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Stereoselective Photoinduced Electron-Transfer Reaction between Zinc Myoglobin and New Chiral Quinolinium Ions

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New chiral quinolinium derivatives, 1-[(*S*)- or (*R*)-(1-phenylethyl)carbamoylmethyl]-6-methoxyquinolinium hexafluorophosphate ([PMQ]PF_6, **2**) and 1-[(*S*)- or (*R*)-(1-phenylethyl)carbamoylmethyl]quinolinium hexafluorophosphate ([PQ]PF_6, **3**), were synthesized and characterized. A cyclic voltammetry in MeCN shows an irreversible redox behavior at −0.88 V and −0.85 V vs SCE (saturated calomel electrode) for **2** and **3**, respectively. Fluorescence from the quinolinium moiety of **2** and **3** was observed at 455 nm and 440 nm, respectively. The fluorescence lifetime of **2** was longer than that of **3**: τ_f = 30 ns (**2**) and 20 ns (**3**) in MeCN and τ_f = 26 ns (**2**) and 16 ns (**3**) in H_2O). The excited triplet state of zinc-substituted myoglobin (^{3}(ZnMb)*) was quenched by chiral $[PMQ]^+$ and $[PQ]^+$ ions; thereafter, the back electron-transfer (ET) reaction from a quinoline radical ($PMQ^\bullet$ or $PQ^\bullet$) to a zinc myoglobin radical cation was detected. The stereoselectivity was observed for both ET quenching and back ET reactions; the (*S*)-isomers preferentially quench ^{3}(ZnMb)* ($k_q(S)/k_q(R)$ = 1.3 and 1.4 at 25 °C for $[PMQ]^+$ and $[PQ]^+$, respectively); in contrast, the (*R*)-isomers react faster than the (*S*)-isomers in the back ET reaction ($k_b(R)/k_b(S)$ = 1.3 and 1.4 for $PMQ^\bullet$ and $PQ^\bullet$, respectively). From a comparison of the rate constants with those for the previously reported systems we suggest that both the quenching and back reactions are controlled by ET, but not by conformational gating.

Specific recognition and binding between a biomacromolecule, such as protein and enzyme, and a small molecule or ion must play an important role in biological functions. Especially, stereoselectivity in the electron-transfer (ET) reactions between metalloproteins and chiral materials is one of the important problems to elucidate the mechanisms of redox catalysis in biological systems. Although a number of studies on stereoselective ET reactions between metalloproteins and chiral metal complexes have been reported,[1–6] there are no reports on the stereoselectivity in the ET reactions of metalloproteins with chiral organic redox reagents, except for ours on chiral viologens.[7–10] One of the reasons is a lack of systematic synthesis of such chiral materials.

Quinoline is a basic unit of an antimalarial agent, such as quinoline alkaloid found in citrus fruits. *N*-Substituted quinolinium compounds are useful agents to detect intracellular Cl^- ions by using fluorescence quenching of the quinolinium moiety through an electron-transfer (ET) mechanism.[11] Zwitter ionic [1-(6-methoxy)quinolinio]propyl-3-sulfonate (SPQ)[12–16] and halide salts, 1-ethyl-6-methoxyquinolinium iodide ([MEQ]I)[17,18] and 1-(ethoxycarbonylmethyl)-6-methoxyquinolinium bromide ([MQAE]Br)[18,19] have been widely used to measure intracellular Cl^- activity. Studies on 1-methylquinolinium perchlorate ([MQ]ClO_4) and 1-methyl-6-methoxyquinolinium perchlorate ([MMQ]ClO_4) have also been reported.[11,20–24] However, because few examples of biologically important chiral quinolinium ions have been reported, it is necessary to develop artificial chiral agents.

In this work we report on new chiral quinolinium derivatives (Chart 1) which show interesting luminescence properties and stereoselectivity in both ET quenching and back ET reactions.

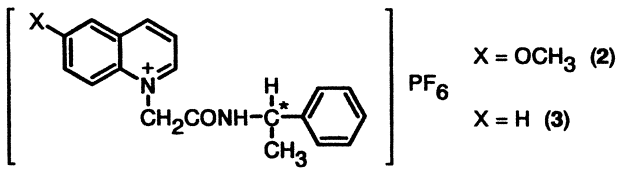


Chart 1. Structure.

