Highly Efficient Energy Dissipation by a Carotenoid in Face-to-Face Porphyrin−Carotenoid Dyads

Satoshi Shinoda,*,† Hiroshi Tsukube,† Yoshinobu Nishimura,‡ Iwao Yamazaki,‡ and Atsuhiro Osuka§

Department of Chemistry, Graduate School of Science, Osaka City University, Sugimoto, Sumiyoshi-ku, Osaka 558-8585, Japan, Department of Molecular Chemistry, Graduate School of Engineering, Hokkaido University, Sapporo 060-8628, Japan, and Department of Chemistry, Graduate School of Science, Kyoto University, Kyoto 606-8502, Japan

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Introduction

Since carotenoids function as light-harvesting and photoprotective pigments during photosynthesis,[1,2] considerable efforts have focused on understanding the interaction mechanism between carotenoids and chlorophylls (or bacteriochlorophylls) in the light-harvesting complexes.[3] Recently, the dissipation of excess light energy by carotenoids during photosynthesis has been attracting attention as a photoregulation function of the carotenoid.[4−9] It is proposed that, in higher plants, the formation of zeaxanthin via "xanthophyll cycle" (biochemical interconversion among violaxanthin, antheraxanthin, and zeaxanthin by epoxidation/de-epoxidation) leads to the thermal dissipation of the excess light energy absorbed by chlorophylls.[10,11] A similar interconversion between carotenoids has also been found in many species of microalgae.[12] The role of zeaxanthin is not clear at the present time, but one plausible mechanism is the singlet-to-singlet energy transfer from the chlorophyll to the carotenoid.[13,14]

Because direct measurement of the $S_0 \leftrightarrow S_1$ transition of a carotenoid is generally difficult due to the small transition dipole moment and rapid nonradiative decay to the ground state, the detailed analysis of the energy transfer mechanisms of carotenoids has not been obtained.[15] Recently, reasonable values were obtained for the S_1 state energies of carotenoids from an extrapolation using a series of shorter polyenes and by applying the energy-gap law to various carotenoids.[13,14] Indeed, the S_1 energy of zeaxanthin, which has 11 conjugated double bonds, was estimated to be lower than that of chlorophyll-*a*, and therefore considered as an energy-accepting state.

To achieve highly efficient photoreactions concerning carotenoids, the distance between a carotenoid and chlorophyll must be short enough to allow large interactions. The structure determination of some light-harvesting complexes in higher plants and purple bacteria has revealed that the carotenoids are in a proximity with the chlorophylls to satisfy the structural demand for effective interactions.[16−18] Although artificial dyad models in which carotenoids are covalently linked with chlorophyll-related molecules have been utilized to mimic photosynthesis, the role of the linkage bonds as a mediator of through-bond electronic interaction cannot be missed, especially when the aromatic molecule was used for the linkage.[19,20] The discrimination of through-space interactions from through-bond ones should be done to develop an effective model compound.

We present here three kinds of porphyrin−carotenoid dyad molecules **3c**−**5c**, in which conformationally rigid spacer molecules such as cyclohexane, Kemp's triacid, and xanthenedicarboxylic acid were introduced to separate two chromophores by saturated bonds and minimize the through-bond interaction, while keeping them in proximity in space. The distance between two adjacent functional groups in the spacer molecules is estimated to be 2.9 Å when using Kemp's triacid and 4.5 Å for the xanthenedicarboxylic acid. Although the amide bonds provide a stiff linkage between the porphyrin and the carotenoid, the center-to-center distance between these two chromophores will be variable since the distance and orientation of the carotenoid relative to the porphyrin have a broad distribution due to free rotaion around the single bonds. On the basis of the measurements of the steady-state fluorescence and fluorescence lifetime of porphyrin in the dyad, we successfully demonstrated highly efficient fluorescence quenching behavior of the attached carotenoid. In particular, dyad **5c** offered the highest quenching efficiency among the reported dyad system. Thus, the obtained results described below, that the carotenoid close to a porphyrin primarily works as a quencher of the porphyrin excited state, give significant insights into the carotenoid's photoregulation function in vivo.

* Corresponding author. Phone: +81-6-6605-3139. Fax: +81-6-6605-2522. E-mail: shinodas@sci.osaka-cu.ac.jp.
† Osaka City University.
‡ Hokkaido University.
§ Kyoto University.

