Electrochemical Study on Dihydrolipoamide-Iron(II) Complex and Its Chemical Reactivity

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Electrochemical properties of dihydrolipoamide (DHLAm)-Fe(II) complex are investigated by use of cyclic voltammetry on a gold electrode in buffer–ethanol solution. A reversible peak based on the DHLAm-Fe(II) complex is observed at –0.65 V by mixing DHLAm and ferrous ion. The redox potentials suggest that the DHLAm-Fe(II) complex is more easily oxidized than DHLAm or ferrous ion. While hydroxylamine derivatives, isoxazoles, and nitrobenzene are reduced by DHLAm-Fe(II), as reported already, reduction potentials of these compounds are more negative than that of the DHLAm-Fe(II) complex. It is suggested that these compounds are reduced through coordination to the DHLAm-Fe(II) complex.

Lipoamide (LAm) works as a coenzyme in acyl-transfer and redox reactions: LAm ⇌ dihydrolipoamide (DHLAm).

We have recently reported[1] that hydroxylamines are cleaved reductively to corresponding amines and alcohols by DHLAm in the presence of ferrous ion under slightly alkaline conditions, as shown in eq 1. It has also been

$$Fe^{2+} + RNH_2 + HOR' \quad (1)$$
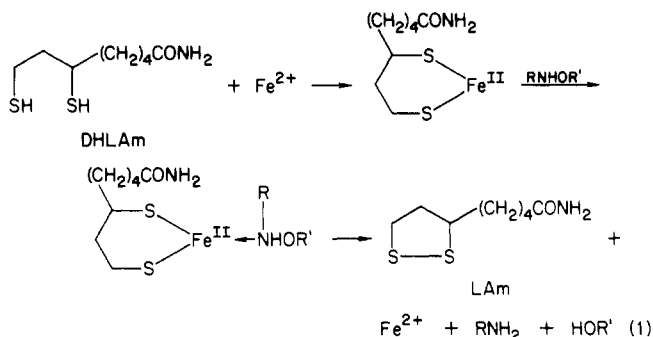
reported[2,3] that azo- and nitrobenzenes are reduced to hydrazobenzene and anilines, respectively, by DHLAm-Fe(II) under mild conditions. In every case, these compounds are not reduced either by DHLAm or ferrous ion alone. It was proposed that in the reduction of these compounds by the DHLAm-Fe(II) a DHLAm-Fe(II) complex is the active species. These specific reactivities of DHLAm-Fe(II) are interesting in connection with non-heme iron proteins such as rubredoxins and ferredoxins in living systems.

In this paper, electrochemistry is used to investigate the reactivity of the DHLAm-Fe(II) complex.

It has been reported[4] that electrochemical oxidations of thiols at a dropping mercury electrode do not give the corresponding disulfides but form mercury thiolates instead. On the other hand, an electrochemical study of lipoic acid and related compounds was achieved by use of a gold electrode in aprotic and aqueous media.[5] For these reasons, electrochemical measurements are conducted by use of a gold electrode in the present studies.

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Table I. Cyclic Voltammetric Data of Some Compounds[a]

compounds	E_p^a, V	E_p^c, V	$E_{1/2}$, V
FeSO$_4$(NH$_4$)$_2$SO$_4$	–0.44	–0.52	–0.48
DHLAm	0	–0.10	
DHLAm-Fe(II)	–0.62	–0.69	–0.65
PhN=NPh	–0.52	–0.53	–0.52
NH$_2$OCH$_2$Ph		–0.80	
PhNO$_2$		–0.88	
PhNHOH		<–1.20[b]	
3,5-dimethyl isoxazole		<–1.20[b]	

[a] Conditions: 1 mM solution (substrate) in 0.4 M KCl plus 0.1 M carbonate buffer (pH 9.8)-EtOH (2:1) on a gold electrode, scan rate 0.3 V/s, potentials are referrenced to SCE. [b] The reduction peak was not observed in the range of +0.20 to –1.20 V.

Results and Discussion

The cyclic voltammograms of dihydrolipoamide (DHLAm), ferrous ion (FeSO$_4$(NH$_4$)$_2$SO$_4$), and the DHLAm-Fe(II) system in carbonate buffer (pH 9.8)–EtOH (2:1), which was used in the reduction of hydroxylamine derivatives, are shown in Figure 1.

