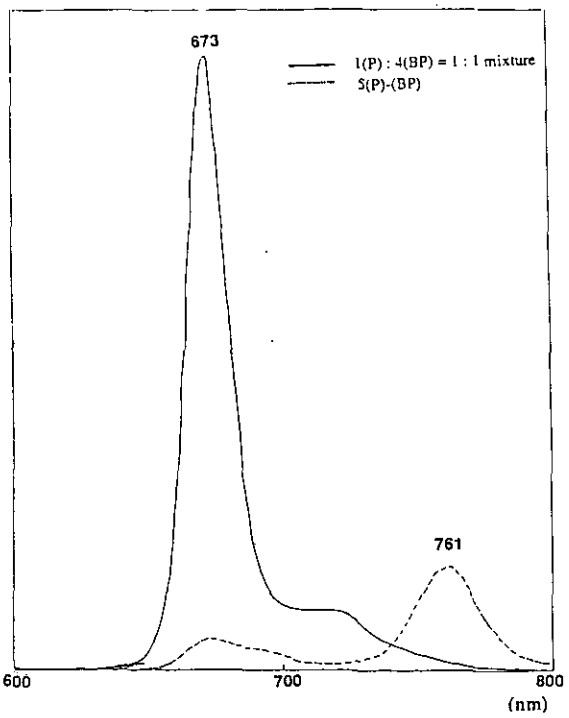


Figure 1 Absorption spectra of 1, 4, and 5 in THF.

The absorption spectrum of 5 is a simple sum of those of the respective subunits (Figure 1). In the fluorescence spectrum of 5, the fluorescence from P is strongly quenched and that from BP is enhanced (Figure 2), indicating the occurrence of intramolecular singlet energy transfer. The energy transfer has been confirmed by measuring the fluorescence lifetimes by the single photon counting technique.[5] The reference molecules (1) and (4) exhibit the fluorescence decays which can be both analyzed in terms of single exponential functions with time constants of 6.7 and 2.8 ns, respectively. The fluorescence of 5 at 680 nm where the main emitting species is P exhibits a rapid decay with a time constant of 22 ps. On the

basis of these data, the energy transfer rate (k_{EN}) has been determined to be 4.5 x 10^{10} s^{-1} by the following equation, $k_{EN} = 1/\tau - 1/\tau_0$, where τ and τ_0 are the fluorescence lifetime of 5 and 1, respectively. This large rate can be accounted for in terms of the Förster mechanism since the fluorescence spectrum of P is well overlapped with the absorption of BP.[6] The dimer (5) has a potential use as a detection probe for the energy migration from large aggregates of natural and modified bacteriochlorophyll *c*.[7]



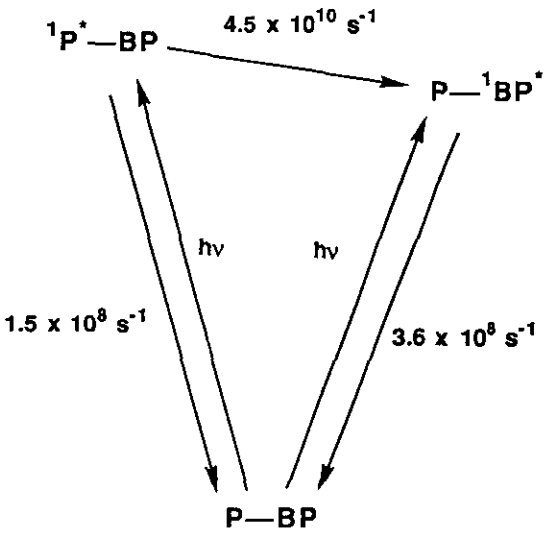


Figure 2 Fluorescence spectra of (5) and a 1:1 mixture of (1) and (4) in THF for excitation at 412 nm.

