Cytochrome c_3, a Tetrahemoprotein Electron Carrier Found in Sulfate-Reducing Bacteria

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This Account reviews the unique characteristics of the tetrahemoprotein cytochrome c_3 obtained from the sulfate-reducing bacteria *Desulfovibrio vulgaris*. This cytochrome, originally reported by Postgate in England[1] and Ishimoto in Japan[2] in 1954, was the first cytochrome isolated from strictly anaerobic bacteria. Before its discovery, the cytochrome family had been thought to be confined to aerobic creatures including facultative anaerobes.[3,4] Discovery of a cytochrome in strictly anaerobic sulfate-reducing bacteria extended the role of cytochromes as more general electron carriers working in various kinds of electron-transfer chains.

In *Desulfovibrio*, cytochrome c_3 is situated at the entrance of an electron-transfer chain starting from H_2 and terminating at sulfur oxy acids, sulfite, or adenylyl sulfate (an active form of sulfate). The content of cytochrome c_3 in *D. vulgaris* ranges from 10 to 20 nmol/g of wet packed cells.[5] As the cell is a rod, 0.4 μm in diameter and 2~5 μm long,[6] the number of cytochrome c_3 molecules in a single cell is between 2000 and 7000. Cytochrome c_3 is one of the essential constituents of sulfate-reducing bacteria belonging to genus *Desulfovibrio*.[7,8] *Desulfotomaculum*, another genus of sulfate-reducing bacteria, is characterized by the presence of a *b*-type cytochrome instead of cytochrome c_3.[7,8] So far, tetrahemoprotein cytochrome c_3, which has a very negative redox potential, has been reported only in *Desulfovibrio*.[9-14] Trihemo-protein cytochrome c_7,[10] which also has a very negative redox potential and is genetically correlated to cytochrome c_3,[15] has been reported in *Desulfuromonas*, which reduces sulfur instead of sulfate.[16] While cytochrome c_7 may well turn out to be a member of the cytochrome c_3 family, only the tetrahemoprotein cytochrome c_3 will be dealt with in this Account.

Function of Cytochrome c_3

Cytochrome c_3 is an electron carrier or mediator in the reactions of very negative redox potentials operating in cells of *Desulfovibrio*. When the cell-free extract of this genus is passed through a column of Amberlite CG 50 (NH_4^+) to adsorb cytochrome c_3, the extract loses its ability to absorb H_2 in the presence of thiosulfate,[17,18] sulfite,[17,19] hydroxylamine,[18] colloidal sulfur,[18,20] and adenylylsulfate.[21] Similarly evolution of H_2 from formate and $Na_2S_2O_4$ is also dependent on the presence of cytochrome c_3. These experiments suggested that cytochrome c_3 might be a natural electron carrier of *Desulfovibrio* hydrogenase. Direct proof that this was true came from experiments with partially purified hydrogenase[22,23] or highly purified hydrogenase[24] of *D. vulgaris* Miyazaki, where evolution of H_2 could be observed only in the presence of either cytochrome c_3 or methylviologen and not in the presence of ferredoxin. Cytochrome c_3 could also be reduced directly under an atmosphere of H_2 in the presence of hydrogenase.[23,24] The reaction of *Desulfovibrio* hydrogenase (to which the name hydrogen:ferricytochrome c_3 oxidoreductase was given, and EC 1.12.2.1) can therefore be formulated as shown.

$$2H_2 + (\text{cyt } c_3)(Fe^{3+})_4 \rightleftharpoons 4H^+ + (\text{cyt } c_3)(Fe^{2+})_4$$

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Table I
Spectral Properties of the Pure Cytochrome c_3 Preparation[a]

form	peak	wavelength of peak, nm	absorptivity, mM
ferri	α-β	530	37
	γ	410	460
	δ	350	86
	protein	280	37
ferro	α	552	110
	β	524	57
	γ	419	688
	δ	323	126

[a] The absorption spectrum of ferricytochrome c_3 could be recorded routinely. That of the ferro form could be recorded after enzymic reduction of cytochrome c_3 under H_2 in the presence of 1% by weight of the purified hydrogenase, whose absorbance at 280 nm was less than 2% that of cytochrome c_3, and much less in other region of the spectrum. See ref 9 for details.