In case of DHLAm (Figure 1, a) an oxidation peak was observed around 0 V, which corresponds to the oxidation of DHLAm to LAm, referring to the similar result.[5] In case of ferrous ion (Figure 1, b), a reversible wave, which corresponds to the redox process between ferrous ion and ferric ion, is observed at –0.48 V. By mixing DHLAm with 0.02 equiv of ferrous ion (Figure 1, c), the reversible wave at –0.48 V disappears and a new peak, thought to be based on a redox reaction of the DHLAm-Fe(II) complex, appears at –0.65 V. The DHLAm-Fe(III) system, made by addition of Fe$_2$(SO$_4$)$_3$·(NH$_4$)$_2$SO$_4$ to the DHLAm solution, also shows the same peak as the DHLAm-Fe(II) system.

These results suggest that a new species which has a higher reducing ability than DHLAm or ferrous ion is formed by mixing DHLAm with ferrous ion in a slightly alkaline solution.

The following relations hold in the voltammogram of the DHLAm-Fe(II) system (Figure 2); $\Delta E_p = E_p^a - E_p^c = 70$ mV and $i_p \propto V^{1/2}$ (V = sweep rate). These results suggest that electron transfer between the DHLAm-Fe(II) and DHLAm-Fe(III) complexes is reversible.[6]

The solution of the DHLAm-Fe(II) system showed a new absorption spectrum having shoulders at 315, 340, and 415 nm which might be caused by the formation of a DHLAm-Fe(II) complex. The composition of the DHLAm-Fe(II) complex was determined to be 1:1 by the con-

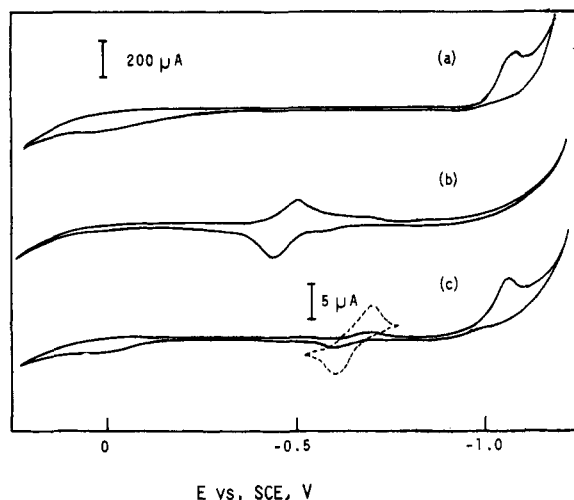(6) Matsuda, H. *Z. Elektrochem.* 1957, *61*, 489.

Figure 1. Cyclic voltammograms of DHLAm, ferrous ion, and DHLAm–Fe(II) solution in 0.4 M KCl + 0.1 M sodium carbonate buffer (pH 9.8)–EtOH (2:1) on a gold electrode, scan rate 0.3 V/s: (a) [DHLAm] = 50 mM; (b) [Fe^{2+}] = 10 mM; (c), [DHLAm] = 50 mM, [Fe^{2+}] = 1 mM. The dotted line was obtained at a higher sensitivity (×40).
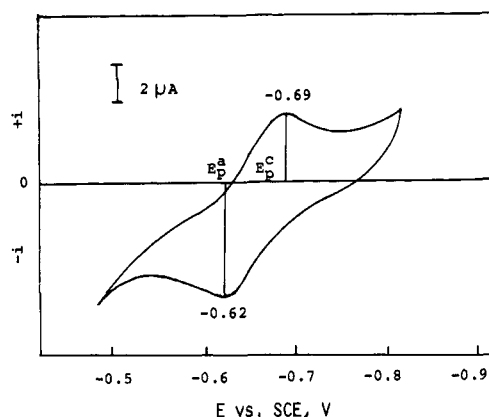

Figure 2. Cyclic voltammogram of DHLAm–Fe(II) complex in buffer (pH 9.8)–EtOH (2:1) solution on a gold electrode with a scan rate of 50 mV/s.

tinuous variation method by plotting the absorbance at 315 nm (Figure 3). Thus, this reversible wave at –0.65 V is thought to be based on a redox reaction between the DHLAm–Fe(II) (1:1) complex and the DHLAm–Fe(III) complex.

Electrochemical data for the complex and substrates reduced by this complex are listed in Table I.