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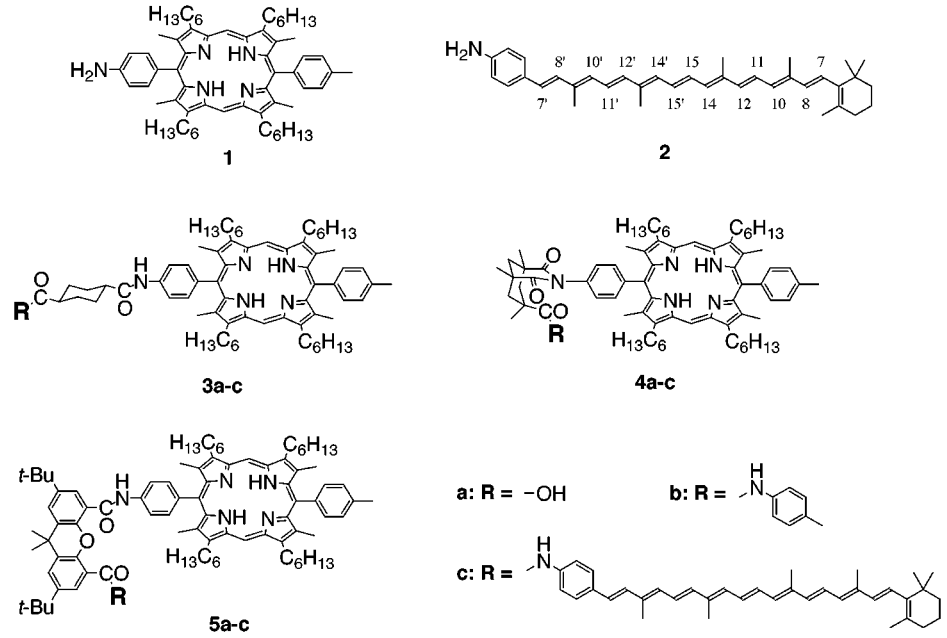
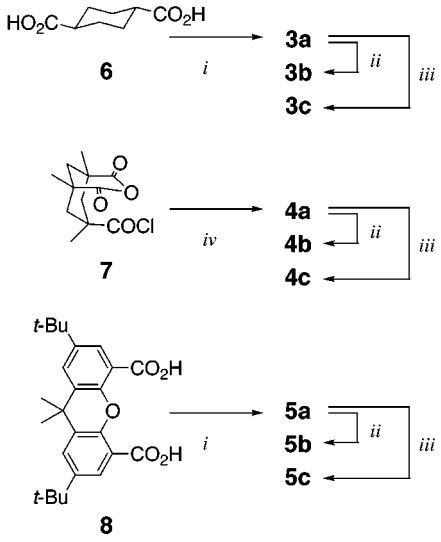


Figure 1.

Scheme 1[a]



[a] (i) CMPI, DMAP, CH_2Cl_2, reflux; (ii) *p*-toluidine, CMPI, DMAP, CH_2Cl_2, reflux; (iii) **2**, CMPI, DMAP, CH_2Cl_2, reflux; (iv) **1**, 2,6-di-*tert*-butylpyridine, toluene, reflux.

Results and Discussion

Aminoporphyrin 1[21] and aminocarotenoid **2**[19] were synthesized according to the reported methods. Three kinds of porphyrin−carotenoid dyads **3c**−**5c** and their reference compounds lacking the carotenoid moieties **3b**−**5b** were synthesized according to Scheme 1. Both chromophores were linked through three kinds of spacer molecules **6**−**8** via amide/imide bonds. Kemp's triacid anhydride−chloride **7**[22] and 2,7-bis(1,1-dimethylethyl)-9,9-dimethylxanthene-4,5-dicarboxylic acid **8**[23] were prepared, respectively, from Kemp's triacid and 9,9-dimethylxanthene as previously reported. The anhydride−chloride **7** was first linked with **1** in refluxing toluene in the presence of 2,6-di-*tert*-butylpyridine to yield the porphyrin-carboxylic acid **4a**. Porphyrincarboxylic acids **3a** and **5a** were synthesized, respectively, by esterification of **6** and **8** with 1 equiv of aminoporphyrin **1** using 2-chloro-1-methylpyridinium iodide (CMPI) and DMAP in refluxing dichloromethane. The porphyrincarboxylic acids **3a**−**5a** were then condensed with aminocarotenoid **2** under similar reaction conditions. Each dyad was separated from the reaction mixture by flash silica gel column chromatography until it showed only one spot on TLC under irradiation by UV/visible light (254/366 nm). In the case of **4c**, the product yield was low (30%) due to the formation of an acid anhydride as a byproduct. The low yield of **4c** probably comes from the steric hindrance around the carboxylic group.

The time-averaged conformation of each dyad in chloroform-*d* was predicted from its ^{1}H NMR chemical shifts. Since the motion of the pigments was faster than the NMR time scale, only one time-averaged conformation could be observed in **3c**−**5c** at room temperature. Upfield shifts due to the ring current effect of the porphyrin π-electron system were definitely observed in the signals for the vinyl protons of the carotenoid moiety in **4c** and **5c**, while only slight shifts were seen in dyad **3c**. These indicate that the carotenoids of dyads **4c** and **5c** lay well above the deshielding region of the porphyrin ring current. The largest upfield shifts (ca. 1 ppm) were observed for the proton signals of 10′-, 11′-, and 12′-H of **5c**, suggesting the nearly parallel conformation of the carotenoid relative to the porphyrin (see Figure 1 for the numbering system). Since the corresponding upfield shifts in **4c** were smaller (ca. 0.4 ppm) than those of **5c**, the center-to-center distance between the porphyrin and carotenoid is inferred to decrease in the order of **3c** > **4c** > **5c**.