Hydrogenases of other species of *Desulfovibrio* also use cytochrome c_3 as their natural electron carriers.[25,26] While the overall features of electron transfer in these bacteria have not been elucidated, cytochrome *c*-553, not cytochrome c_3, is known to accept electrons from formate[27] and lactate[28] in the presence of the respective dehydrogenases. Cytochrome c_3 is also one of the essential components in phosphoroclastic decomposition of pyruvate to produce H_2, and this reaction system needs ferredoxin[29] or flavodoxin[30] as an additional component. Another enzyme, sulfite reductase, can accept electrons from flavodoxin.[31] Another potential member of this electron transport chain is adenylyl sulfate reductase, which seems to accept electrons from cytochrome c_3,[32] although this must be confirmed by experiments using a purified enzyme preparation. Indeed, the mechanism of the enzyme transfer system involving cytochrome c_3, cytochrome *c*-553, flavodoxin, ferredoxin, and so on is not yet thoroughly understood.

Purification and Spectral Properties of Cytochrome c_3

When the cells of *D. vulgaris* Miyazaki are disrupted by sonication and centrifuged, about 80% of the total cytochrome c_3 in the cells are brought into the supernatant.[5] The cytochrome c_3 can be adsorbed on an Amberlite CG 50 (NH_4^+) column, eluted by 0.1 M aqueous NH_3, separated from other *c* cytochromes by gel filtration chromatography on Sephadex G-50, dialyzed against distilled water, and finally rechromatographed on an Amberlite CG 50 (NH_4^+) column to obtain highly purified cytochrome c_3 preparation.[9] Table I illustrates some spectral properties of the pure cytochrome c_3 preparation.

One of the spectral characteristics that has been overlooked is the sharpness of the γ peak in the ferro form

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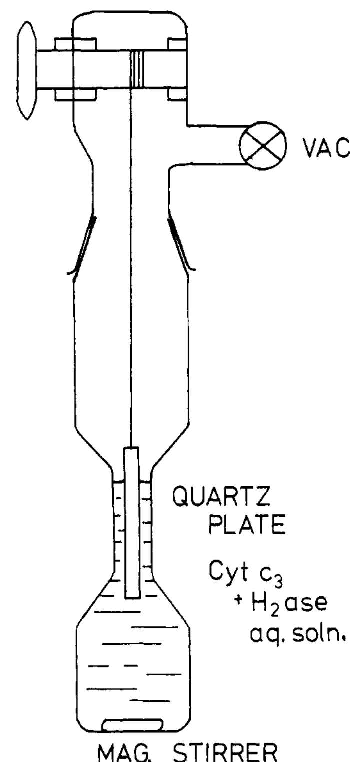

Figure 1. Apparatus for preparing cytochrom c_3 film.

(T. Yagi and K. Kimura, (unpublished observations). The width between the half-maximal points on both sides of the γ peak of ferrocytochrome c_3 is 15 nm, which is only 60% that of yeast cytochrome *c* (25 nm). It seems that the sharpness of the γ peak in the ferro form is a reflection of its strong heme–heme interaction in the excitonic structure. When the cytochrome c_3 loses its biochemical characteristics, such as the ability to act as an electron carrier for hydrogenase by any means, its γ peak width broadens considerably. For example, the γ peak width of the CO complex of ferrocytochrome c_3 is 19.4 nm and that of pyridine ferrohemochrome is 30 nm.

Purity of the Cytochrome c_3 Preparation

The purity of the cytochrome c_3 preparation is routinely expressed in terms of the purity index, which is defined as the height (absorbance) of the α peak of the ferro form relative to that at 280 nm of the ferri form. The purity index of the cytochrome c_3 preparation of the highest purity obtained was 3.0 for *D. vulgaris* Miyazaki,[9] 2.9–3.0 for *D. gigas*,[33,34] 3.30 for *D. desulfuricans* Norway,[13], and 3.1 for *D. vulgaris* Hildenborough.[35]

