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Intermolecular interaction between multifunctional porphyrin and ubiquinone analogues

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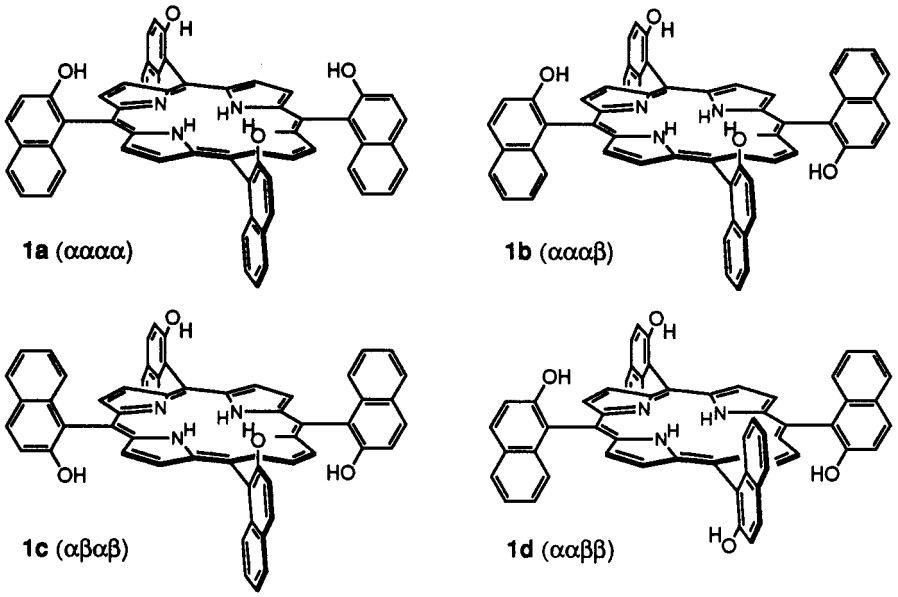
Abstract: Face-to-face complexation between *meso*-tetra(2-hydroxynaphthyl)porphyrin (**1**) and *p*-benzoquinone derivatives via hydrogen bonds has been clarified by spectroscopic measurements. Formation of quinone-porphyrin complex depends upon orientation of hydroxyl groups above and below porphyrin ring. Among them, α,α,α,α-atropisomer (**1a**) having four convergent hydroxyl groups shows most efficient multipoint interaction with ubiquinone analogues. The binding constants (K_a) and thermodynamic parameters (ΔG°, ΔH°, $T\Delta S^\circ$) of quinones with **1** largely depend on the number and position of methoxy substituents on quinone ring. Tetramethoxy-*p*-benzoquinone (**2e**) is most favorable guest among ubiquinone analogues and its binding constant is determined as $K_a = 2.0 \times 10^4$ M^{-1} at 298 K in $CHCl_3$. The present porphyrin-quinone pair, which is mainly governed by specific hydrogen bonding fixation, is quite difference from the system of two-point fixation by 5,15-*cis*-bis(2-hydroxynaphthyl)-octaethylporphyrin (**3**) reported in previous work.

INTRODUCTION

In respiratory system, it is well known that ubiquinone (coenzyme Q_{10}) plays an important role as electron and proton carrier. The quinone derivatives having a long isoprenoid tail are freely movable to carry the electron and/or proton from reductase to oxidase in the lipid layer (ref. 1). Thus, noncovalent interaction of ubiquinone at the both redox reaction sites in enzymes may be essential to regulate the rate of respiratory electron transfer via molecular recognition (ref. 2). Many porphyrins covalently linked with quinone have been synthesized to elucidate mechanism of electron transfer relevant to photosynthesis (ref. 3). However, investigations for noncovalent porphyrin-quinone interactions are very few (ref. 4,5). Recently we have reported the intermolecular interaction between various quinones **2a-i** and porphyrin, *meso*-tetra(α,α,α,α-2-hydroxynaphthyl)porphyrin (**1a**), via multipoint hydrogen bonds (ref. 6). In this paper, the complexation properties of **1a** are reviewed. The interactions between ubiquinone analogues and other atropisomers, α,α,α,β- and α,β,α,β-isomers (**1b,c**) of *meso*-tetra(2-hydroxynaphthyl)porphyrin (**1**) are described. In addition, the binding mode with quinones between present host **1a** and previous host molecule, 5,15-*cis*-bis(2-hydroxynaphthyl)octaethylporphyrin (**3**), are compared.