It has been suggested that a conformational gating controls the intermolecular ET reactions of ZnMb, zinc hemoglobin, and the other metalloproteins.[25–35] Is there any ET-controlled quenching reaction for ZnMb other than intramolecular ET reactions? If the driving force of the reaction becomes small and, therefore, the ET rate decreases, it might be possible that the reaction is not controlled by a conformational change, but by ET. We have chosen quinolinium compounds to find the ET-controlled photoinduced reaction of ZnMb, because the redox potential of the quinolinium ions is much lower than that of viologens and close to that of the couple of the radical cation ($ZnMb^{\bullet+}$) with the excited triplet state of ZnMb (^{3}(ZnMb)*).

Experimental

Materials. Horse heart metmyoglobin (metMb, Sigma) was purified as previously described.[36,37] Zinc-substituted myoglobin (ZnMb) was prepared at 4 °C in the dark by the same method as reported previously.[28,29,38,39] The concentrations of ZnMb were determined spectrophotometrically ($\varepsilon_{428} = 1.53 \times 10^5$ dm^3 mol^{-1} cm^{-1}).[39] Chiral quinolinium compounds were synthesized by the reaction of quinoline or 6-methoxyquinoline with chiral iodo[(1-phenylethyl)carbamoyl]methane. Hexafluorophosphate salts of

chiral quinolinium compounds were converted to chloride salts by ion-exchange chromatography for kinetic and the other measurements in aqueous solutions. All other chemicals used were of reagent grade. All of the aqueous solutions were prepared from redistilled water. The ionic strength (I) of the solution was adjusted with NaCl.

Synthesis of Iodo[(*S*)-(1-phenylethyl)carbamoyl]methane ((*S*)-PI, 1a) and Iodo[(*R*)-(1-phenylethyl)carbamoyl]methane ((*R*)-PI, 1b). To a stirred solution of iodoacetic acid (11.2 g, 0.0602 mol) in dichloromethane (DCM, 60 cm^3) was slowly added *N*,*N*-dicyclohexylcarbodiimide (12.4 g, 0.0601 mol) in DCM (40 cm^3) at −5 °C. After the mixture was stirred for 30 min, chiral 1-phenylethylamine (7.21 g, 0.0600 mol) was added dropwise. Stirring was continued for a further 90 min, after which the mixture was warmed to room temperature. After the mixture was filtered, 60 cm^3 of ice water was added into the DCM solution. The organic layer was washed successively with 2 mol dm^{-3} HCl, saturated $NaHCO_3$, and saturated NaCl aqueous solutions (100 cm^3 each). After the DCM solution was dried over anhydrous Na_2SO_4 overnight, removal of solvent gave a pale-yellow powder. Recrystallization from MeOH yielded pale-yellow crystals. Yield, 6.91 g (39.8%) for **1a**, 6.32 g (36.4%) for **1b**. ^{1}H NMR (270 MHz, DMSO-d_6, TMS) δ 1.51 (3H, d, J = 6.8 Hz, CH_3), 3.70 (2H, s, CH_2), 5.04–5.14 (1H, m, CH), 6.28 (1H, br, NH), 7.27–7.40 (5H, m, C_6H_5). IR (KBr, ν/cm^{-1}) 1583 (C=C), 1658 (C=O), 3330 (NH). Found: C, 42.01; H, 4.32; N, 4.93% (**1a**), C, 42.02; H, 4.27; N, 4.90% (**1b**). Calcd for $C_{10}H_{12}NOI$: C, 41.54; H, 4.19; N, 4.85%.

Synthesis of (*S*)- and (*R*)-Isomers of Chiral Quinolinium Compounds. An iodo derivative, **1a** or **1b** (5.78 g, 0.0200 mol), in *N*,*N*-dimethylformamide (DMF, 6 cm^3) was heated at 85 °C under N_2, to which quinoline or 6-methoxyquinoline (0.010 mol) in DMF (10 cm^3) was slowly added over a period of 2 h. After the solution was further heated for 24 h, the solvent was removed by a rotary evaporator. The residue was dissolved in MeOH (20 cm^3) and passed through a Dowex 1-X8 (a Cl^- form) column to convert to chloride salts. After the solvent was removed, the residue was dissolved in water (30 cm^3), and then washed with the same volume of DCM several times. The aqueous phase was evaporated to dryness and the residue was redissolved in a small amount of water. The addition of a saturated aqueous solution of $NaPF_6$ gave a white powder. Recrystallization from MeOH gave white crystals.