Experimental Section

General methods. The instruments and procedures were as previously reported.[8] Methyl pyropheophorbide *a* (1) obtained in large scale from Spirulina Maxima (Nippon INK) was transformed into 2-hydroxyethylpyropheophrobide *a* by the ester exchange reaction in 75% yield. Bacteriochlorophyll *a* obtained from large-scale (25 l) culture of *Rhodobactor sphaeroides* was transformed into bacteriopyropheophorbide essentially the same procedure as those for pyropheophorbide.[3]

Bacteriopyropheophorbide *a*—pyropheophorbide *a* hybrid dimer (5). A mixture of 2 (35 mg, 0.063 mmol), 3 (32 mg, 0.058 mmol), 2-chloro-1-methylpyridinium iodide (59 mg, 0.232 mmol), and 4-

dimethylaminopyridine (35 mg, 0.286 mmol) in CH_2Cl_2 (20 ml) was refluxed for 2 h under argon. The reaction mixture was poured into water and extracted with CH_2Cl_2 The extract was washed with 7% $NaHCO_3$, and dried over anhydrous Na_2SO_4. The solvent was evaporated and the residue was separated by flash silica gel column (eluting with a mixture of methanol and CH_2Cl_2 (0.2/99.8)) to give 5 (49 mg, 75%). ^{1}H-Nmr ($CDCl_3$); -1.73, 0.42 (s, 1H+1H, NH), -1.12, 0.26 (s, 1H+1H, NH^{Bp}), 1.07 (t, 3H, J = 8.0 Hz, 8^2-CH_3^{Bp}), 1.66 (t, 3H, J = 8.0 Hz, 8^2-CH_3), 1.68, 1.72 , 1.78 (each d, 3H, J = 7.0 Hz, 7-CH_3, 18-CH_3, 18-CH_3^{Bp}), 1.97-2.11, 2.18-2.34, 2.41-2.64 (m, 2H+4H+2H+2H, 8^1-CH_2^{Bp} + 17-CH_2CH_2 + 17-$CH_2CH_2^{Bp}$), 3.09 (s, 3H, 3-$CO\underline{CH_3}^{Bp}$), 3.19, 3.34, 3.37, 3.41, 3.62 (each s, 3H, 2-, 7-, 12-CH_3, 2-^{Bp}, 12-CH_3^{Bp}), 3.60 (dq, 2H, J = 6, 7.5 Hz, 8^2-CH_2), 3.94, 3.97, 4.15, 4.26 (m, 1H+1H+1H+1H, 7-, 8-, 17-, 18-H^{Bp}), 4.12 (s, 4H, $CO_2\underline{CH_2CH_2}$), 4.75, 4.96 (d, 1H+1H, J = 20.0 Hz, 13^2-H_2^{Bp}), 5.00, 5.18 (d, 1H, J = 20.0 Hz, 13^2-H_2), 6.14 (dd, 1H, J = 1.5, 11.5 Hz, 3-CH=$\underline{CH_2}$), 6.24 (dd, 1H, J = 1.5, 18.0 Hz, 3-CH=$\underline{CH_2}$), 7.91, 7.94 (dd, 1H, J = 11.0, 18.0 Hz, 3-$\underline{CH}$=CH_2), 8.30 (s, 1H, 20-H^{Bp}), 8.44 (s, 1H, 10-H^{Bp}), 8.50 (s, 1H, 20-H), 8.96 (s, 1H, 5-H^{Bp}), 9.32 (s, 1H, 5-H), 9.44 (s, 1H, 10-H). Ir (KBr) ν_{max} 1738 (CO_2CH_3), 1695 (C=O), 1687 (C=O) 1662 (C=O) cm^{-1}. Ms (FAB) Found: m/z 1112. Calcd for $C_{68}H_{72}N_8O_7$: M^++2, 1112.

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REFERENCES AND NOTES

1. This paper is dedicated to Dr. Shigeru Oae, Professor Emeritus of Tsukuba University, on the occasion of his 77th birthday.
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