PREPARATION AND CHARACTERIZATION OF HOST MOLECULES

These host molecules are prepared by the usual porphyrin synthesis on the condensation of 2-methoxy-1-naphthaldehyde and pyrrole followed by demethylation of four methoxy groups in 6.1% yield. The porphyrin thus obtained includes mixture of four atropisomers (α,β,α,β-, α,α,β,β-, α,α,α,β-, and α,α,α,α-isomer in the ratio of 1:2:4:1). Each atropisomer was easily separated by silica gel column chromatography (R_f = 0.63, 0.38, 0.08, and 0.01, respectively; benzene/AcOEt = 10 v/v) and determined by ^1H NMR and FAB Mass spectroscopies. Especially, β-proton signals of pyrrole ring of host **1** enable us to differentiate four atropisomers: coupling patterns of these proton signals in α,β,α,β-, α,α,β,β-, α,α,α,β-, and α,α,α,α-atropisomers appear as singlet, doublet, multiplet, and singlet, respectively. Atropisomerization of **1** due to carbon-carbon bond rotation between porphyrin ring and naphthyl group was not detected after boiling in toluene for 2h.

COMPLEXATION BETWEEN PORPHYRIN 1 AND QUINONE 2e

Porphyrin **1a**, having four convergent hydroxyl groups, can specifically bind with tetramethoxy-*p*-benzoquinone (**2e**) at 1:1 stoichiometry. Face-to-face complexation between **1a** and **2e** was confirmed by the downfield shift (+1.91 ppm) of the hydrogen-bonding OH of **1a** and upfiled shift (-0.93 ppm) of the OCH_3 of quinone due to ring current of the porphyrin macrocycle in the ^1H NMR spectra. Furthermore, IR spectrum of an approximately 2:1 mixture of **1a** and **2e** in $CHCl_3$ showed two absorptions at 3449 cm^{-1} and 3544 cm^{-1} assignable to the hydrogen-bonding OH and free OH stretching vibrations, respectively. According to these results and CPK molecular modeling, the distance between porphyrin and quinone ring is approximately estimated about 3.5 Å. These affinities were determined by titrimetric measurement of visible spectra, which show clear isosbestic points in the region of 550-700 nm. Figure 1 represents the schematic complexation of top view and side view of **1a-2e** adducts. Two other host porphyrins, **1b** and **1c**, which are atropisomers of **1a**, also show the binding affinities with ubiquinone analogues via face-to-face interaction mode observed by ^1H NMR and electronic absorption spectroscopies.

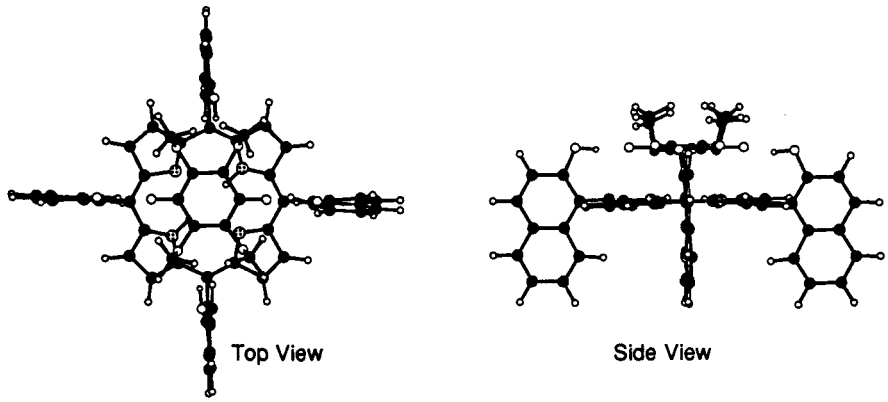
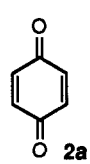
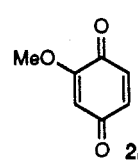
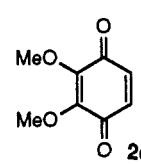
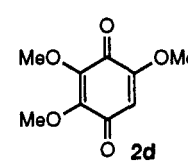
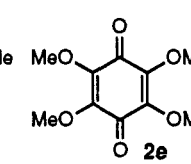