1-[(*S*)-(1-Phenylethyl)carbamoylmethyl]-6-methoxyquinolinium Hexafluorophosphate ((*S*)-[PMQ]PF_6, 2a) and 1-[(*R*)-(1-Phenylethyl)carbamoylmethyl]-6-methoxyquinolinium Hexafluorophosphate ((*R*)-[PMQ]PF_6, 2b). Yield, 1.82 g (39.0%) for **2a**, 1.90 g (40.7%) for **2b**. ^{1}H NMR (270 MHz, DMSO-d_6, TMS) δ 1.43 (3H, d, J = 6.8 Hz, CH_3), 3.99 (3H, s, OCH_3), 4.87–4.93 (1H, m, CH), 5.82 (2H, s, CH_2), 7.20–7.39 (5H, m, C_6H_5), 7.88 (1H, d, J = 2.4 Hz, 5-quinolinium), 7.90 (1H, dd, J = 2.4, 9.8 Hz, 7-quinolinium), 8.15 (1H, d, J = 9.8 Hz, 8-quinolinium), 8.18 (1H, dd, J = 5.8, 8.3 Hz, 3-quinolinium), 9.13 (1H, br, NH), 9.16 (1H, d, J = 8.3 Hz, 4-quinolinium), 9.28 (1H, d, J = 5.8 Hz, 2-quinolinium). UV-vis (MeCN, λ_{max}/nm (ε/dm^3 mol^{-1} cm^{-1})) 253 (3.24×10^5), 318 (1.02×10^5), 355 (6.54×10^4). ORD (c 0.25 in MeCN, 20 °C) $[\Phi]_{589}$ 89° for **2a**, −88° for **2b**. IR (KBr, ν/cm^{-1}) 829 (PF), 1534 (C=C), 1686 (C=O), 3417 (NH). Found: C, 51.51; H, 4.55; N, 5.98% (**2a**), C, 52.02; H, 4.57; N, 6.00% (**2b**). Calcd for $C_{20}H_{21}N_2O_2PF_6$: C, 51.51; H, 4.54; N, 6.01%.

1-[(*S*)-(1-Phenylethyl)carbamoylmethyl]quinolinium Hexafluorophosphate ((*S*)-[PQ]PF_6, 3a) and 1-[(*R*)-(1-Phenylethyl)carbamoylmethyl]quinolinium Hexafluorophosphate ((*R*)-[PQ]PF_6, 3b). Yield, 1.75 g (40.1%) for **3a**, 1.65 g (37.8%) for **3b**. ^{1}H NMR (270 MHz, DMSO-d_6, TMS) δ 1.45 (3H, d, J = 6.8 Hz, CH_3), 4.88–4.94 (1H, m, CH), 5.88 (2H, s, CH_2), 7.25–7.37 (5H, m, C_6H_5), 8.03 (1H, d, J = 9.8 Hz, 8-quinolinium), 8.06 (1H, dd, J = 4.8, 9.8 Hz, 6-quinolinium), 8.23 (1H, dd, J = 5.8, 8.3 Hz, 3-quinolinium), 8.27 (1H, dd, J = 4.8, 9.8 Hz, 7-quinolinium), 8.49 (1H, d, J = 4.8 Hz, 5-quinolinium), 9.23 (1H, br, NH), 9.35 (1H, d, J = 8.3 Hz, 4-quinolinium), 9.48 (1H, d, J = 5.8 Hz, 2-quinolinium). UV-vis (MeCN, λ_{max}/nm (ε/dm^3 mol^{-1} cm^{-1})) 237 (3.68×10^5), 317 (8.21×10^4). ORD (c 0.25 in MeCN, 20 °C) $[\Phi]_{589}$ 91° for **3a**, −91° for **3b**. IR (KBr, ν/cm^{-1}) 847 (PF), 1532 (C=C), 1699 (C=O), 3422 (NH). Found: C, 52.39; H, 4.33; N, 6.41% (**3a**), C, 52.19; H, 4.23; N, 6.40% (**3b**). Calcd for $C_{19}H_{19}N_2OPF_6$: C, 52.30; H, 4.39; N, 6.42%.