The magnetic susceptibility (χ) of cytochrome c_3 was determined by the rf-Squid method in the temperature range between 1.5 and 4.2 K under an external magnetic field of 176 Oe. It has been confirmed[36] that the magnetic susceptibility obeys the Curie–Weiss law, i.e., $\chi = C/(T - \theta)$, where $C = 216 \pm 18$ nJ K G^{-2} mol^{-1}, $\theta = 0.00 \pm 0.04$ K. It is remarkable to have θ equal to

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Table II
Mössbauer Effect of Cytochrome c_3

form	temp, K	isomer shift,[a] mm s^{-1}	quadrupole splitting, mm s^{-1}
ferri	300	0.18 ± 0.04	2.02 ± 0.04
	77	0.30 ± 0.04	2.04 ± 0.04
	4.2[b]	0.28 ± 0.04	2.12 ± 0.04
ferro	300	0.35 ± 0.04	1.08 ± 0.04

[a] Relative to iron metal absorber. [b] The spectrum at 1.6 K was the same as that obtained at 4.2 K.

zero; this shows that our cytochrome c_3 preparation was perfectly free from any paramagnetic and ferromagnetic impurities such as iron compounds. The g value calculated from these experiments was 2.39 ± 0.10, which is consistent with those obtained from Mössbauer spectrometery[37] or ESR measurements.[12]

Redox Reactions of Cytochrome c_3 in the Solid State

Purified hydrogenase from *D. vulgaris* was found to catalyze the reversible oxidoreduction of cytochrome c_3 not only in the aqueous solution but also in the solid state.[38,39] An anhydrous film containing purified ferricytochrome c_3 and purified hydrogenase at the molar ratio of 400:1 was prepared on a quartz plate in vacuo; a device to prepare this film is shown in Figure 1. When this film was kept under an atmosphere of H_2 at 102 kPa (1 atm), almost all the cytochrome c_3 molecules on the film were reduced in 3 days. The α peak position of the dry ferrocytochrome c_3 film was red-shifted by 1 nm compared to that in the aqueous solution. The time evolution curve of the reduction of cytochrome c_3 measured spectrophotometrically was found to obey first-order kinetics. When the H_2 atmosphere was evacuated to 13 mPa, the ferrocytochrome c_3 on the film was reoxidized to some extent. This reversible redox reaction could be repeated. Since almost all of the cytochrome c_3 molecules were reduced on the film and since the molar ratio of hydrogenase to cytochrome c_3 was as high as 1:400, it must be concluded that the cytochrome c_3 molecules not directly in contact with hydrogenase molecules must have been reduced catalytically.

The reduction of 400 molecules of cytochrome c_3 by a single hydrogenase molecule in the solid state strongly suggests that electrons as well as protons propagate among cytochrome c_3 molecules; i.e., the electron may be transferred from the H_2 to the nearest-neighbor cytochrome c_3 molecules and then propagate to other cytochrome c_3 molecules. A similar phenomenon was observed by Mössbauer effect studies on a cytochrome c_3–hydrogenase mixture at a molar ratio of 7000:1.[39]

Mössbauer Effect of Cytochrome c_3

The Mössbauer spectra of ferricytochrome c_3 were recorded at 300, 77, and 4.2 K[37] together with that of ferrocytochrome c_3 at 300 K,[39] and the results are summarized in Table II. Although the spectra at 300 and 77 K showed the same features as those for eucaryotic cytochrome c, the spectrum at 4.2 K showed no obvious magnetic hyperfine structure, in contrast to that of eucaryotic cytochrome c. The spectrum at 4.2 K was drastically affected by a small external magnetic field of less than a few hundred Oe, whereas that at 77 K was not affected by an external magnetic field of 120 Oe.

Careful examination of the spectra indicated that the distance between the iron ions is on the order of 1.0 nm (10 Å), which is about a half of that calculated with a simple cubic arrangement, that there is a superexchange interaction between the ferric ion that is a few times as strong as the dipolar interactions, and that the neighboring ferric ions have different g axes from one another even though they have the same principal values.[37] That is to say the orientation of the hemes is definitely anisotropic with respect to one another.