Figure 2a shows the absorption spectra of **3c**−**5c** in THF. No differences in the porphyrin absorption wavelength among the reference compounds **3b**−**5b** were observed, indicating that attachment of the cyclohexane,

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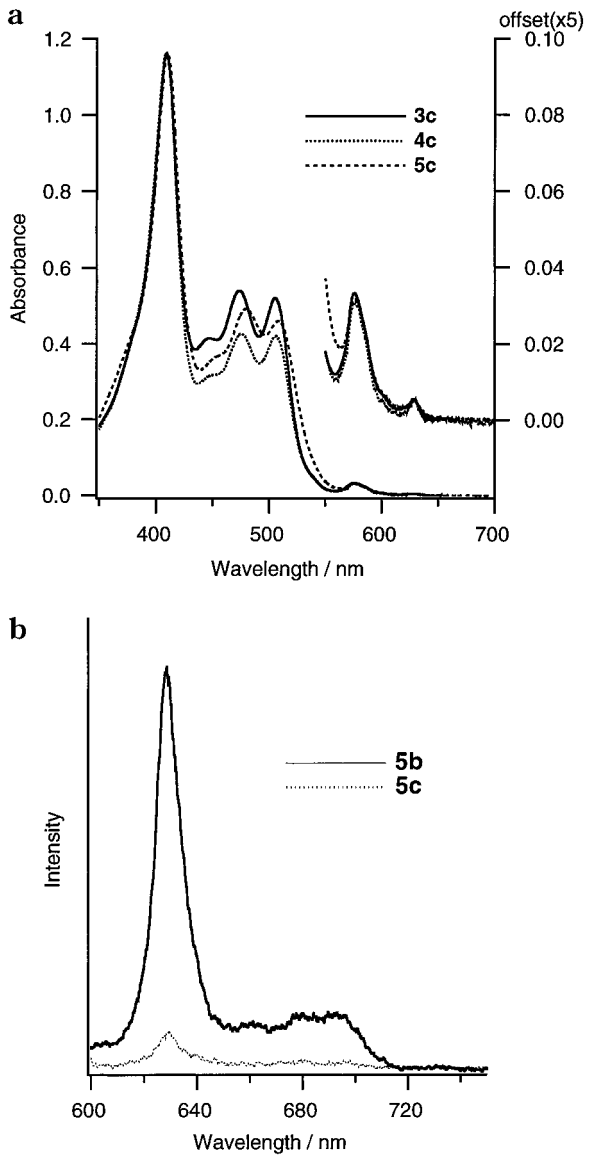

Figure 2. (a) Absorption spectra of the dyads in THF at room temperature. They are magnified in the 550−700 nm region (offset, right axis). (b) Steady-state fluorescence spectra in THF at room temperature. The blank value was subtracted from the shown spectra.

Kemp's triacid, and xanthene spacers gave practically no perturbation to the porphyrin moieties. Both the B and Q absorption bands of the porphyrin moieties of the dyads **3c**−**5c** were also unaffected by the attached carotenoid moieties compared with those of the reference porphyrin monomers **3b**−**5b**. On the other hand, the absorbance of the carotenoid moiety showed a red shift by 5 nm at the maximum absorbance in **5c** that may be due to the π-conjugation of the carotenoid with the introduced xanthene group to some extent through the amide bond. Such shifts of the carotenoid absorption bands were not obvious in **3c** and **4c**. Since the phenyl ring substituted at the meso-position of the porphyrin is perpendicular against the porphyrin plane in the present models, the spacer xanthene moiety only slightly purturbs the electronic state of the porphyrin.

When the porphyrin moieties of dyad **5c** and its reference **5b** were selectively excited at the Q_y bands (580 nm) in THF, their steady-state fluorescence spectra (Figure 2b) showed the emission bands from the porphyrin moiety which have their maxima at 628 nm. The concentration of **5b** and **5c** were adjusted to be the same by the absorbance at 628 nm, where the contribution from carotenoid is small and negligible, but **5b** exhibited a higher fluorescence intensity than **5c**. Dyads **3c** and **4c** similarly showed reduced fluorescence spectra compared with references **3b** and **4b**.

Table 1. Relative Fluorescence Quantum Yields of Dyads[a]

compd	DMF	THF	benzene
3c	0.82	0.86	0.86
4c	0.28	0.39	0.33
5c	0.076	0.097	0.11

[a] Fluorescence quantum yield of the corresponding references **3b**−**5b** in the same solvent was set to unity.

Singlet energy transfer from an excited carotenoid to porphyrin or pyropheophorbide has been observed in several "dyad" systems,[19,21,24−27] which has been studied in relation to the light-harvesting function of carotenoids. We have demonstrated with zeaxanthin−pyropheophorbide **9** that singlet energy transfer from zeaxanthin to pyropheophorbide (4−15%) could be assigned as an energy transfer from the zeaxanthin S_2 state via a dipole−dipole interaction (Förster mechanism). Recently, energy transfer from the carotenoid S_2 state on a femtosecond time scale in the light-harvesting complexes have been reported.[28−31] To estimate the possibility of energy transfer from the carotenoid S_2 excited state to the porphyrin in dyads **3c**−**5c**, their steady-state corrected fluorescence excitation spectra were measured in THF by monitoring the porphyrin fluorescence. The fluorescence excitation spectra of **3c** and **4c** were consistent with the absorption spectra of the corresponding reference porphyrins **3b** and **4b**, suggesting that energy transfer from the carotenoids to the porphyrins did not occur at all in dyads **3c** and **4c**. Since the molar extinction coefficients of the porphyrin Q-bands, which may serve as energy acceptors, are smaller than those of the pheophorbide Q_y bands, the lack of energy transfer in **3c** and **4c** can be explained by the small rate constant of energy transfer due to the poor spectral overlap between donor emission and acceptor absorption. Through-bond interaction between two chromophores is probably weak as a consequence of the larger number of saturated bonds between them. The fluorescence intensity of **5c** was too weak, thus not allowing an estimation of the energy transfer efficiency from the carotenoid to the porphyrin. Further measurements of the lifetime of the carotenoid excited state are necessary to precisely determine the rate constant of energy transfer.