Figure 1. Schematic representation of porphyrin **1a** - quinone **2e** adduct

AFFINITIES OF UBIQUINONE ANALOGUES WITH HOST 1a

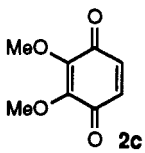
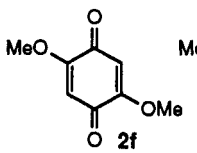
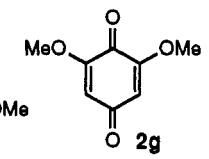
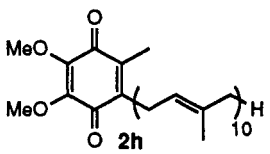
The binding constants (K_a) determined from the non-linear curve fitting analysis and the thermodynamic parameters (ΔG°, ΔH°, $T\Delta S^\circ$) for **1a-2** are listed in Table 1 and 2. Characteristic properties of binding affinities are summarized as follows. (i) The binding constants of quinone with **1a** increase with the number of substituted methoxy groups on quinone ring (**2a** < **2b** < **2c** < **2d** < **2e**). (ii) The favorable negative changes of enthalpy increase in the same order as above. In contrast, the remarkable entropy changes are not observed among **2a-e** ($T\Delta S^\circ$ = -3.8 ~ -4.7 kcal/mol). (iii) According to the calculation of free energy, the difference in the negative gains of free energy change ($\Delta\Delta G^\circ$) upon the substitution of methoxy groups at the adjacent positions ($\Delta\Delta G^\circ$(**1a-2b** → **1a-2c**) = ΔG°(**1a-2c**) — ΔG°(**1a-2b**) and $\Delta\Delta G^\circ$(**1a-2d** → **1a-2e**) = ΔG°(**1a-2e**) — ΔG°(**1a-2d**)) are determined as approximately -1.5 kcal/mol, whereas these values at the separated positions ($\Delta\Delta G^\circ$(**1a-2a** → **1a-2b**) = ΔG°(**1a-2b**) — ΔG°(**1a-2a**) and $\Delta\Delta G^\circ$(**1a-2c** → **1a-2d**) = ΔG°(**1a-2d**) — ΔG°(**1a-2c**)) are about -0.5 kcal/mol. Table 2 shows that two adjacent methoxy groups in 2,3-dimethoxy-*p*-benzoquinone (**2c**) cooperate to form the favorable hydrogen

TABLE 1. Binding constants (K_a) and thermodynamic parameters (ΔG°, ΔH°, $T\Delta S^\circ$) for porphyrin **1a** and ubiquinone analogues **2** complexation in $CHCl_3$ at 298 K[a].

quinone:	 **2a**	 **2b**	 **2c**	 **2d**	 **2e**
K_a (M^{-1})	3.0 x 10	7.4 x 10	8.3 x 10^2	1.6 x 10^3	2.0 x 10^4
ΔG° (kcal/mol)	-2.0	-2.5	-4.0	-4.4	-5.8
ΔH° (kcal/mol)	-6.5	-6.8	-7.8	-9.0	-10.5
$T\Delta S^\circ$ (kcal/mol)	-4.5	-4.2	-3.8	-4.7	-4.7

[a] Binding constants and thermodynamic parameters were obtained from electronic absorption studies at 584 nm.