The reduction peak potentials (E_p^c) of hydroxylamine derivatives, nitrobenzene, and isoxazole are more negative than that of DHLAm–Fe(II). From these results, these substrates might not be reduced by DHLAm–Fe(II). However, these compounds are chemically reduced by DHLAm–Fe(II) to amines and alcohols,[1] anilines,[2] and β-amino enone,[7] respectively. These N–O compounds are, therefore, thought to be activated by coordination to the DHLAm–Fe(II) complex and reduced.

Further, the redox potentials of various thiol–Fe(II) complexes and oxidation potentials of thiolates are shown in Table II together with the yields[1] of benzyl alcohol in the reduction of *O*-benzylhydroxylamine by various thiol–Fe(II) systems.

The oxidation potentials of thiolates correspond to a one-electron oxidation from S anion to S radical. The
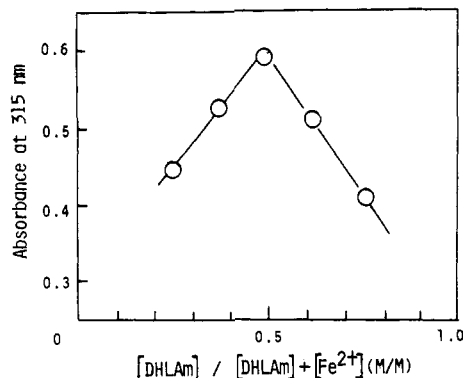

Figure 3. Continuous variation curve of DHLAm and ferrous ion: [DHLAm] + [Fe^{2+}] = 5 × 10^{-8} M, in buffer (pH 9.8)–EtOH (1:1) under Ar.

Table II. Electrochemical Data and Chemical Reactivities of Thiol–Fe(II) Complexes[a]

thiol	E_p^a, V (thiolate)	$E_{1/2}$, V (thiol–Fe(II))	yield of PhCH$_2$OH,[b] %
(CH$_2$)$_4$CONH$_2$, SH SH	–0.10	–0.65	100
(CH$_2$)$_4$COOH, SH SH	+0.10	–0.35	93
CH$_2$SH / CH$_2$SH (benzene)	–0.05	–0.54	53
HO OH SH SH	–0.10	–0.61	58
SH SH OH	–0.20	–0.76	0
SH (benzene)	+0.23	–0.36	5

[a] Conditions are the same as those in Table I. [b] Yields were determined by GLC. Reaction conditions: [NH$_2$OCH$_2$Ph] = 10 mM, [dithiol] = 20 mM ([monothiol] = 40 mM), [Fe^{2+}] = 0.05 mM in 0.1 M carbonate buffer–EtOH (1:1) for 4 h at room temperature.[1]

redox potentials of thiol–Fe(II) complexes correspond to the redox reaction between thiol–Fe(II) complexes and thiol–Fe(III) complexes, and the redox potentials correlate well with the oxidation potentials of thiolates. However, chemical reactivities do not correlate with these potentials but correlate with the ease of formation of cyclic disulfides in the oxidative cyclization of Bunte salts (eq 2).[8]

$$\text{(5-ring)} \quad > \quad \text{(6-ring)} \quad \gg \quad \text{(4-ring)} \qquad (2)$$
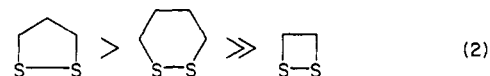
As a thiol–Fe(II) complex has an increasingly negative redox potential, the *O*-benzylhydroxylamine which coordinates to the thiol–Fe(II) complex should be subjected to one-electron reduction more easily. However, the 2,3-dimercaptopropanol–Fe(II) complex which has the most negative redox potential in Table II does not reduce the hyroxylamine derivative, because 2,3-dimercaptopropanol cannot be oxidized to a cyclic disulfide and two electron transfer from the complex to the substrate would be impossible. DHLA–Fe(II) and thiophenol–Fe(II) complexes have similar redox potentials. DHLA–Fe(II) reduces the hydroxylamine, but thiophenol–Fe(II) scarcely reduces this substrate, because thiophenol is more difficultly oxidizable to a disulfide than is DHLA.

