Stabilising Effects on the 4Fe–4S Core Analogues for High-potential Iron–Sulphur Proteins with a Hydrophobic Environment Provided by Macrocycles

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Studies of redox potentials and reactions with molecular oxygen of a series of 4Fe–4S complexes attached to a 36-membered macrocycle show that all three electron transfer processes, 1−/2−, 2−/3−, and 3−/4−, are reversible for the cyclic aryl-substituted clusters, and that half-wave potentials for alkyl derivatives with hydrophobic macrocycles showed largely positive shifts of the 1−/2− couples as given in high-potential iron–sulphur proteins as well as a stabilising effect of the cores towards molecular oxygen.

The Fe_4S_4 cubane-type clusters are of great interest because they are good analogues for the active site in ferredoxins and other iron–sulphur proteins. Studies on redox behaviour and stability of synthetic Fe_4S_4 cores surrounded by a hydrophobic environment are of relevance to the biological systems.[1] We have studied the characteristics of the new clusters (**1**)—(**3**), in particular their electrochemical behaviour and dissolution reactions with molecular oxygen. We now report that the use of SH ligands attached to the hydrophobic 36-membered macrocycle causes a positive shift of redox potentials for the 2−/1− couple and leads to increased stability of the 4Fe–4S core towards oxygen.

The model complexes (**1**)—(**3**) were prepared, as black powders,† using novel tetrathiol compounds anchored to a 36-membered ring consisting of a methylene framework as previously reported.[2]

Cyclic voltammetric electrochemical studies demonstrated that the clusters (**1**) and (**2**) can be oxidized and reduced by one electron in dimethyl sulphoxide (DMSO).‡ While Pickett

† Complex (**1**): n.m.r. (270 MHz), δ (CD_3SOCD_3), 1.07—1.49 [br., 24H + 48H, CH_3(cation) + CH_2 (skeleton)], 3.16—3.37 [br., 16H, N_α–(skeleton)–CH_2], 3.18—3.20 [br., 16H, N_α(cation)–CH_2], 5.8 (br., 8H, ArH *ortho* to S), and 8.0 (br., 8H, ArH *meta* to S); $\lambda_{max.}$ (DMF) (ε) 442 (14 300) and 365 (sh, 21 100) nm. Satisfactory elemental analyses (Et_4N salt) were obtained.

Complex (**2**): n.m.r. (270 MHz), δ (CD_3SOCD_3), 1.10—1.49 [br., 24H + 48H, CH_3(cation) + CH_2 (skeleton)], 2.8 (br., 16H, N_α (skeleton)–CH_2], 3.17—3.19 [br., 16H, N_α(cation)–CH_2], 7.3 and 7.5 (br. × 2, 16H, ArH), and 13.2 (br., 8H, Ph–CH_2–S); $\lambda_{max.}$ (DMF) (ε) 420 (15 300) and 315 (sh, 22 300) nm.

Complex (**3**): n.m.r. (270 MHz), δ (CD_3SOCD_3), 1.02—1.77 [br., 24H + 48H, CH_3(cation) + CH_2(skeleton)], 2.50 (s, 8H, CO–CH_2), 2.59 [br., 24H, CH_3(ligand)], and 3.23—3.30 [br., 16H, N_α(cation)–CH_2]; $\lambda_{max.}$ (ε) 416 (16 200) and 302 (sh, 21 100) nm.

Table 1. Redox potentials of 4Fe–4S clusters in DMSO solution.[a]

Compound[b]	$E_{1/2}$/V 1−/2−	2−/3−	3−/4−
		$(E_{p,c} - E_{p,a})$/mV	
(**1**)	−0.36	−0.85	−1.64
	(36)	(48)	(50)
(**4**)	—	−0.92	−1.70
	—	(74)	(134)
(**2**)	−0.35	−1.12	−1.86
	(70)	(20)	(30)
(**5**)	—	−1.14	—
	—	(70)	—
(**3**)	+0.25	−1.13	—
	(180)	(60)	—
(**6**)	−0.11	−1.40	—
	(46)	(70)	—

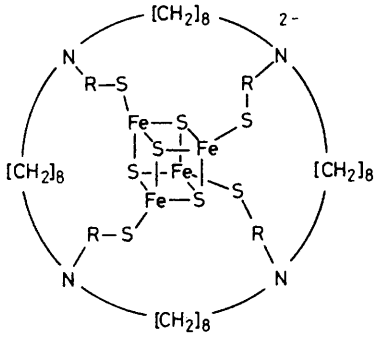
[a] Potentials *vs.* S.C.E. See ‡ footnote for more details. [b] Et_4N^+ salt.

‡ Electrochemical data were obtained for a cluster concentration of 1 mM in 0.2 M solutions of $Bu^n_4NBF_4$ using platinum as the working electrode and a saturated calomel electrode (S.C.E.) as the reference.

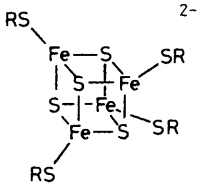
Table 2. Rate constants and lifetimes ($1/k_{obs.}$) for cluster decomposition with O_2.

Compound[a]	O_2/[cluster] = 10 $k_{obs.} \times 10^3/min^{-1}$	$(1/k_{obs.})$/min	O_2/[cluster] = 2 $k_{obs.} \times 10^3/min^{-1}$	$(1/k_{obs.})$/min
(1)	16.6	60.3	4.0	251
(4)	26.9	37.2	6.2	162
(2)	21.9	45.7	5.0	200
(5)	126	7.9	11.7	85.1
(3)	8.1	123	2.6	389
(6)	30.1	33.1	4.7	214

[a] Et_4N^+ salt, 0.5 mM in DMF.



