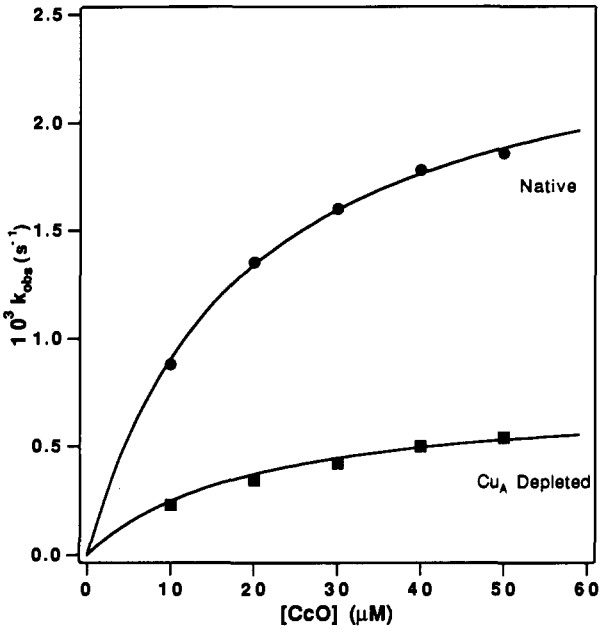

Figure 3. Kinetics of ET between ferrocytochrome *c* and CcO at various CcO concentrations and an ionic strength of 110 mM. The reaction conditions are as shown in Figure 1, and the concentration of cytochrome *c* is unchanged as the concentration of CcO is varied. The pseudo-first-order rate constants for the reduction of cytochrome *a* during the fast phase are plotted as a function of the concentration of CcO. The solid curves represent the best fits of the data to eq 3.

ferrocytochrome *c* to cytochrome *a*. Since the reduction potential of cytochrome *a* is significantly higher than that of Cu_A when the enzyme is fully oxidized, cytochrome *a* is expected to provide the ultimate disposition of the electron prior to subsequent ET to the dioxygen reduction site. Our experiments with the Cu_A-depleted enzyme show, however, that cytochrome *a* can also accept an electron directly from cytochrome *c*, albeit at a decreased kinetic rate. Thus we have the distinct possibility that there exist two distinct electron-input ports for ET from ferrocytochrome *c* to CcO, with the more facile ET proceeding via Cu_A.

New Type of Charge-Transfer Complex from an Antiaromatic Electron Donor. Possible Radical Cation Stabilization by the Captodative Effect

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One of the requirements for formation of conducting organic charge-transfer (CT) salts is the formation of resonance-stabilized radical-ion species from reaction of donor and acceptor molecules.[1] Design of suitable component molecules might incorporate aminocarboxy captodative stabilization as in the Kosower radical[2] or 3,5,5-trimethyl-2-oxomorpholinyl radical.[3] We report the synthesis and characterization of a novel electron donor for CT salts based on the 1,4-dihydropyrazine ring system, 4a,8a-diaza-2,6-dioxa-3,4,7,8-tetrahydro-4,4,8,8-tetramethylanthracene-1,5-dione (**1**, DDTTA), and its oxidation to an exceptionally persistent, captodatively stabilized radical cation. We also report

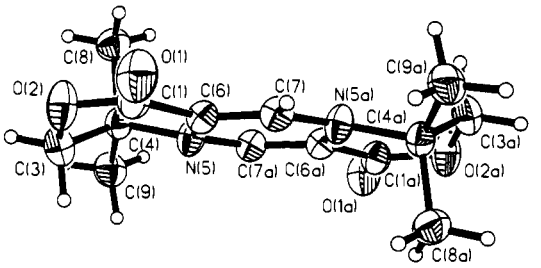
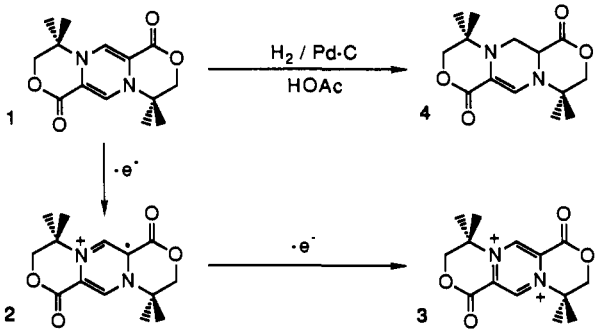


Figure 1. Thermal ellipsoid plot of **1** showing the numbering scheme adopted.

the structure and electrical properties of the complex of **1** with the electron acceptor tetracyanoquinodimethane (TCNQ).



