Molecular Assembly of Zinc–Nickel Hybrid Porphyrin Dimer Using Synthetic 4α-Helix Polypeptides

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Molecular assembly of hybrid porphyrin dimer containing zinc and nickel mesoporphyrins using synthetic 4α-helix polypeptides induced an efficient energy-transfer between the porphyrins due to the presence of the polypeptide.

Synthetic porphyrin dimer analogs can be helpful in investigating the effect of porphyrin structure on energy-transfer process in photosynthesis.[1,2] Especially, porphyrins play a key role in the energy transfer in light-harvesting (LH) polypeptide complexes of photosynthetic bacteria, where porphyrins such as bacteriochlorophylls (BChls) are assembled according to cooperative interactions between the BChls and the LH polypeptides.[3–6] Several laboratories have demonstrated self-assemblies of porphyrins using synthetic polypeptides to organize an artificial hemoprotein models. However, there has been little study of molecular assembly of porphyrins using synthetic polypeptides to organize an artificial LH model complex.[2,7] Furthermore, several covalently-linked porphyrin dimers have been studied as models for charge separation, electron-transfer process, and recombination that are involved in early photosynthetic events. However, an artificial design of an energy-transfer system of porphyrin dimer using synthetic LH model polypeptides is not well studied.[2,7]

Herein covalently-bridged mesoporphyrin dimers with an L-lysine residue (Figure 1) are assembled using synthetic polypeptide, HAHA and HHHH (Figure 2). HAHA and HHHH were designed as models of LH polypeptides from photosynthetic bacteria.[2] It is considered that these synthetic polypeptides display a 4α-helical bundled structure in an aqueous condition,

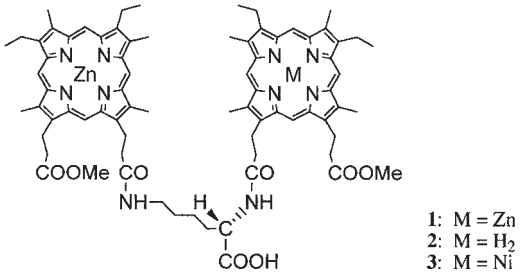

Figure 1. Chemical structures of mesoporphyrin dimers.

DAPGELLKAHAELLK-DAPGELLKAAAELLK-DAPGELLKAHAELLK-DAPGELLKAAAELLK-Naf
DAPGELLKAHAELLK-DAPGELLKAHAELLK-DAPGELLKAHAELLK-DAPGELLKAHAELLK-Naf

Figure 2. Amino acid sequences of HAHA and HHHH. DA and Naf represent D-alanine and naphthylalanine, respectively.

and the histidine residues are located at the hydrophobic region in the helical rods. Previous studies indicated that the molecular assembly with the LH-model polypeptides, in particular HAHA, supported a well-ordered BChl aggregation with axial coordination comparable to that in a native BChls/LH polypeptide complex.[2,7] Our interest in dimeric donor–acceptor hybrid metalloporphyrins assembled with these synthetic model polypeptides is further motivated by the fact that this assembly will be useful for constructing artificial efficient energy-transfer systems in aqueous solutions. L-lysine-bridged mesoporphyrin dimers **1–3** were prepared as described previously.[2,8] Zinc porphyrin as a donor pigment possessed not only an excellent axial coordination ability for a histidine residue but also the ability of the high fluorescence. Davis et al. have reported a successful reconstitution of nickel-substituted BChl analog with LH polypeptides,[9] so that nickel porphyrins will be utilized in constructing the molecular assembly using these LH polypeptides. Furthermore, nickel complexes act as an acceptor pigment due to the ability of the strong fluorescence quenching. Synthetic 4α-helix polypeptides, HHHH and HAHA were prepared and purified by Sephadex LH-60 gel chromatography and then HPLC.[7] These polypeptides containing 57 amino acids were analyzed by TOF-MS given the expected molecular mass.

Initially, intramolecular energy transfer efficiencies for **2** and **3** were characterized in DMSO by fluorescence lifetime measurements as shown in Table 1. The results indicated that an unidirectional rapid energy transfer was detected at 100 ps in **3**, while the energy transfer of **2** was found to be slower in comparison to that of **3**. The data shows that an efficient energy transfer between porphyrins occurrs for **3** rather than for **2**. However, the intramolecular energy transfer efficiencies for **2** and **3** in the presence of HHHH and HAHA were not characterized because the decay of the fluorescence for **2** or **3** was too fast to detect the lifetime due to the presence of these polypeptides in the aqueous solutions. Thus, the efficient energy transfer in mesoporphyrin derivatives was evaluated by fluorescence emission spectrum as shown in Table 2. As is apparent from Table 2, a significant energy transfer occurred in assembled **3** with HAHA in *n*-octyl-β-D-glucopyranoside (OG) micelle, detecting relative $I_{H|A} = 3.60$, rather than with HHHH, detecting relative $I_{H|A} = 2.02$. Figure 3 shows the CD spectra of **3** in the presence of HAHA or HHHH. The splitted CD spectrum was observed due to the presence of HAHA and HHHH at 4 °C, where a large splitted CD was observed especially in the presence of HAHA rather than HHHH. No CD signal was detected for **3** in the

Table 1. Fluorescence Lifetimes and Quantum Efficiencies of Energy Transfer in DMSO

Compound[a]	λ_{ex}[c]/nm	Donor			Acceptor		
		λ_{em}[d]/nm	τ[e]/ns	Φ[f]	λ_{em}[d]/nm	τ[e]/ns	Φ[f]
ZnMPMM							
E[b]	400	579	2.40	100%	—	—	—
3	400	579	0.95	043%	687	02.50	100%
			0.10	057%			
2	532	581	2.60	037%	624	10.50	060%
			0.40	063%		00.46	040%

[a]All concentrations of mesoporphyrin were 3.45×10^{-6} mol dm^{-3}. [b]Zinc mesoporphyrin monomethyl ester. [c]Excitation wavelength. [d]Fluorescence emission wavelength. [e]Fluorescence lifetime. [f]The quantum efficiency of energy transfer.