Structure of Cytochrome c_3

The relative molecular mass of cytochrome c_3 from *D. vulgaris* Hildenborough has been shown to be about 14 000.[17] The iron content was reported to be two[17,40,41] or three.[42,43] Direct iron analysis as well as spectrophotometric heme analysis of the highly purified cytochrome c_3 revealed that the cytochrome c_3 from *D. vulgaris* Miyazaki has four hemes in the molecule.[9] Other members of the cytochrome c_3 family isolated from various species of bacteria in genus *Desulfovibrio* are now believed to have a similar tetrahemoprotein structure.[10,11,12,13,44,-46]

Amino acid sequences of cytochrome c_3 from *D. vulgaris* Miyazaki,[47] *D. vulgaris* Hildenborough,[48,49] *D. gigas*,[50] *D. salexigens*,[51] *D. desulfuricans*,[11] and *D. desulfuricans* Norway[52] have been determined and are illustrated in Figure 2. There is rather poor homology among them; the closest ones, i.e., those from *D. vulgaris* Miyazaki and *D. vulgaris* Hildenborough, differ from each other by 14 residues out of 107 amino acid residues.[47] In the case of eucaryotic cytochromes c, those of human and pigeon differ from each other by only 12 out of 104 amino acid residues. However, poor homology in the case of the *Desulfovibrio* cytochrome c_3 family would not be surprising because these bacteria must have diverged very early during evolution.

Mössbauer spectroscopy of [^{57}Fe]cytochrome c_3 suggested that the four hemes were completely unequivalent in the molecule, that heme–heme interaction existed among the four hemes, and that the distances between the heme irons were of the order of 1 nm.[37] Unequivalence of the four hemes was also indicated by

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                     1                        2         3         4         5
                5    0           5            0    5    0    5    0    5    0    5
DvM         APKAPADGLKMD       KTK       QPVV FNHSTHKAVKCGDCHHPVNGKENYQKCATAGCHDNM
                * *              *          * * *  ●    ●  ***   *      *   ***
DvH         APKAPADGLKME       ATK       QPVV FNHSTHKSVKCGDCHHPVNGKEDYRKCGTAGCHDSM
                * *              ●          * * *  ●    *  ***   *      ●   ***
Dg          V DVPADGAKIDFIAG   GEK       NLVV FNHSTHKDVKCBBCHHBP GBKQYAGCTTDGCHNIL
                * *              *          * * *  *    ●  ***   *      ●   ***
Ds          V DAPAD MVLKAPAGAK MTK       AP VDFSHKGHAALDCTKCHHKWDGKAEVKKCSAEGCHVBT
                * *              ●          * * *  *    *  ***   ●      ●   ***
Dd          V DAPAD MVIKAPAGAK VTK       AP VAFSHKGHASMDCKTCHHKWDGAGAIQPCGASGCHANT
                * *              *          * * *  ●    *  ***   *      *   ***
DdN         A DAPGDDYVISAPEGMKAKPKGDKPGALQKTVPEPHTKHATVECVQCHHXADG GAVKKCTTSGCHDSL