TABLE 2. Binding constants (K_a) and free energy ($\Delta G°$) for porphyrin **1a** and dimethoxy substituted quinone **2** complexation in $CHCl_3$ at 298 K[a].

quinone:	 **2c**	 **2f**	 **2g**	 **2h**
K_a (M^{-1})	8.3×10^2	1.9×10^2	1.7×10^2	1.6×10^2
$\Delta G°$ (kcal/mol)	-4.0	-3.1	-3.0	-2.9

bond with **1a** compared with two separated methoxy groups in 2,5-dimethoxy-*p*-benzoquinone (**2f**) and 2,6-dimethoxy-*p*-benzoquinone (**2g**). (iv) Although a long isoprenoid tail of ubiquinone (coenzyme Q_{10}) (**2h**) may bring about steric repulsion with naphthyl group, its binding constant with **1a** is larger than those of **2a** and **2b**. These results indicate that the OCH_3 substituents at 2- and 3-positions of the *p*-benzoquinone ring cooperatively act as the effective third or fourth interaction site via "bifurcated" hydrogen bonding between hydroxyl group and two oxygens of methoxy groups. It is most likely that simultaneous multipoint hydrogen bonds give rise to an extremely large binding constant of **2e** with **1**.

INTERACTION BETWEEN QUINONE AND EACH ATROPISOMER OF PORPHYRIN 1

Ubiquinone analogues, **2a-e**, also interact with α,α,α,β- and α,β,α,β-isomers (**1b,c**), but the binding affinities are smaller than that with α,α,α,α-isomer (**1a**). Table 3 shows the comparison of the binding constants of **2a**, **2c**, **2e** for three atropisomers, **1a-c**. Face-to-face interaction operating in pairs of atropisomers and quinones are mostly hydrogen bonds and steric repulsion. The large difference in these affinities results from the following two factors. Firstly, **1a** has four cooperative interaction sites through hydrogen bonds, whereas **1b** and **1c** have three and two interaction sites, respectively. Secondly, the overturned naphthyl groups as are seen in α,α,α,"β"- or α,"β",α,"β"-isomers bring about significant steric repulsion with methoxy groups of quinone. Low binding constants and small free energy changes of simple benzoquinone **2a** for three atropisomers **1a-c** imply that two hydrogen bonds between the carbonyl

TABLE 3. Binding constants of quinones with each atropisomer[a].

host	quinone		
	2a	**2c**	**2e**
1a (αααα)[b]	$K_a = 3.0 \times 10\ M^{-1}$ $\Delta G° = -2.0$ kcal/mol	$K_a = 8.3 \times 10^2\ M^{-1}$ $\Delta G° = -4.0$ kcal/mol	$K_a = 2.0 \times 10^4\ M^{-1}$ $\Delta G° = -5.8$ kcal/mol
1b (αααβ)[b]	$K_a = 1.2 \times 10\ M^{-1}$ $\Delta G° = -1.5$ kcal/mol	$K_a = 6.7 \times 10\ M^{-1}$ $\Delta G° = -2.5$ kcal/mol	$K_a = 2.6 \times 10^2\ M^{-1}$ $\Delta G° = -3.3$ kcal/mol
1c (αβαβ)[c]	$K_a = 4\ M^{-1}$ $\Delta G° = -0.8$ kcal/mol	$K_a = 5\ M^{-1}$ $\Delta G° = -1.0$ kcal/mol	$K_a = 1.1 \times 10\ M^{-1}$ $\Delta G° = -1.4$ kcal/mol

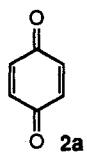
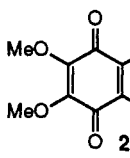
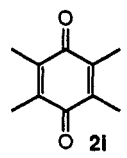
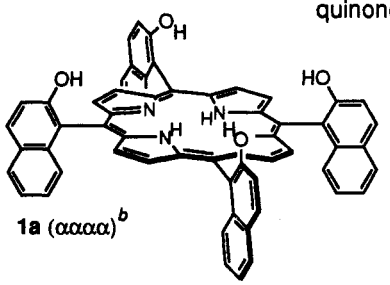
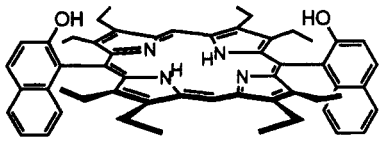
[a] at 298 K, in $CHCl_3$. [b] Binding constants and free energies were obtained from electronic absorption studies at 584 nm. [c] Binding constants and free energies were obtained from ^1H NMR studeis in $CDCl_3$.

groups of quinone and hydroxyl groups of porphyrin are not enough to stabilize the face-to-face adduct. Steric hindrance of the naphthyl group at the binding face is not negligible in spite of no bulky substituents in **2a**. Thus, the number and position of binding sites of both host and guest molecules are crucial for complex formation.