Kinetic Measurements. A ZnMb solution was gently purged with Ar gas and then carefully degassed by freeze-pump-thaw cycles. Single-flash photolysis was performed in degassed solutions containing ZnMb (3.00×10^{-6} mol dm^{-3}) and chiral quinolinium ions (0–2.00×10^{-3} mol dm^{-3}) at 25.0 °C, pH 7.0 (0.010 mol dm^{-3} sodium phosphate buffer), and I = 0.020 mol dm^{-3} using a Photal RA-412 pulse flash apparatus with a 30 μs pulse-width Xe lamp ($\lambda > 450$ nm; a Toshiba Y-47 glass filter). Absorption spectral changes during the reaction were monitored at 460 nm for the decay of ^{3}(ZnMb)* and 680 nm for the formation and decay of $ZnMb^{\bullet+}$.

Other Measurements. ^{1}H NMR spectra were recorded on a JEOL JNM-GX270 FT NMR spectrometer (270 MHz). UV-vis spectra were measured with Shimadzu UV-240 and MultiSpec-1500 spectrophotometers. The fluorescence spectra and lifetimes were measured in Ar saturated solutions with a Shimadzu RF-5300PC spectrofluorometer and a Horiba NAES-500 nano-second fluorometer, respectively. The pHs of the solutions were measured on a Hitachi-Horiba F-14RS pH meter. Cyclic voltammetry was performed in an N_2-saturated 0.050 mol dm^{-3} tetrabuthylammonium perchlorate ($[Bu_4N]ClO_4$) MeCN solution and 0.050 mol dm^{-3} KCl aqueous solutions with a Yanaco Model P-900 instrument. A three-electrode system (BAS Inc.) was used with a Pt auxiliary electrode and a glassy carbon working electrode against Ag/$AgClO_4$ (0.10 mol dm^{-3} $[Bu_4N]ClO_4$ in MeCN) and Ag/AgCl (3.33 mol dm^{-3} KCl in water) reference electrodes. The potentials were calibrated by using 1,1′-dimethyl-4,4′-bipyridinium perchlorate ($[MV](ClO_4)_2$ (E^0 = −0.45 V vs SCE (saturated calomel electrode)) in MeCN and its chloride (E^0 = −0.45 V vs NHE (normal hydrogen electrode)) in water.

Results and Discussion

Characterizations of Chiral Quinolinium Compounds. New chiral quinolinium compounds (**2** and **3**) were characterized by elemental analyses and UV-vis, ^{1}H NMR, and IR spectroscopies. The redox and photophysical properties are summarized in Table 1 along with the previously reported compounds. Cyclic voltammograms of **2** and **3** showed irreversible reduction waves with relatively small oxidation peak currents. Therefore, the redox potentials were estimated from the mean values between the reduction and oxidation peak potentials: −0.88 V and −0.85 V vs SCE for **2** and **3**, respectively. The phenylethylcarbamoylmethyl group shifted the redox potential slightly positive compared with that of the *N*-alkyl-

Table 1. Redox and Photophysical Properties of Quinolinium Compounds in Acetonitrile and Aqueous Solutions

Quinolinium	$E^{a)}$/V		τ_f/ns		Ref.
PMQ^+ (**2**)	−0.88[b)]	−0.88[c)]	30[b)]	26[c)]	This work
PQ^+ (**3**)	−0.85[b)]	−0.85[c)]	20[b)]	16[c)]	This work
MMQ^+		−0.82[c)]		26[c)]	Ref. 11
MQ^+	−0.90[b)]	−0.96[b)]	20[b)]	15[c)]	Refs. 21 and 22
$PrMQ^+$	−0.89[b)]		25[b)]		This work

a) The potentials are against SCE and NHE in MeCN and aqueous solutions, respectively. b) In MeCN. c) In an aqueous solution.

quinolinium compounds ($[MMQ]^+$, $[MQ]^+$, and 1-(3-propyl)-6-methoxyquinolinium ($[PrMQ]^+$) ions). The fluorescence from the quinolinium moiety of **2** and **3** was observed at 455 nm and 440 nm, respectively. The fluorescence lifetime of **2** (30 ns) is longer than that of **3** (20 ns), which is in good agreement with the fact that the fluorescence intensity of **2** is larger than that of **3**. The fluorescence lifetimes in MeCN are longer than in water (26 ns and 16 ns for **2** and **3**, respectively). These results are the same tendency as those for the *N*-alkylquinolinium ions.