Porphyrin fluorescence quenching was evident in dyads **3c**−**5c**. Table 1 summarizes the relative fluorescence

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Table 2. Fluorescence Lifetimes of Dyads and References

	τ/ns[a]		
compd	DMF	THF	benzene
3b	12	11	11
4b	13	11	11
5b	12	11	11
3c	12 (0.88)	11 (0.90)	11 (0.88)
	1.8 (0.12)	1.9 (0.10)	2.1 (0.12)
4c	10 (0.07)	8.8 (0.10)	9.0 (0.08)
	3.2 (0.70)	3.8 (0.70)	3.5 (0.69)
	0.45 (0.23)	0.77 (0.20)	0.52 (0.23)
5c	10 (0.05)	10 (0.07)	10 (0.07)
	1.6 (0.48)	1.7 (0.48)	1.6 (0.47)
	0.19 (0.47)	0.19 (0.45)	0.17 (0.47)

[a] Values in the parentheses are the relative amplitudes of each component.

quantum yields of dyads **3c**−**5c** determined from the peak heights of the fluorescence spectra in DMF, THF, and benzene. They were significantly dependent on the natures of the spacer groups, and the most significant decrease (larger than 90%) in fluorescence intensity was found for dyad **5c** (**5c** > **4c** > **3c**). Probably, the center-to-center distance between the porphyrin and carotenoid influences the degree of fluorescence quenching.

The fluorescence lifetime of a porphyrin was measured using the time-correlated photon counting technique.[32] The porphyrin moiety was selectively excited with 2 ps pulses of a dye laser at 620 nm, and the decaying of the fluorescence from the porphyrin was monitored at 690 nm. The lifetime of the carotenoid S_1 state in toluene has been reported by Gust et al. with the *N*-acetylated form of **2** to be 16.2 ps.[25] The fluorescence decay curves of the reference compounds **3b**−**5b** were well fitted with a single-exponential function to give almost the same lifetimes (t = 11−13 ns; Table 2), indicating that the quenching is merely caused by the attached carotenoids. In contrast, the fluorescence decays of dyads **3c**−**5c** were well fitted with bi- or triexponential functions (χ^2 < 1.3), represented as

$$I(t) = A_1 \exp(-t/\tau_1) + A_2 \exp(-t/\tau_2) + A_3 \exp(-t/\tau_3)$$

where t_i and A_i are the lifetime and the preexponential amplitude of the ith component (Figure 4), respectively. The lifetimes became short and the initial amplitude of the fastest decaying component increased in the order of **3c** > **4c** > **5c**.[33] The observed bi- or triphasic fluorescence decay may be derived from the distribution of lifetimes for various conformers of carotenoid relative to the porphyrin. That would be possible if the time scale of some rotational modes of a large molecule such as carotenoid were comparable to or longer than the lifetime of the S_1 state of the porphyrin.[34]

Fluorescence quenching by the linked carotenoid was also observed in other dyad model compounds.[19,21,35,36] The efficient fluorescence quenching (ca. 75%) was reported for the porphyrin−carotenoid ester **10**, in which the carotenoid was attached at the ortho-position of the meso-phenyl group and located 4−5 Å above the tetraphenylporphyrin. Photoinduced charge separation was suggested for this quenching, but no direct evidence for formation of the charged species was reported.[37] On the contrary, we have demonstrated that fluorescence quenching in the porphyrin−carotenoid dyad **11** occurred via energy transfer from the excited porphyrin to the carotenoid S_1 state followed by a rapid decay of the excited carotenoid to the ground-state based on the results of transient absorption spectroscopy.[21] In the present dyads

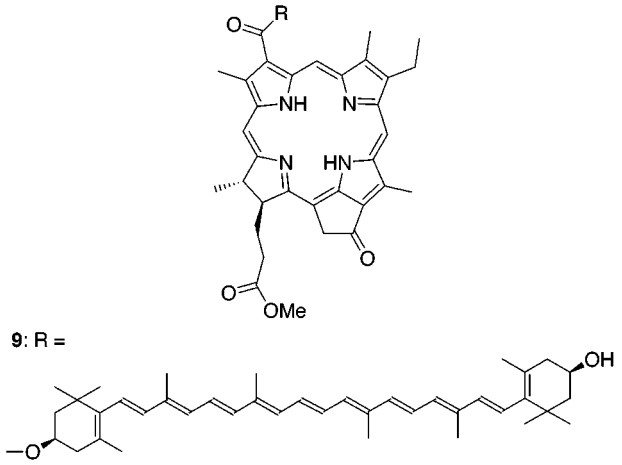

Figure 3.

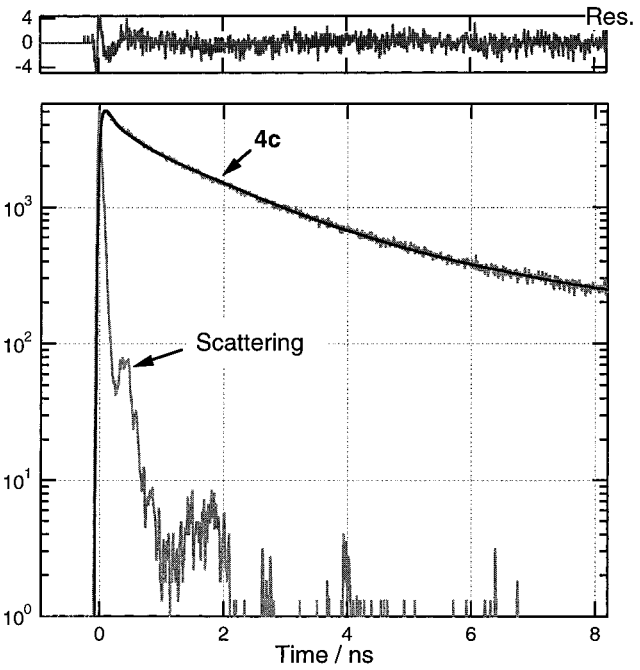

Figure 4. Fluorescence decay profile of dyad **5c** in THF. The best fit result with three components and the residuals are also shown.