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For these reasons, it is suggested that the reduction of O-benzylhydroxylamine by DHLAm–Fe(II) proceeds as shown in eq 3. The hydroxylamine coordinates to the
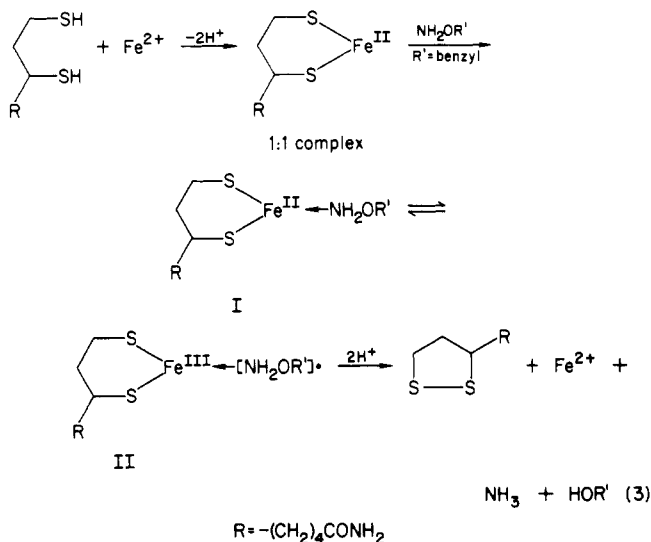
thiol–Fe(II) complex to produce intermediates I. The coordinated hydroxylamine then undergoes one-electron reduction to produce intermediate II, which is presumed to be in equilibrium with intermediate I. In the final stage, one-electron transfer from thiol–Fe(III) complex to the hydroxylamino radical occurs by oxidative cyclization of dithiol to disulfide to produce ammonia, the alcohol, ferrous ion, and disulfide.

In conclusion, it is confirmed by spectroscopic and electrochemical methods that a reactive DHLAm–Fe(II) (1:1) complex is formed by mixing DHLAm and ferrous

ion. Hydroxylamine derivatives, nitrobenzene, and isoxazole coordinate to the active complex and are reduced, respectively, with oxidative cyclization of DHLAm to LAm.

Experimental Section

Materials and Measurements. The cyclic voltammetric experiments were accomplished by use of a Hokuto Denko Potentiostat PS-1000 and a Function Generator HB-104. A gold-inlay electrode was used as the working electrode in 0.4 M KCl + 0.1 M carbonate buffer (pH 9.8)–ethanol (2:1) solution. The solution in the electrochemical cell was degassed with argon.

Absorption spectra were recorded on a Hitachi 200-10 spectrometer.

Dihydrolipoamide (DHLAm) was prepared by the reduction of lipoamide with sodium borohydride according to the procedure of Reed et al.[9] o-Xylene-α,α'-dithiol was prepared as reported already.[10]

Other chemicals used in this study were reagent grade and solvents were purified by the usual methods.

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Synthesis and X-ray Crystal Structure Analysis of a Vicinally Dinitro-Substituted Bishomopentaprismane

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Oximation of [4]peristylane-2,6-dione (6) followed by bromination in buffered aqueous dimethylformamide and trifluoroperacetic acid oxidation delivered chiefly the caged dinitro dibromide 9. Upon treatment with tri-*n*-butyltin hydride in refluxing benzene, 9 was transformed into the title compound (4). No evidence for free radical chain elimination of the nitro groups was seen. Single-crystal X-ray analysis was performed on 4. Highly strained carbon–carbon single bonds are in evidence as expected. Significantly, the pair of nitro groups bring to 4 a density (1.63 g cm^{-3}) markedly enhanced relative to values exhibited by closely related structures.

The search for energetic compounds having high density has very recently focused on polynitro-substituted cage molecules.[1–3] Properties such as heightened strain energy, molecular compactness, and self-contained oxidizability have been deemed especially important. To this time,

1,4-dinitrocubane (1),[1,2] 3,5,5-trinitropentacyclo-
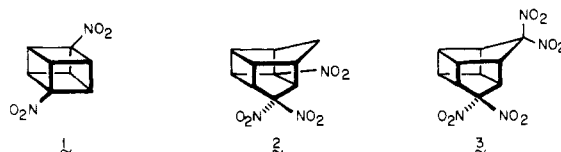
[5.3.0.0^{2,6}.0^{3,10}.0^{4,8}]decane (2),[3a] and 5,5,9,9-tetranitropentacyclo[5.3.0.0^{2,6}.0^{3,10}.0^{4,8}]decane (3)[3b] represent the most advanced candidates to have yielded to synthesis.

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(2) Griffin, G. W., private communication.

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