                 6             7          8          9         0
                 0     5       0     5    0     5    0    5    0    5
DvM         D KKDK SAKGYY   HAMHD KGTKFKSCVGCH LETAGADAAKKKELTGCKGSKCHS
                               ●         *  **               * *    **
DvH         D KKDK SAKGYY   HVMHD KNTKFKSCVGCH VEVAGADAAKKKDLTGCKKSKCHE
                               *         ●  **               * *    **
Dg          D KADK SVNSWY   KVVHDAKGGAKPTCISCHK DKAGDDKELKKKLTGCKGSACHPS
                               *         *  **               * *    **
Ds           SKKGKKSTPKFY   SAFHS KSDI  SCVGCHKALKK      ATGPTKC G DCHPKKK
                               *         *  **               * *    **
Dd          ESKKGDDS   FY   MAFHERKSEK  SCVGCHKSMKK        GPTKC T ECHPKN
                               *         *  **               * *    **
DdN         E FRDKANAKDIKLVESAFHT       QCIDCH ALKKKD  KKPTGPTAC G KCHTTN
```

Figure 2. Sequence alignment of cytochrome c_3 family based on a new proposal by Higuchi et al.[56] DvM stands for *D. vulgaris* Miyazaki; DvH, *D. vulgaris* Hildenborough; Dg, *D. gigas*; Ds, *D. salexigens*; Dd, *D. desulfuricans*; and DdN, *D. desulfuricans Norway*.

ESR spectroscopy and the chemical reduction and reoxidation cycle of cytochrome c_3.[45] The remarkably low ionization potential of cytochrome c_3[53] as well as its NMR paramagnetic shift[54] suggested the close proximity of the heme irons. Proton magnetic resonance studies of cytochromes c_3 from *D. vulgaris* Hildenborough and from *D. gigas* in ^2H_2O indicated that all the heme irons in each molecule were in the low spin state and that histidine coordinated to each heme iron at the fifth and sixth ligands.[46] Complete three-dimensional structures of two members of the cytochrome c_3 family, one from *D. desulfuricans* Norway[52] and the other from *D. vulgaris*, Miyasaki,[55] have been elucidated. In spite of their poor homology in amino acid sequences, the relative orientations of the four hemes are strikingly alike.[55] Deletion and insertion of peptide fragments occur only in the outer loop regions of the molecules and have little influence on the relative heme orientations.[56] The heme–heme distances and heme–heme angles of these two cytochromes c_3 are given in Table III. This table shows that the distances between the heme irons are somewhat longer than expected, but the four hemes are unequivalent, as expected by physicochemical analyses as described in the preceding sections. The established heme orientations would help us in discussing some of the characteristics of cytochrome c_3 molecules presented in this Account.

Table III
Iron-Iron Distances and Heme-Heme angles[a]

heme no.	heme no. 1 (*1*)	2 (*4*)	3 (*3*)	4 (*2*)
1		1.63	1.81	1.28
1		*1.63*	*1.73*	*1.27*
2	71		1.24	1.62
4	*60*		*1.28*	*1.68*
3	22	89		1.13
3	?	*80*		*1.09*
4	80	64	82	
2	*90*	?	*85*	

[a] Distances in nm; angles in deg. Data for *D. vulgaris* Miyazaki are indicated in roman type; those for *D. desulfuricans* Norway are in italics.

Electrochemical Characteristics of Cytochrome c_3

One of the characteristic features of cytochrome c_3 is its very negative standard redox potential. It was first reported to be −205 mV[17,40,42] for *D. vulgaris* or −225 mV[33] for *D. gigas*. Redox equilibrium of cytochrome c_3 from *D. vulgaris* Miyazaki under specific H_2 pressure in the presence of purified hydrogenase revealed that the standard redox potential was −290 mV.[9] Precise analysis of the reversible voltammetric response of cytochrome c_3 revealed that the standard redox potentials for the four hemes are different (−226, −278, −298, and −339 mV), with their average being 285 mV.[57] Other members of the cytochrome c_3 family are now believed to have lower redox potentials that were reported earlier and are thus very close to that of cytochrome c_3 from *D. vulgaris* Miyazaki, some examples being −303 mV (average of −284, −310, −319, and −324 mV) for *D. vulgaris* Hildenborough,[58] −250 to −340 mV for *D. vulgaris* Hildenborough,[35] −160 to −340 mV for *D. desulfuricans*,[35] −273 mV (average of −235, −235, −306, and −315 mV) for *D. gigas*,[59] and −276 mV for *D. africanus*.[14]

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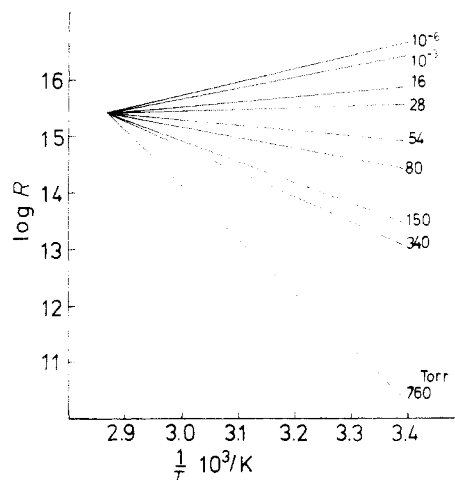

Figure 3. Temperature dependence of the resistance as function of hydrogen partial pressure. The figures on the right side of the straight line represent hydrogen pressure in torrs.