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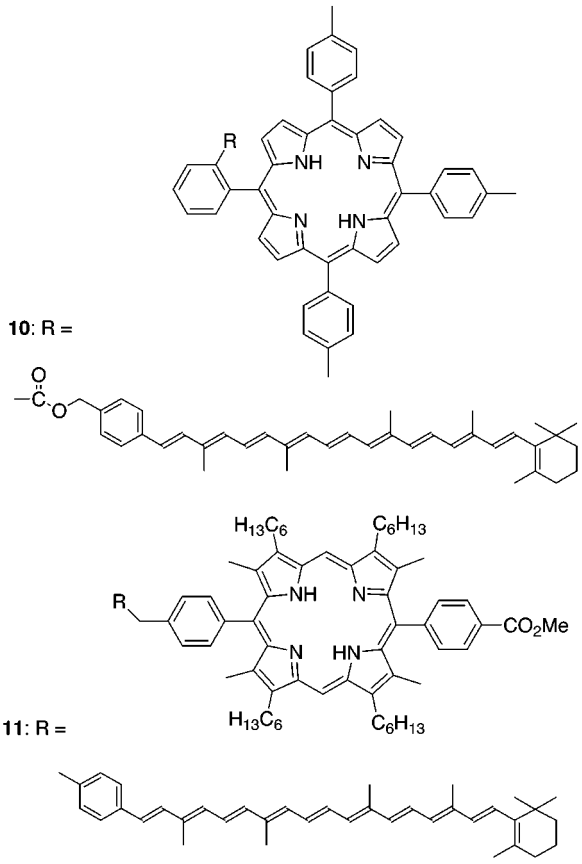

Figure 5.

3c−**5c**, only a weak dependence on solvent polarity was observed with respect to both the lifetimes and the initial amplitudes of all the components. If electron transfer is involved in this fluorescence quenching, the solvent polarity must have a large influence on the quenching rate. Thus, we consider a smaller contribution by electron transfer in this quenching process. The S_1 energy level of the carotenoid used here must be lower than that of the β-carotene, which has 11 conjugated double bonds, as the conjugation spreads over the phenyl ring on its one side, and the S_1 state of the porphyrin is energetically higher than that of chlorophyll. Therefore, the energy transfer from the porphyrin to the carotenoid should be an exothermic process.

Conclusion

Conformationally restricted porphyrin−carotenoid dyads display efficient intramolecular fluorescence quenching phenomena which resemble the energy dissipation by carotenoids in biological light-harvesting complexes. Dyad **5c**, in which the rigid xanthene-type spacer-linked carotenoid and porphyrin are in close proximity, offered more than a 90% fluorescence quenching. Since this is the highest quenching efficiency reported among the dyad systems, dyad **5c** can be regarded as the most suitable model compound for investigating the energy dissipation by a carotenoid. The solvent effects on the fluorescence properties of the dyads **3c**−**5c** suggest that the decrease in fluorescence intensity is due to the intramolecular energy transfer from the porphyrin to carotenoid rather than electron transfer.

Experimental Section

Materials. THF, DMF, and benzene used for spectroscopic measurements were of spectroscopic grade (Nacalai Tesque) and used without further purification. Dichloromethane and toluene (Nacalai Tesque) for synthetic use were distilled twice from calcium hydride under nitrogen. Silica gel used for the purification of the porphyrin−carotenoid molecules by flash column chromatography was purchased from Merck (Art. 7736). All other reagents were reagent grade and used as received.

Porphyrin 3a. A mixture of 1,4-cyclohexanedicarboxylic acid (**6**, 50.0 mg, 0.290 mmol), porphyrin **1** (59.2 mg, 0.0669 mmol), CMPI (78.0 mg, 0.305 mmol) and DMAP (74.2 mg, 0.607 mmol) was refluxed for 3 h in CH_2Cl_2. The reaction mixture was washed with water and dried over Na_2SO_4. After evaporation of the solvent, the residue was chromatographed on silica gel (5%MeOH/CH_2Cl_2) to give **3a** (21.4 mg, 31%).