(1) R = $-C(:O)C_6H_4-p-$
(2) R = $-C(:O)C_6H_4CH_2-p-$
(3) R = $-C(:O)CH_2CMe_2-$



(4) R = Ph
(5) R = CH_2Ph
(6) R = Bu^t

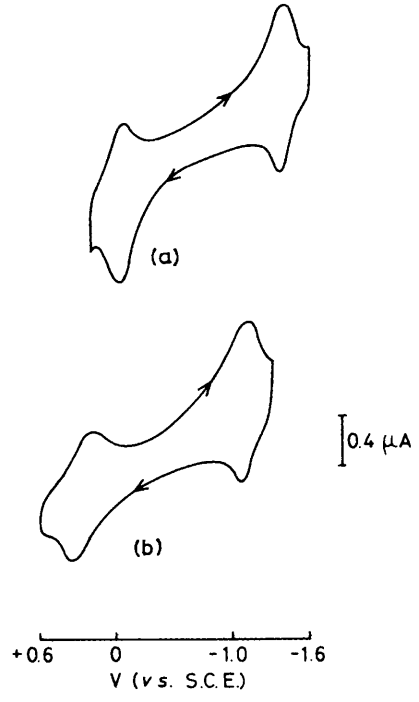


Figure 1. Cyclic voltammograms of 4Fe–4S clusters (a), (3), and (b), (6) (1 mM) in Me_2SO, scan rate 100 mV s^{-1}.

reported that a particular medium was required for measuring the complete electron-transfer series of the cluster $[Fe_4S_4(SPh)_4]^{2-}$ (4),[3] all three redox processes, 1−/2−, 2−/3−, and 3−/4−, with notably good reversibilities, were readily obtained using the complex (1) in DMSO solution under ordinary conditions. This phenomenon seems to be reasonably general, since similar behaviour was observed for the benzyl analogues (2) and $[Fe_4S_4(SCH_2Ph)_4]^{2-}$ (5). It should be emphasized that the half-potentials of (1) and (2) tend to shift in a positive direction (*e.g.*, *ca.* 70 and 60 mV, respectively, for the 2−/3− and 3−/4− cycles for the phenyl derivatives) compared with the corresponding complexes (4) and (5) containing small thiol ligands. Since $[Fe_4S_4(SBu^t)_4]^{2-}$ (6) is less hydrophobic than (4), the 2−/3− redox potential is more negative than that of (4). However, (6) shows a potential due to the 1−/2− process, which is considered to be attributable to its bulky ligand. The cyclic cluster (3) which is both hydrophobic and bulky showed the most marked positive shift of the redox potentials for the 2−/3− and 2−/1− processes; *i.e.* positive shifts of 0.27 V for the former and 0.38 V for the latter compared to (6) (Figure 1). Data are collected in Table 1. The above results suggest that the increased hydrophobic environment has a marked influence, and neither use of sterically highly hindered ligands such as 2,4,6-trimethyl- or 2,4-6-tri-isopropyl-benzenethiolato[4] nor the presence of NH · · · · S hydrogen bonding such as in $[Fe_4S_4(Z\text{-}Cys\text{-}Gly\text{-}Ala\text{-}OMe)_4]^{2-}$ (Z = benzyloxycarbonyl)[5] is an absolutely necessary factor in the stabilization of analogues of the biological high potential ferredoxin, HP_{ox}.[6] Steric protection from solvent attack afforded by the hydrophobic environment is at least as important a contributing factor.§

In further studies, in as much as the active sites of the iron–sulphur proteins are known not to be particularly stable towards oxygen,[7] we have examined the decomposition of the synthetic complexes (0.5 mM clusters in dimethylformamide, DMF) with molecular oxygen by observing the decrease in absorbance at $\lambda_{max.}$¶ Plots of the logarithms of the first-order rate constant ($k_{obs.}$) for the decomposition as a function of O_2 concentration showed the marked stabilizing effect of the Fe–S cores in (1), (2), and (3) compared to the corresponding unclad clusters. For example, with a 10 molar excess of oxygen

§ Preliminary results for a more hydrophobic ligand ($-COCH_2-CMe_2SH$) in a cyclophane-type macrocycle gave $E_{1/2}$ +0.25 V with a much smaller difference between $E_{p,a}$ and $E_{p,c}$ values (80 mV) for the 2−/1− couple.

¶ Reaction rates of the O_2 oxidation were obtained in the form of initial rates (slope = $\Delta A/\Delta t$). $k_{obs.}$ values were computed from the initial rate of change of absorbance with time, $k_{obs.}/min^{-1}$ = (initial slope/A_1), A_1 = initial absorbance.

concentration [O_2/(Fe–S) = 10], the stability of complexes (**4**), (**6**), and (**5**) decreased in this order (relative ratios 4.7 : 4.2 : 1), and the clusters contained within the macrocycle, (**1**), (**2**), and (**3**), were 1.4, 5.8, and 3.7 times more stable than the corresponding non-macrocyclic clusters (**4**), (**5**) and (**6**), respectively. Five-fold changes in oxygen concentration were found to provide only 3.1—4.1-fold changes in $k_{obs.}$ for the disappearance of the macrocyclic clusters, but 4.3—10.8-fold changes for the unclad clusters. The data are summarized in Table 2 together with the lifetimes obtained as the reciprocal of the rate constants.

The above observations are pertinent to the situation in biological systems, where the uncommon stability of the $[Fe_4S_4(SCys)_4]^-$ core of the HP proteins has been attributed to the hydrophobic spheres of the iron–sulphur centres.[1,6,8]

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