3-(Chloromethyl)-5,6-dihydro-5,5-dimethyl-1,4-oxazin-2-one, synthesized via the method of Himmelsbach et al.[4] from 5,6-dihydro-3,5,5-trimethyl-1,4-oxazin-2-one and *tert*-butyl hypochlorite, underwent base-promoted self-condensation in *N,N*-dimethylformamide to give DDTTA in 22% yield, ^{1}H NMR ($CDCl_3$) δ 1.22 (s, 12 H), 3.79 (s, 4 H), 6.28 (s, 2 H). DDTTA was isolated as a bottle green, crystalline solid, soluble in a variety of solvents to give blue to blue-violet solutions. The color of **1** appears to be due to a symmetry-forbidden π-π^* band in the 590–630-nm region. Both the position and extinction coefficient of this band are highly solvent dependent. Increasing the Lewis acidity of the solvent (as measured by acceptor numbers)[5] increased both the wavelength and extinction coefficient, suggesting strong coordination of the first excited state to the solvent. Such a visible absorption band has not been observed for any other stable 1,4-dihydropyrazine derivative. Though the 1,4-bis(trialkylsilyl) and 1,4-bis(trimethylgermyl) derivatives are colored (yellow and red, respectively), this was attributed to intramolecular charge transfer from the ring to low-lying orbitals at the trialkylsilyl or trialkylgermyl substituents.[6] X-ray diffraction measurements on a single crystal of **1** (Figure 1) revealed that the central ring is somewhat elongated (cf. cyclobutadiene[7] and push-pull substituted cyclobutadienes[8]) and close to planar, the mean deviation of the six ring atoms from a least-squares plane being 0.02 Å. The central ring can thus be considered to be an "antiaromatic" eight-π-electron ring system. Further evidence for this came from breaking the annular conjugation of the system. Thus, catalytic hydrogenation of **1** gave 4a,8a-diaza-2,6-dioxa-3,4,7,8,9,9a-hexahydro-4,4,8,8-tetramethylanthracene-1,5-dione (**4**), in which the remaining vinylic proton resonates at δ = 7.14 ppm. Similar paratropic shifts have been observed in other stable 1,4-dihydropyrazines;[9] such an effect is considered evidence for an "antiaromatic ring" in a single-ring system.[10]

(1) Cowan, D. O. *New Aspects of Organic Chemistry I*; VCH: Basel, 1990; pp 177–225.
(2) Kosower, E. M.; Waits, H. P.; Teverstein, A.; Butler, L. *J. Org. Chem.* **1978**, *43*, 800.
(3) Olson, J. B.; Koch, T. H. *J. Am. Chem. Soc.* **1986**, *108*, 756.
(4) Himmelsbach, R. J.; Barone, A. D.; Kleyer, D. L.; Koch, T. H. *J. Org. Chem.* **1983**, *48*, 2989.
(5) Gutmann, V. *Electrochim. Acta* **1976**, *21*, 661.
(6) Baumgaten, J.; Bessenbacher, C.; Kaim, W.; Stahl, T. *J. Am. Chem. Soc.* **1989**, *111*, 2126.
(7) Masamune, S.; Souto-Bachiller, F. A.; Machiguchi, T.; Bertie, J. E. *J. Am. Chem. Soc.* **1978**, *100*, 4889.
(8) Gompper, R.; Holsboer, F.; Schmidt, W.; Seybold, G. *J. Am. Chem. Soc.* **1973**, *95*, 8479.
(9) Kaim, W. *J. Am. Chem. Soc.* **1983**, *105*, 707.
(10) Mallion, R. B. *Pure Appl. Chem.* **1980**, *52*, 1541.