Porphyrin 3b. A mixture of porphyrin **3a** (7.7 mg, 0.0074 mmol), *p*-toluidine (1.0 mg, 0.0093 mmol), CMPI (3.5 mg, 0.014 mmol), and DMAP (3.5 mg, 0.029 mmol) was refluxed for 7 h in CH_2Cl_2. The reaction mixture was washed with 1 N HCl and then sat. aq $NaHCO_3$, and the organic phase was dried over Na_2SO_4. After evaporation, the residue was chromatographed on silica gel (CH_2Cl_2) to give **3b** (3.0 mg, 35%). UV−vis (λ_{max}/nm, THF) 408 (1), 506 (0.082), 538 (0.020), 576 (0.029), 629 (0.005). ^{1}H NMR ($CDCl_3$) δ 10.20 (s, 2H), 8.00 (d, J = 8.4 Hz, 2H), 7.93 (d, 2H), 7.92 (d, J = 7.8 Hz, 2H), 7.65 (s, 1H), 7.54 (d, J = 7.5 Hz, 2H), 7.45 (d, J = 8.4 Hz, 2H), 7.19 (s, 1H), 7.15 (d, J = 8.1 Hz, 2H), 3.95 (m, 8H), 2.72 (s, 3H), 2.52 (s, 6H), 2.50 (s, 6H), 2.31 (s, 3H), 2.16 (m, 8H), 1.70 (m, 8H), 1.45 (m, 8H), 1.35 (m, 8H), 0.89 (t, 12H), −2.48 (s, 2H). HRMS (FAB, positive) found m/z 1126.7693, calcd for $C_{76}H_{98}N_6O_2$ = 1126.7751 (M^+).

Dyad 3c. A mixture of porphyrin **3a** (8.7 mg, 0.0084 mmol), carotenoid **2** (4.9 mg, 0.0097 mmol), CMPI (4.2 mg, 0.016 mmol), and DMAP (4.1 mg, 0.034 mmol) was refluxed in CH_2Cl_2 for 3 h under N_2. After washing with water and drying over Na_2SO_4, the solvent was evaporated. The residue was purified by silica gel flash column chromatography (CH_2Cl_2) to give **3c** (6.7 mg, 52%). UV−vis (λ_{max}/nm, THF) 409 (1), 448 (0.36), 474 (0.47), 505 (0.45), 576 (0.029), 628 (0.004). ^{1}H NMR ($CDCl_3$) δ 10.21 (s, 2H), 8.02 (d, J = 8.4 Hz, 2H), 7.94 (d, J = 8.7 Hz, 2H), 7.92 (d, J = 7.8 Hz, 2H), 7.59 (d, J = 7.8 Hz, 2H), 7.57 (d, J = 8.1 Hz, 2H), 7.54 (d, J = 8.1 Hz, 2H), 7.53 (s, 1H), 7.42 (d, J = 8.4 Hz, 2H), 6.86 (d, J = 15.6 Hz, 1H), 6.56 (d, 16.2 Hz, 1H), 3.62 (m, 8H), 2.72 (s, 3H), 2.53 (s, 6H), 2.50 (s, 6H), 2.2 (m, 8H), 2.17 (s, 3H), 2.04 (s, 3H), 1.99 (s, 3H), 1.98 (s, 3H), 1.72 (s, 3H), 1.7 (m, 8H), 1.5 (m, 8H), 1.4 (m, 8H), 1.03 (s, 6H), 0.89 (t, J = 7.2 Hz, 12H). HRMS (FAB, positive) found m/z 1525.0656, calcd for $C_{106}H_{136}N_6O_2$ = 1525.0724 (M^+).

Porphyrin 4a. Kemp's acid chloride−anhydride **7** (41.9 mg, 0.162 mmol), porphyrin **1** (117 mg, 0.133 mmol), and 2,6-di-*tert*-butylpyridine (0.10 mL) were dissolved in 10 mL of toluene, and the reaction mixture was refluxed for 10 h under N_2 atmosphere. The solution was washed with water and dried over Na_2SO_4, and then the solvent was evaporated. Purification with silica gel chromatography (1% EtOH/CH_2Cl_2) gave **4a** (121 mg, 82%). ^{1}H NMR ($CDCl_3$) δ 10.24 (s, 2H), 8.10 (d, J = 8.5 Hz, 2H), 7.94 (d, J = 8.0 Hz, 2H), 7.55 (d, J = 7.5 Hz, 2H), 7.44 (br s, 2H), 3.98 (t, J = 7.3 Hz, 8H), 2.93 (d, J = 14.5 Hz, 2H), 2.73 (s, 3H), 2.59 (s, 6H), 2.51 (s, 6H), 2.27 (d, J = 13.5 Hz, 1H), 2.19 (m, 8H), 1.73 (m, 8H), 1.52 (d, J = 13.5 Hz, 1H), 1.47 (m, 8H), 1.44 (s, 6H), 1.37 (m, 8H), 1.29 (s, 3H), 1.28 (d, 2H), 0.91 (t, J = 7.5 Hz, 12H).

Porphyrin 4b. A mixture of porphyrin **4a** (37.2 mg, 0.0336 mmol), *p*-toluidine (37.8 mg, 0.353 mmol), CMPI (19.1 mg, 0.0747 mmol), and DMAP (18.7 mg, 0.153 mmol) was refluxed for 4 h in CH_2Cl_2. After washing with water and then citric acid solution, the organic phase was dried over Na_2SO_4, and the solvent was evaporated. The residue was chromatographed on silica gel (CH_2Cl_2) to give **4b** (22 mg, 55%). UV−vis (λ_{max}/nm, THF) 408 (1), 505 (0.085), 537 (0.021), 576 (0.030), 629 (0.005). ^{1}H NMR ($CDCl_3$) δ 10.20 (s, 2H), 7.92 (d, J = 7.8 Hz, 2H), 7.74 (br s, 2H), 7.53 (d, J = 8.1 Hz), 7.40 (d, J = 8.4 Hz, 2H), 7.38 (s, 1H), 7.22 (d, J = 8.7 Hz, 2H), 7.00 (d, J = 8.4 Hz, 2H), 3.96 (t, J = 7.5 Hz, 8H), 2.97 (d, J = 14.4 Hz, 2H), 2.72 (s, 3H), 2.50 (s, 6H), 2.42 (s, 6H), 2.37 (d, J = 13.5 Hz, 1H), 2.17 (m, 8H), 1.94 (s, 3H), 1.72 (m, 8H), 1.7 (1H), 1.53 (s, 6H), 1.5 (2H), 1.5 (m,

8H), 1.46 (s, 3H), 1.36 (m, 8H), 0.90 (t, J = 7.1 Hz, 6H), 0.89 (t, J = 7.2 Hz, 6H), −2.42 (s, 2H). HRMS (FAB, positive) found m/z 1194.7941, calcd for $C_{80}H_{102}N_6O_3$ = 1194.8013 (M^+).