Another characteristics is the ability of cytochrome c_3 to interact with an electrode of an enzymic electric cell without the aid of any intermediate electron carrier of small size.[60] Cytochrome c_3 can be readily oxidized and reduced electrochemically in conventional polarography, cyclic voltammetry, and potentiometry.[61] Cytochrome c_3 molecules in solution are freely and reversibly oxidized and reduced through the strongly adsorbed layer of cytochrome c_3 on a mercury electrode, on which the electrochemical redox reaction is diffusion controlled.[61] These observations are remarkable, because in the case of other proteins including eucaryotic cytochrome c, the electrochemical redox reaction is confined to the layer of protein that is strongly adsorbed on the mercury electrode, and therefore no reversible reaction can be observed for the molecules in solution. These results also suggested to us that a layer of cytochrome c_3 molecules might conduct electrons fairly freely; this has shown to be true and is discussed in the next section.

Electrical Conduction of a Cytochrome c_3 Layer

In biological systems, there are several instances in which proteins are carrying electric charges. In mitochrondrial respiratory electron-transfer systems, flavoproteins and cytochromes transfer electrons from organic substrates to the final electron acceptor, O_2. In nerve tissues, nervous impulse is transmitted by means of a charge-transfer system involving proteins such as acetylcholine receptor. Eley and his school have found the semiconductive behavior of proteins from direct measurement of their electric conduction.[62] In spite of these electron-transfer functions of proteins, isolated simple protein molecules are rather insulative.[63]

A thin film of anhydrous cytochrome c_3 containing trace amount of hydrogenase was thus prepared on a

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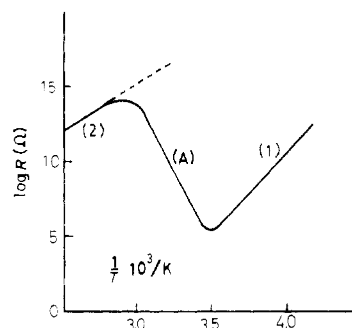

Figure 4. Temperature dependence of the resistance under a hydrogen pressure of 200 kPa.

quartz plate, and its electrical conductivity was measured in vacuo or under H_2.[64] As shown in Figure 3, the electrical conductivity of cytochrome c_3 in the temperature range between 295 and 350 K was a function of H_2 pressure and temperature.

As a ferro form/ferri form ratio is a function of the H_2 pressure in the presence of hydrogenase, this figure suggested to us that the conductivity of the ferrocytochrome c_3 would be much higher than that of ferricytochrome c_3. Accordingly, the conductivity of cytochrome c_3 at a fixed temperature of 296 K under various H_2 pressure was measured. It was confirmed that the conductivity of the cytochrome was proportional to the third power of H_2 pressure, as shown:

$$I = 0.11(P/102\ \text{kPa})^{3.1}\text{p}A$$

where P is the H_2 pressure and I is the electric current of the cytochrome c_3 film measured under the specified conditions described in ref 65.

The change in electrical conductivity as a function of temperature under a fixed pressure (200 kPa) of H_2 (Figure 4) revealed puzzling characteristics of electric conduction of this electron carrier molecule. The curve in this figure consists of three parts: below 286 K (part 1) the electric conduction is that of ferrocytochrome c_3 and is of a semiconductor type with very high activation energy (ΔE = 370 kJ/mol); above 346 K (part 2) the conduction is that of ferricytochrome c_3 in spite of the fact that the cytochrome is in equilibrium under 200 kPa of H_2 and is also of semiconductive type (ΔE = 160 kJ/mol). Absorption spectroscopic measurement revealed that ferri- and ferrocytochrome c_3 equilibrium is a function of temperature and that the equilibrium is inclined to ferriform above 346 K. Between 286 and 346 K (part A) the conduction is due to an equilibrium mixture of the ferri and ferro forms of cytochrome c_3 with an apparently negative activation energy of electric conduction. At critical point at 286 K where two lines (1 and A) cross, the proportion of the ferrocytochrome c_3 was spectroscopically estimated to be 99.97%. Under the higher H_2 pressure, we could extend line 1 a little longer to obtain much better conductivity, such as 56 Ω cm at 102 kPa and 8 Ω cm at 200 kPa. However, we were unable to change the other critical point at 346 K. The significance of this critical point remains to be elucidated, but it must be emphasized that the process of conductivity measurements with changing tempera-

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Table IV
Ionization Potentials of Some Metalloporphyrin Thin Film

	divalent form, kJ/mol	trivalent form, kJ/mol
horse cytochrome *c*	560	589
cytochrome c_3	444	521
zinc tetraphenylporphyrin	482	
protohemin		511

ture was perfectly reproducible either from 243 to 393 K or from 393 to 243 K.