0002-7863/91/1513-5910$02.50/0

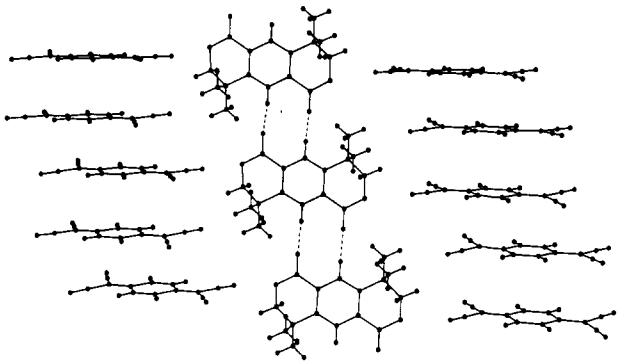


Figure 2. Packing diagram of the TCNQ salt of 1. The short intermolecular O-H distances are shown as dotted lines.

Table I. Bond Lengths and Angles for the Central Ring of 1 and 2

	bond lengths, Å			bond angles, deg	
	1	2		1	2
N(5)-C(6)	1.443 (1)	1.393 (3)	N(5)-C(6)-C(7)	119.5 (1)	119.3 (2)
C(6)-C(7)	1.343 (2)	1.355 (3)	C(6)-C(7)-N(5a)	123.9 (1)	122.4 (2)
N(5)-C(7a)	1.351 (2)	1.343 (3)	C(6)-N(5)-C(7a)	116.3 (1)	118.3 (2)

Cyclic voltammetry on a solution of 1 in methylene chloride with tetrabutylammonium perchlorate supporting electrolyte revealed two reversible oxidation waves at -0.33 V and 0.61 V vs ferrocene/ferricinium corresponding to oxidation of 1 to the radical cation 2 and dication 3, respectively. The former was characterized by its five-line ESR signal (relative intensity 1:2:3:2:1, $g = 2.0035$, $a_N = 6.9$ G) when an acetonitrile solution of 1 was oxidized with 1 equiv of ferrin, $[Fe(phen)_3]^{3+}$. The high stability of 2 (calculation gives $K_{disproportionation} = 1.5 \times 10^{-16}$) is probably due in part to captodative stabilization[11] or merostabilization[12] and also to the high coulombic destabilization of 3 and the "antiaromatic destabilization" of 1.

When 1 was added to 1 equiv of TCNQ in hot acetonitrile under an inert atmosphere, an emerald green, paramagnetic, air-sensitive solution was formed, presumably containing 2 and the TCNQ radical anion. An ESR spectrum of this solution revealed only a broad unresolved signal. Upon slow cooling of the solution, purplish-black, opaque needles were deposited having the stoichiometry $DDTTA(TCNQ)_2$. X-ray analysis of a single crystal (Figure 2) revealed chains of stacked TCNQ dimers with "ribbons" of 2 aligned edge to edge in between the chains. Within these ribbons, the distance between the carbonyl oxygen and ring hydrogen on a neighboring molecule is rather short (2.38 Å), suggesting a small "edge on" interaction along the ribbon. At ambient temperature the TCNQ chains exhibit a Peierls distortion, which gives rise to the semiconducting nature of the material.[1] Initial (two-probe) measurements gave a room temperature conductivity on the order of 10^{-3} Ω^{-1} cm^{-1}. Measurement of the degree of charge transfer[13] gave $Z = 0.57$ per TCNQ molecule. Interestingly, this corresponds to a positive charge of 1.14 per DDTTA molecule. The central ring of the DDTTA became planar and the N(5)-C(6) bond became shorter by 0.05 Å as DDTTA became "partially aromatic" (Table I). The mean deviation of the six central ring atoms from a least-squares plane is now 0.001 Å.

Though the electrical properties of this complex are not exceptional, the donor represents an interesting new organic structure with three stable one-electron redox states. Electron acceptors other than TCNQ may give rise to higher conductivities. Variation of the substitution on the 1,4-dihydro-2,5-bis(alkoxycarbonyl)-pyrazine unit may lead to further novel CT complexes and provide insight into the electronic structure of this interesting chromophore.