Dyad 4c. A mixture of porphyrin **4a** (11.6 mg, 0.0105 mmol), carotenoid **2** (10.6 mg, 0.0209 mmol), CMPI (6.8 mg, 0.027 mmol), and DMAP (6.6 mg, 0.054 mmol) was refluxed in CH_2Cl_2 for 11 h under N_2 atmosphere. After washing with water and drying over Na_2SO_4, the solvent was evaporated. The residue was purified by silica gel flash column chromatography (CH_2Cl_2) to give **4c** (5.0 mg, 30%). UV−vis (λ_{max}/nm, THF) 409 (1), 475 (0.37), 506 (0.36), 577 (0.026), 630 (0.003). ^{1}H NMR ($CDCl_3$) δ 10.17 (s, 2H), 7.91 (d, J = 8.1 Hz, 2H), 7.77 (br s, 2H), 7.52 (d, J = 7.8 Hz, 2H), 7.51 (d, J = 8.7 Hz, 2H), 7.45 (s, 1H), 7.29 (d, J = 8.7 Hz, 2H), 7.25 (d, 2H), 6.65 (dd, J = 14.7 Hz, 11.4 Hz, 1H), 6.56 (d, 15.3 Hz, 1H), 6.56 (2H), 6.34 (d, J = 15.0 Hz, 1H), 6.34 (dd, J = 14.7 Hz, 11.3 Hz, 1H), 6.20 (d, J = 16.5 Hz, 1H), 6.2 (d, 1H), 6.15 (2H), 6.14 (1H), 6.08 (d, 1H), 6.06 (d, J = 15.0 Hz, 1H), 5.95 (d, J = 11.4 Hz, 1H), 3.94 (m, 8H), 2.98 (d, J = 14.4 Hz, 2H), 2.71 (s, 3H), 2.50 (s, 6H), 2.39 (s, 6H), 2.37 (d, 1H), 2.14 (m, 8H), 2.0 (m, 2H), 1.97 (s, 3H), 1.96 (s, 3H), 1.80 (s, 3H), 1.72 (s, 3H), 1.6 (m, 2H), 1.6 (d, 1H), 1.58 (s, 3H), 1.55 (s, 6H), 1.5 (d, 2H), 1.5 (m, 2H), 1.47 (s, 3H), 1.03 (s, 6H), 0.92 (t, J = 7.2 Hz, 6H), 0.89 (t, J = 7.2 Hz, 6H), −2.49 (s, 2H). HRMS (FAB, positive) found m/z 1593.0927, calcd for $C_{110}H_{140}N_6O_3$ = 1593.0986 (M^+).

Porphyrin 5a. A mixture of porphyrin **1** (301 mg, 0.340 mmol), xanthenedicarboxylic acid **8** (136 mg, 0.331 mmol), CMPI (104 mg, 0.407 mmol), and DMAP (141 mg, 1.15 mmol) was refluxed in CH_2Cl_2 for 1 h under N_2 atmosphere. After washing with water and then citric acid solution, the organic phase was dried over Na_2SO_4 and the solvent was evaporated. The residue was chromatographed on silica gel (20% hexane/CH_2Cl_2) to give **5a** (282 mg, 65%). ^{1}H NMR (acetone-d_6) δ 10.94 (s, 1H), 10.13 (s, 2H), 8.44 (d, J = 2.4 Hz, 1H), 8.35 (d, J = 8.0 Hz, 2H), 8.10 (s, 1H), 7.99 (s, 1H), 7.92 (d, J = 8.0 Hz, 2H), 7.92 (d, J = 2.7 Hz, 1H), 7.80 (d, J = 7.8 Hz, 2H), 7.52 (d, J = 7.6 Hz, 2H), 3.86 (m, 8H), 2.82 (br s, 3H), 2.66 (s, 3H), 2.54 (s, 6H), 2.42 (s, 6H), 2.14 (m, 8H), 1.82 (s, 3H), 1.72 (m, 8H), 1.47 (s, 9H), 1.44 (m, 8H), 1.42 (s, 9H), 1.37 (m, 6H), 0.89 (t, J = 7.2 Hz, 12H).