Ionization Potential of Cytochrome c_3

The electrochemical characteristics of cytochrome c_3 described in the preceding sections might be a reflection of the electronic state of the hemes in the cytochrome c_3 molecule that is not shared by other heme groups in single-heme proteins such as eucaryotic cytochrome *c*. Electronic energy states of porphyrins in hemoproteins can be investigated by spectroscopic methods such as optical absorption, emission, magnetic resonance, or Mössbauer effect, but measurement of ionization potentials of the ferri and ferro forms of cytochrome c_3 together with those of monoheme-type cytochrome *c* would provide us with more crucial information for the determination of absolute energy levels. Accordingly they were measured, giving the results summarized in Table IV.[52,66] This table also includes the ionization potentials of zinc tetraphenylporphyrin and protohemin as the standards for divalent and trivalent metal derivatives of porphyrin.

In the case of eucaryotic cytochrome *c*, the difference in ionization potentials between the ferri and ferro forms was 29 kJ/mol, which is in accord with the difference between typical trivalent and divalent metal porphyrins, protohemin and zinc tetraphenylporphyrin. This means that the polarization energy of the metalloporphyrin structure induced by the protein moiety of eucaryotic cytochrome *c* was identical in both redox states. This is in accord with the fact that the conformation change during the reduction of eucaryotic cytochrome *c* is minor, as indicated by X-ray crystallographic measurements.[67]

By contrast, in the case of cytochrome c_3 where the four hemes are unequally oriented to form a heme cluster, the difference of ionization potentials between the ferri and ferro forms was as much as 77 kJ/mol. This remarkable difference may well be ascribed to the change in polarization energy induced by the protein moiety, change in electronic states of heme clusters, or an extraordinarily large intermolecular interaction in cytochrome c_3 film, which can be associated with the unusual electrical properties of cytochrome c_3 as described in the preceding sections. It has to be pointed out that the ionization potential of ferrocytochrome c_3 (444 kJ/mol) is the lowest in several porphyrin derivatives and phthalocyanines (near or over 482 kJ/mol)[68] and even lower than that of graphite (453 kJ/mol).[69,70]

Closing Remarks

In this Account we have presented some characteristics of cytochrome c_3 that make this molecule unique among organometallic compounds from either biological or nonbiological origins. An understanding of the theoretical basis of these characteristics of the tetrahemoprotein system would help us to discover as yet unexpected characteristics from biological materials.

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Biosynthetic Studies of Macrolide and Polyether Antibiotics

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There are two schools of thought concerning the value of heuristic studies of the biosynthesis of natural products. One is that the formation of natural products like alkaloids, antibiotics, and terpenoids must involve complex and, perhaps, unusual biochemical transformations. Investigation of their biosynthesis, consequently, is worthwhile to broaden our insight about the scope of biochemical events. A similar argument can be made for many other scientific endeavors: it is the diversity of scientific achievement, rather than just its singularity, that enriches the basis of our knowledge. The other school is that many of these natural products are useful to mankind as chemotherapeutic, insecticidal, color, and flavor, etc., agents. Information about their biosynthesis, therefore, has practical value to organi-

Charles Richard (Dick) Hutchinson was born in Dayton, Ohio, in 1943 and remained near there until receiving his undergraduate degree at the Ohio State University in 1966. After completing a Ph.D. in organic Chemistry with Professor Edward Leete at the University of Minnesota (1970), he spent a brief period as a postdoctoral fellow with Professor Alan Battersby at Cambridge University (1971). His independent research career began at the University of Connecticut (1971-1974) and has continued since 1974 at the University of Wisconsin, where he is a professor of medicinal chemistry. Major research interests in his laboratory have been the biosynthesis of natural products (alkaloids, antibiotics) and the medicinal chemistry of naturally occurring antitumor agents.

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