Photoinduced ET Reaction of ^{3}(ZnMb)* with Chiral Quinolinium Ions. The excited singlet state of ZnMb was not quenched by the quinolinium ions (**2** and **3**) under the present experimental conditions. In contrast, the excited triplet state, ^{3}(ZnMb)*, was quenched by **2** and **3**. Figure 1a shows the absorption spectral changes after the irradiation by light of a degassed solution containing ZnMb and **3** at 25 °C, pH 7.0 (0.010 mol dm^{-3} phosphate buffer), and I = 0.020 mol dm^{-3}. The decay of ^{3}(ZnMb)* in the presence of **2** or **3** was of first order, and was faster than the spontaneous decay of ^{3}(ZnMb)*. Plots of the first-order rate constant of the quenching of ^{3}(ZnMb)*, k_{obsd}, vs the concentrations of **2** and **3** were linear (see Fig. 2), indicating no appreciable complex formation between ^{3}(ZnMb)* and the quinolinium ions. When the reaction was monitored at 680 nm, we observed the formation and decay of the radical cation of ZnMb$^{\bullet+}$ (Fig. 1b),[26,27,29,30,38] indicating ET quenching followed by a thermal back ET reaction. Therefore, the photoinduced ET reaction of ZnMb with the quinolinium ions is represented in Scheme 1.

The quenching rate constants (k_q) obtained from the slope of the plots of k_{obsd} vs the concentrations of the quinolinium ions are listed in Table 2. The ET quenching reactions of ^{3}(ZnMb)* by **2** and **3** are slightly endothermic: ΔG^0 = 0.08 eV and 0.05 eV for **2** and **3**, respectively, based on the redox potentials of the couples, $[PMQ]^+/PMQ^\bullet$ (−0.88 V vs NHE), $[PQ]^+/PQ^\bullet$ (−0.85 V), and ZnMb$^{\bullet+}$/^{3}(ZnMb)* (−0.80 V).[40] Under the present experimental conditions, in which the quinolinium ions are used in large excess over ^{3}(ZnMb)*, the latter of which concentration is 1.8 × 10^{-6} mol dm^{-3}, the ET quenching was almost completed: > 93% for **2** and > 98% for **3**. The thermal back ET reaction was much slower than the quenching reaction of ^{3}(ZnMb)*, and the second-order rate constant of the back ET reaction (k_b) was evaluated at the latter portion of the decay of ZnMb$^{\bullet+}$ at 680 nm after the quenching reaction was completed (see Fig. 1b), based on

$$A_t = (A_0 + k_b[ZnMb^{\bullet+}]_0A_\infty t)/(1 + k_b[ZnMb^{\bullet+}]_0 t). \quad (1)$$