COMPARISON OF BINDING CONSTANTS BETWEEN 1a AND PREVIOUS PORPHYRIN HOST 3

Recently, we have reported the host molecule 5,15-*cis*-bis(2-hydroxynaphthyl)octaethylporphyrin (**3**) capable of binding affinities with various quinones via two-point hydrogen bonds (ref. 4). Thus, it is of particular interest to compare the binding constants and thermodynamic parameters of quinones for **1** and the previous host **3** substituted with two hydroxynaphthyl groups at the *meso*-positions and eight peripheral ethyl groups at the β-positions of pyrrole ring. Table 4 summarizes the binding constants and their thermodynamic parameters for **1a** and **3** with quinone **2a**, **2e** and **2i**. Binding affinities of the substituted quinones, **2e** and **2i** with **1a** and **3** show sharp contrast, whereas no significant difference was found for complexation between **2a** and two host porphyrins. The binding constant of **1a**-**2e** pair is ca. 2500 times larger than that of **3**-**2e** pair. Negative enthalpy change on complexation of **1a**-**2e** is also surprisingly larger than that of the **3**-**2e** pair. Although the four OCH_3 groups of **2e** may bring about the repulsive interaction to the 2-hydroxynaphthyl groups and weakening charge-transfer type interaction to some extent, efficient and cooperative hydrogen bonds in **1a**-**2e** pair contribute to marked stabilization of cofacial adduct. In contrast, tetramethyl-*p*-benzoquinone (**2i**) can form less favorable complexation with **1a** compared with **3** due to a repulsive interaction between 2-hydroxynaphthyl groups and four methyl groups of quinone. These results indicate that the interaction mode of **1a** with quinone is quite different from that of previous host **3**. Interaction operating in the complex of **3** and quinones is mostly due to electronic effect of substituents of quinones and charge-transfer interaction. Thus, particular porphyrin **1a** is regarded as a good host molecule for adjacent di- and/or tetramethoxy substituted quinones like ubiquinone (coenzyme Q_n).

TABLE 4. Comparison of Binding Constants (K_a)[a] and Thermodynamic Parameters ($\Delta G°$, $\Delta H°$, $T\Delta S°$) [a] between **1** and **3** at 298 K.

host molecule	quinone:  **2a**	 **2e**	 **2i**
 **1a** (αααα)[b]	K_a = 3.0 x 10 $\Delta G°$ = -2.0 $\Delta H°$ = -6.5 $T\Delta S°$ = -4.5	K_a = 2.0 x 10^4 $\Delta G°$ = -5.8 $\Delta H°$ = -10.5 $T\Delta S°$ = -4.7	K_a = 1.0 x 10 $\Delta G°$ = -1.4 $\Delta H°$ = -5.6 $T\Delta S°$ = -4.2
 **3** (*cis*)[c]	K_a = 5.5 x 10 $\Delta G°$ = -2.4 $\Delta H°$ = -5.6 $T\Delta S°$ = -3.3	K_a = 7.8 $\Delta G°$ = -1.2 $\Delta H°$ = -4.3 $T\Delta S°$ = -3.1	K_a = 4.2 x 10^2 $\Delta G°$ = -3.6 $\Delta H°$ = -9.0 $T\Delta S°$ = -5.5

[a] In kcal/mol. [b] Binding constants and thermodynamic parameters were obtained from electronic absorption studies in $CHCl_3$. [c] Binding constants and thermodynamic parameters were obtained from ^1H NMR studies in $CDCl_3$.

The sequence analysis of the ubiquinone binding protein, QCP-C, has indicated high population of tyrosine at the binding site (ref. 7). Consequently the movable ubiquinone seems to interact with the tyrosine residues of particular membrane-bound protein via hydrogen bonds and accept electrons from reductase. The present porphyrin host having multi-interaction sites is considered to be a suitable model of the binding site for ubiquinones in the respiratory electron transfer system.

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