Porphyrin 5b. A mixture of porphyrin **5a** (50.4 mg, 0.0395 mmol), *p*-toluidine (42.7 mg, 0.399 mmol), CMPI (24.8 mg, 0.0971 mmol), and DMAP (23.5 mg, 0.192 mmol) was refluxed for 9 h in CH_2Cl_2. It was washed with 1 N HCl and then sat. aq $NaHCO_3$, and the organic phase was dried over Na_2SO_4. After evaporation, the residue was chromatographed on silica gel (CH_2Cl_2) to give **5b** (33.5 mg, 62%). UV−vis (λ_{max}/nm, THF) 408 (1), 505 (0.082), 537 (0.019), 576 (0.029), 628 (0.005). ^{1}H NMR ($CDCl_3$) δ 10.23 (s, 2H), 9.13 (s, 1H), 8.82 (s, 1H), 8.06 (d, J = 2.2 Hz, 1H), 7.95 (d, J = 7 Hz, 2H), 7.93 (d, J = 8 Hz, 2H), 7.91 (d, J = 2.4 Hz, 1H), 7.78 (d, J = 8.3 Hz, 2H), 7.69 (d, J = 2.4 Hz, 1H), 7.69 (d, J = 6 Hz, 2H), 7.67 (d, J = 2.4 Hz, 1H), 7.54 (d, J = 7.8 Hz, 2H), 7.06 (d, J = 8.1 Hz, 2H), 3.99 (t, J = 6.5 Hz, 8H), 2.72 (s, 3H), 2.51 (s, 6H), 2.45 (s, 6H), 2.19 (m, 8H), 1.78 (s, 6H), 1.75 (s, 3H), 1.73 (m, 8H), 1.5 (m, 8H), 1.46 (s, 9H), 1.41 (s, 9H), 1.4 (m, 8H), 0.90 (t, J = 7.2 Hz, 6H), 0.90 (t, J = 7.2 Hz, 6H), −2.38 (br s, 2H). ^{13}C NMR ($CDCl_3$) δ 164.9, 164.8, 146.5, 145.8, 145.8, 145.3, 145.3, 143.1, 143.1, 141.4, 141.4, 139.3, 138.2, 137.9, 136.4, 136.3, 135.4, 134.4, 132.9, 132.8, 130.5, 130.4, 129.3, 128.3, 126.2, 126.1, 125.8, 122.8, 122.0, 120.3, 118.1, 117.5, 96.8, 34.8, 34.8, 34.7, 33.3, 33.3, 32.6, 32.0, 32.0, 31.5, 31.4, 30.0, 30.0, 26.8, 22.8, 21.7, 20.6, 15.0, 14.7, 14.1, 14.1. HRMS (ESI, positive) found m/z 1365.9199, calcd for $C_{93}H_{117}N_6O_3$ = 1365.9187 ($M + H^+$).

Dyad 5c. A mixture of porphyrin **5a** (10.6 mg, 0.0210 mmol), carotenoid **2** (26.3 mg, 0.0206 mmol), CMPI (10.7 mg, 0.0418 mmol), and DMAP (10.8 mg, 0.0883 mmol) was refluxed in CH_2Cl_2 for 3 h under N_2. After washing with water and drying over Na_2SO_4, the solvent was evaporated. The residue was chromatographed on silica gel (10% AcOEt/hexane). Recrystallization from CH_2Cl_2/hexane gave **5c** (19.0 mg, 51%). UV−vis (λ_{max}/nm, THF) 410 (1), 480 (0.42), 508 (0.40), 576 (0.027) and 628 (0.005). ^{1}H NMR ($CDCl_3$) δ 10.18 (s, 2H), 9.07 (s, 1H), 8.93 (s, 1H), 8.04 (d, J = 2.2 Hz, 1H), 8.02 (d, J = 8.3 Hz, 2H), 7.93 (s, 1H), 7.92 (d, J = 7.8 Hz, 2H), 7.89 (d, J = 8.5 Hz, 2H), 7.85 (d, J = 8.3 Hz, 2H), 7.70 (d, J = 2.2 Hz, 1H), 7.68 (d, J = 2.2 Hz, 1H), 7.52 (d, J = 7.6 Hz, 1H), 7.37 (d, J = 8.6 Hz, 2H), 6.68 (m, 1H), 6.47 (d, 2H), 6.39 (d, 1H), 6.39 (d, J = 14.9 Hz, 1H), 6.26 (m, 1H), 6.18 (d, 1H), 6.17 (2H), 6.09 (d, J = 16.1 Hz, 1H), 5.76 (m, 1H), 5.57 (m, 1H), 5.27 (d, J = 11.4 Hz, 1H), 5.22 (d, J = 14.4 Hz, 1H), 3.94 (m, 8H), 2.71 (s, 3H), 2.49 (s, 6H), 2.41 (s, 6H), 2.15 (m, 8H), 2.03 (m, 2H), 2.00 (s, 6H), 1.79 (s, 6H), 1.74 (s, 3H), 1.72 (m, 8H), 1.62 (m, 2H), 1.5 (m, 2H), 1.50 (s, 3H), 1.44 (s, 3H), 1.47 (2, 9H), 1.47 (m, 8H), 1.42 (s, 9H), 1.37 (m, 8H), 1.1 (3H), 1.05 (s, 6H), 0.90 (t, J = 7.2 Hz, 12H), −2.36 (br s, 2H). HRMS (ESI, positive) found m/z 1764.2229, calcd for $C_{123}H_{155}N_6O_3$ = 1764.2161 ($M+H^+$).

Spectroscopic Measurements. Instruments for the measurements of the fluorescence lifetimes were the same as already reported, and the decay time was determined by deconvolution methods.[32]

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Supporting Information Available: ^{1}H NMR spectra of **3**−**5**, 2D COSY spectra of **4c** and **5c**, and the fluorescence decay profiles of **3c**−**5c**. This material is available free of charge via the Internet at http://pubs.acs.org.

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