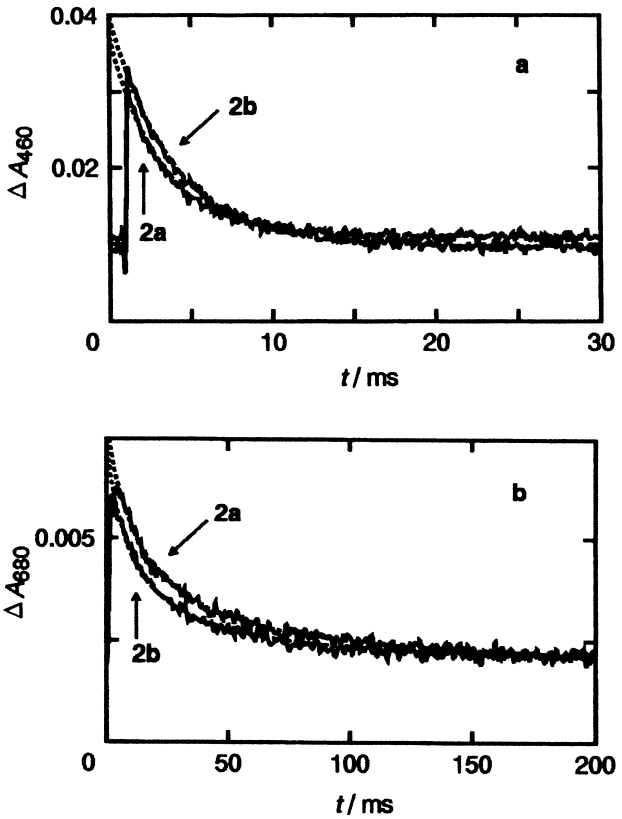


Fig. 1. Absorption spectral changes after irradiation by light of a degassed solution containing ZnMb (3.00 × 10^{-6} mol dm^{-3}) and **3** ($[PQ]^+$) at 25 °C, pH 7.0 (0.010 mol dm^{-3} phosphate buffer), and I = 0.020 mol dm^{-3}. (a) Decay of ^{3}(ZnMb)* at 460 nm in the presence of 1.00 × 10^{-3} mol dm^{-3} **3a** ((*S*)-$[PQ]^+$) and **3b** ((*R*)-$[PQ]^+$). Dotted lines are fitted to the first-order kinetics. (b) Decay of ZnMb$^{\bullet+}$ at 680 nm in the presence of 2.00 × 10^{-3} mol dm^{-3} **3a** ((*S*)-$[PQ]^+$) and **3b** ((*R*)-$[PQ]^+$). Dotted lines are fitted to the second-order kinetics.

Here, A_0, A_t, and A_∞ are the absorbances at time 0, t, and infinity, respectively, and $[ZnMb^{\bullet+}]_0$ is the initial concentration of ZnMb$^{\bullet+}$. The value of k_b was determined by using the value of $[ZnMb^{\bullet+}]_0$ which was estimated from the concentration of ^{3}(ZnMb)* ($\Delta\varepsilon_{428}$ = ε(ground) − ε(triplet) = 1.00 × 10^5 dm^3 mol^{-1} cm^{-1}).[29,41] The values of k_b are listed in Table 2.

Mechanisms of ET Quenching and Back ET Reactions. It has been suggested that a conformational gating controls the intermolecular ET reactions of ZnMb, zinc hemoglobin, and

Table 2. Rate Constants of the Quenching and Back ET Reactions of ZnMb with Chiral Quinolinium and Viologen at 25 °C, pH 7.0, and $I = 0.020$ mol dm^{-3}

	$k_q/10^5$ dm^3 mol^{-1} s^{-1}			k_b[a]$/10^8$ dm^3 mol^{-1} s^{-1}		
Quencher	(*S*)-isomer	(*R*)-isomer	$k_q(S)/k_q(R)$	(*S*)-isomer	(*R*)-isomer	$k_b(S)/k_b(R)$
PMQ^+ (**2**)	4.5 ± 0.3	3.4 ± 0.2	1.3	1.0 ± 0.1	1.3 ± 0.1	0.77
PQ^+ (**3**)	2.1 ± 0.2	1.5 ± 0.1	1.4	0.38 ± 0.03	0.53 ± 0.04	0.72
OAV^{2+} [b]	390 ± 10	290 ± 10	1.3	1.4 ± 0.1	0.98 ± 0.08	1.4

a) Determined at four different concentrations of **2** and **3** ((1.25–2.00) × 10^{-3} mol dm^{-3}).
b) Ref. 8.

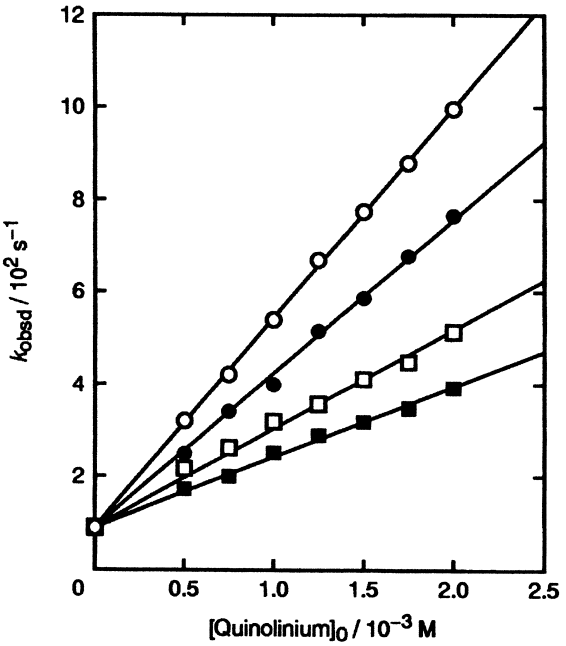
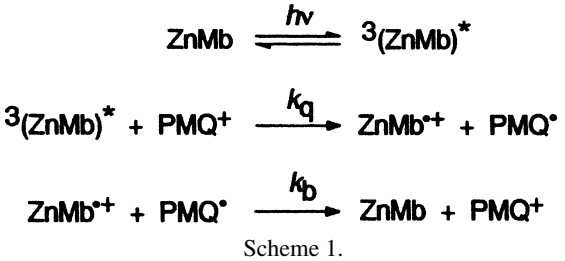


Fig. 2. Plots of k_{obsd} vs [Quinolinium]$_0$ for the quenching of ^{3}(ZnMb)* by the chiral quinolinium ions at 25 °C, pH 7.0, and $I = 0.020$ mol dm^{-3}. (○, ●) **2** ([PMQ]$^+$) and (□, ■) **3** ([PQ]$^+$). Open and closed symbols are for (*S*)- and (*R*)-isomers, respectively.