Table 2. Spectral Parameters of Visible Absorption and Quantum Efficiency of Energy Transfer for Mesoporphyrins in the Presence and Absence of HAHA and HHHH

Compound	Peptide[c]	Soret peaks/	rel
		4 °C	4 °C
2[a]	none	395.0	—[i]
2[a]	HAHA[d]	396.5, (411.5)[e]	—[i]
3[a]	none	392.0	2.31
3[a]	HAHA[d]	391.5, 413.5	3.60
3[a]	HHHH[d]	392.5, 414.0	2.02
monomer[b]	none	391.0	2.14
monomer[b]	HAHA[d]	392.5, 415.0	1.54

[a]1.73×10^{-6} mol dm^{-3}. [b]A heterogeneous mixture of zinc and nickel mesoporphyrins. zinc mesoporphyrin, 1.73×10^{-6} mol dm^{-3}; nickel mesoporphyrin, 1.73×10^{-6} mol dm^{-3}. [c]In phosphate buffer (pH 7.0) in the presence of 26.5×10^{-3} mol dm^{-3} of OG. [d]1.73×10^{-6} mol dm^{-3}. [e]Parentheses represent an inconspicuous shoulder peak. [f]Excitation wavelength is set at 404.0, 414.5, and 416.0 nm in the presence of HAHA or HHHH. rel $I_{H|A} = I_{H|A}/I_{0|A}$. $I_{H|A}$ is the fluorescence emission intensity of **2** and **3** at 625.0 and 690.5 nm, respectively, and $I_{0|A}$ is that of free-base and nickel mesoporphyrin monomers. [i]No significant luminescence was observed.

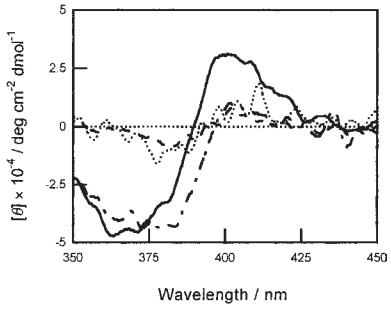

Figure 3. CD spectra of **3** in the presence of HAHA or HHHH. Lines represent **3** in the absence of HAHA at 4 °C (dotted), **3** in the presence of HAHA at 25 °C (dashed), **3** in the presence of HAHA at 4 °C (solid), and **3** in the presence of HHHH at 4 °C (dashed and dotted). **3** was assembled with HAHA or HHHH in phosphate buffer (pH 7.0) in the presence of 26.5×10^{-3} mol dm^{-3} of OG. **3**, 1.73×10^{-6} mol dm^{-3}; HAHA, 1.73×10^{-6} mol dm^{-3}; HHHH, 1.73×10^{-6} mol dm^{-3}.

absence of polypeptides. This result shows that **3** was easily assembled in HAHA rather than HHHH, coordinating with histidine residues of the polypeptides.[7] This coordination was consistent with the red-shifted absorption of the Soret band for **3** due to the presence of the polypeptides (the data not shown).[7b] These results implied that the axial-coordination of the Zn atom in the porphyrin dimer with the histidine residue in the hydrophobic core of HAHA caused the red-shift especially at low temperature, consistent with histidine-linked Zn mesoporphyrin, ZnMPMME-L-HisOMe in $CHCl_3$. Thus, it is assumed that a successful form of the molecular assembly of **3** generated the efficiency of energy transfer especially due to the presence of HAHA. In contrast, an enhanced energy transfer due to the presence of HAHA was not observed for heterogeneous mixtures of donor and acceptor mesoporphyrins as well as for **2** in comparison to **3** (Table 2). The reasons are that 1) the monomeric mixtures as well as **2** were not well assembled with HAHA, because the nickel monomer or free-base mesoporphyrin could not coordinate with the histidine residues of the polypeptide (the data not shown) and 2) the monomeric mixtures have the low energy transfer efficiency between porphyrins as described above (Table 1). No or little splitted CD signal was observed for the dimethyl ester of **3** due to the presence of HAHA. These results implied that the zinc atom and the carboxyl group in the mesoporphyrin dimer played crucial roles on the molecular assembly, where the carboxyl group on the dimer may bind with polar amino acid residues in HAHA through hydrogen bonding.[2]

In conclusion, molecular assemblies of zinc–nickel porphyrin hybrid dimers with HAHA induced an efficient intramolecular energy transfer from zinc porphyrin to nickel porphyrin and these assemblies will be useful for constructing an artificial efficient energy-transfer system in aqueous solutions.

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8 ^{1}H NMR spectra: **1** and **2** were unambiguously supported those assigned structures. Mass spectra: FAB, m/z; 1398 (M) for **1**; 1335 (M) for **2**; 1391 (M) for **3**.

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