Scheme 1.

the other metalloproteins;[25–35] the ET rate is insensitive to the redox potentials of quenchers. In the case of the quinolinium ions (**2** and **3**), the rate constants (k_q) are much smaller than those for the other quenchers, such as viologens (see Table 2). This may arise partly because the driving force of the reaction is slightly positive; namely the ET rate is dependent on the driving force of the reaction, and is not fast enough to be controlled by a conformational change of myoglobin. To confirm this assumption we calculated the rate constants for the quenching of ^{3}(ZnMb)* by **2** and **3**, and the back ET reaction between ZnMb$^{\bullet+}$ and the quinoline radicals by using the Marcus theory (Eq. 2);[42,43] using

$$k_{12} = (k_{11}k_{22}f_{12}K_{12})^{1/2}. \quad (2)$$

Here, k_{12} is the rate constant for the cross reaction, k_{11} and k_{22} are those for the self-exchange reactions of donor and acceptor, respectively, K_{12} is the equilibrium constant for the cross reaction, and f_{12} is given by

$$\ln f_{12} = (\ln K_{12})^2/4\ln(k_{11}k_{22}/10^{22}). \quad (3)$$

Equations 2 and 3 are also represented by

$$\ln k_{12} = 25.3 - (\lambda_{12} + \Delta G^0)^2/4\lambda_{12}RT, \quad (4)$$

where λ_{12} is the reorganization energy for the reaction, and equals the average of those of the donor and acceptor, $(\lambda_{11} + \lambda_{22})/2$. By using the data $k_{11} = 2.6 \times 10^5$ dm^3 mol^{-1} s^{-1} for ZnMb$^{\bullet+}$/^{3}(ZnMb)* ($\lambda_{11} = 1.32$ eV),[44] $k_{22} = 1.0 \times 10^8$ dm^3 mol^{-1} s^{-1} for both PMQ^+/ $PMQ^\bullet$ and PQ^+/ $PQ^\bullet$ couples ($\lambda_{22} = 0.71$ eV),[45] and $K_{12} = 4.4 \times 10^{-2}$ for PMQ^+ and 1.4×10^{-1} for PQ^+ (the redox potential of ZnMb$^{\bullet+}$/ZnMb is 0.98 V),[40] the calculated rate constants, k_{12}, are 1.0×10^6 dm^3 mol^{-1} s^{-1} ($f_{12} = 0.88$) and 1.9×10^6 dm^3 mol^{-1} s^{-1} ($f_{12} = 0.95$) for PMQ^+ and PQ^+, respectively. The calculated rate constants are close to the observed ones (see Table 2 and Fig. 3 (symbols ○, ●, □, and ■)). Similarly we obtained the calculated rate constants for the back ET reactions, $k_{12} = 1.0 \times 10^8$ dm^3 mol^{-1} s^{-1} ($f_{12} = 1.2 \times 10^{-29}$) and 1.7×10^8 dm^3 mol^{-1} s^{-1} ($f_{12} = 1.1 \times 10^{-28}$) for $PMQ^\bullet$ and $PQ^\bullet$, respectively, by using the data $k_{11} = 2.6 \times 10^5$ dm^3 mol^{-1} s^{-1} for ZnMb$^{\bullet+}$/ZnMb ($\lambda_{11} = 1.32$ eV) and $K_{12} = 3.2 \times 10^{31}$ for $PMQ^\bullet$ and 9.9×10^{30} for $PQ^\bullet$. These calculated rate constants for the back ET reaction are also close to those for the observed ones (see Table 2 and Fig. 3).

On the other hand, the viologen system is different from that for the quinolinium ions. The quenching rate constant for a 1,1′-bis(1-phenylethylcarbamoylmethyl)-4,4′-bipyridinium ion (OAV^{2+}) is calculated to be $k_{12} = 1.9 \times 10^9$ dm^3 mol^{-1} s^{-1} ($f_{12} = 9.6 \times 10^{-4}$), by using the data $k_{22} = 1.0 \times 10^8$ dm^3 mol^{-1} s^{-1} for OAV^{2+}/$OAV^{\bullet+}$ ($\lambda_{22} = 0.71$ eV)[46–49] and $K_{12} = 1.5 \times 10^{10}$ (the redox potential of OAV^{2+} is −0.20 V).[9] The calculated value is much larger than the experimentally determined one ((2.9–3.9) × 10^7 dm^3 mol^{-1} s^{-1}),[8] as is shown in Fig. 3 (symbols △ and ▲). Moreover, the calculated rate constant for the back ET reaction ($k_{12} = 7.6 \times 10^{10}$ dm^3 mol^{-1} s^{-1}) is also much larger than the observed one ($k_b = (0.98–1.4) \times 10^8$ dm^3

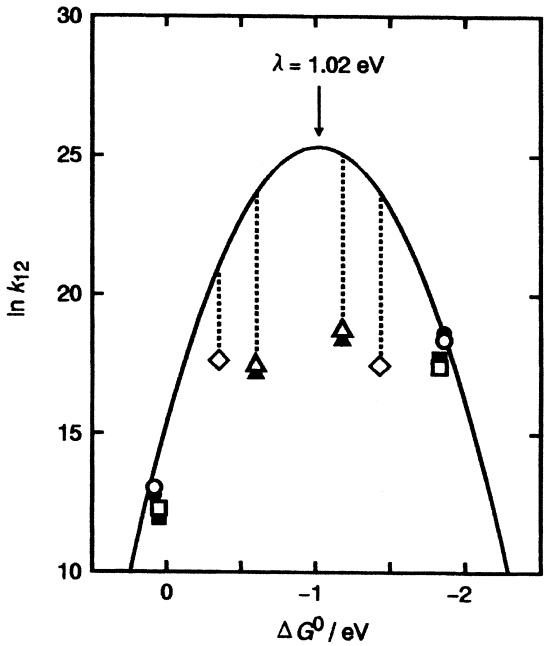


Fig. 3. Driving force dependence of the rate constants for the quenching and back ET reactions of ZnMb with quinolinium ions and viologens. (○) **2a**, (●) **2b**, (□) **3a**, (■) **3b**, (△) (*S*,*S*)-OAV^{2+}, (▲) (*R*,*R*)-OAV^{2+}, and (◇) MV^{2+}. The solid curve is calculated by Eq. 4 with $\lambda_{12} = 1.02$ eV.

$mol^{-1}\ s^{-1}$) with $K_{12} = 9.7 \times 10^{19}$ and $f_{12} = 2.3 \times 10^{-12}$. For a typical viologen, MV^{2+}, the rate constants for the quenching and back ET reactions were also calculated, and are much different from the observed ones (see the symbol ◇ in Fig. 3). From these results we can conclude that the present quinolinium system is controlled by an ET step and that the viologen system is not controlled by an ET, as has been previously reported.[7,8,26–31]

Stereoselectivity in ET Quenching and Back ET Reactions. The k_q values for the (*S*)-isomers of the chiral quinolinium ions are larger than those for the (*R*)-isomers. The ratios of $k_q(S)/k_q(R)$ are 1.3 and 1.4 for **2** and **3**, respectively. Therefore, the (*S*)-isomers preferentially quench $^3(ZnMb)^*$, which is the same tendency as in the viologen system.[8] A similar selectivity of quinolinium to viologen may arise from a decrease in the charge repulsion and the number of chiral groups for the former. The latter has a more positive charge and two chiral moieties. We have suggested from the evidence for the precursor complex formation in the thermal ET reaction of myoglobin with chiral viologen radical cations that the amide bond in the chiral viologen reacts with an amino or carboxylate group of surface amino-acid residues (Lys, Asp, and/or Glu).[10] In the present case, the attacking site by the quinolinium ion might be Lys, Asp, and/or Glu residues near the heme pocket of myoglobin.

On the other hand, interestingly, the selectivity in the back ET reaction was inverted: the ratios of $k_b(S)/k_b(R)$ are 0.77 and 0.72 for **2** and **3**, respectively. The quinoline radical produced by the ET quenching reaction becomes a neutral molecule; therefore, the charge repulsion decreases. Then, the quinoline radical easily attacks myoglobin, especially the hydrophobic heme pocket, which is different from the site for the quenching reaction.

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