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Supplementary Material Available: A complete description of the X-ray crystallographic determinations of 1 and the TCNQ salt of 1 including atomic coordinates, isotropic and anisotropic displacement parameters, bond lengths, bond angles, and torsion angles (17 pages); listing of observed and calculated structure factors for 1 and the TCNQ salt of 1 (10 pages). Ordering information is given on any current masthead page.

(11) Stella, L.; Janousek, Z.; Merényi, R.; Viehe, H. G. *Angew. Chem., Int. Ed. Engl.* **1978**, *17*, 691. Viehe, H. G.; Janousek, Z.; Merényi, R.; Stella, L. *Acc. Chem. Res.* **1985**, *18*, 148.
(12) Baldock, R. W.; Hudson, P.; Katritzky, A. R. *J. Chem. Soc., Perkin Trans. 1* **1974**, 1422. Katritzky, A. R.; Zerner, M. C.; Karelson, M. M. *J. Am. Chem. Soc.* **1986**, *108*, 7213.
(13) Chappell, J. S.; Bloch, A. N.; Bryden, W. A.; Maxfield, M.; Poehler, T. O.; Cowan, D. O. *J. Am. Chem. Soc.* **1981**, *103*, 2442.

Pulsed Endor Study of the Native and High pH Perturbed Forms of the Blue Copper Site in Stellacyanin

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The spectroscopic properties and electronic structure of type 1 (T1) copper sites in blue copper proteins have been intensively studied.[1-3] In the prototypical blue copper proteins plastocyanin[4] and azurin,[5] where X-ray crystal structures have been published, the chromophore is $Cu[N(HIS)]_2S(CYS)S(MET)$ in a distorted T_d coordination geometry. Stellacyanin, a blue Cu protein isolated from the Japanese lacquer tree, *Rhus vernicifera*, is unusual in several respects.[6] Stellacyanin has no protein methionine residues, has the lowest redox potential (+184 mV) of all blue copper proteins, and exists in a reversible perturbed form between pH 9 and 11.5 which has slightly different spectral properties but still has T1 character.[6,7] Neither the fourth ligand on copper which replaces the thioether sulfur ligand in the normal blue copper sites nor the nature of the high pH perturbed form is known.

In this communication, we present data from pulsed ENDOR and a new two-dimensional pulsed electron nuclear electron triple resonance method for recording hyperfine selective ENDOR (HS-ENDOR) spectra[8,9] on the native (pH 6.4) and high pH (pH 11.0) perturbed forms of the blue Cu site in stellacyanin. ENDOR lines for two imino imidazole nitrogens and two strongly coupled methylene protons from a cysteine thiolate ligand are observed at both pH 6.4[10] and pH 11.0. A third nitrogen coupling is observed at pH 11. This fourth ligand, which exhibits much larger hyperfine anisotropy than imidazole imino nitrogen couplings,[10,11]

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(1) Malkin, R.; Malmstrom, B. G. *Adv. Enzymol.* **1970**, *33*, 177-244.
(2) Fee, J. A. *Struct. Bonding (Berlin)* **1975**, *23*, 1-60.
(3) Gray, H. B.; Solomon, E. I. In *Copper Proteins*; Spiro, T. G., Ed.; Wiley-Interscience: New York, 1981.
(4) Colman, P. J.; Freeman, H. C.; Guss, J. M.; Murata, M.; Norris, V. A.; Ramshaw, J. A. M.; Venkatappa, M. P. *Nature (London)* **1978**, *272*, 319-324.
(5) Norris, G. E.; Anderson, B. F.; Baker, E. N. *J. Am. Chem. Soc.* 1986, *108*, 2784-2785.
(6) Peisach, J.; Levine, W. G.; Blumberg, W. E. *J. Biol. Chem.* **1967**, *242*, 2847-2858.
(7) Malmstrom, B. G.; Reinhammer, B.; Vanngard, T. *Biochim. Biophys. Acta* **1970**, *205*, 48-57.
(8) Thomann, H.; Bernardo, M. *Chem. Phys. Lett.* **1990**, *169*, 5-11.
(9) Buhlmann, C.; Schweiger, A.; Ernst, R. R. *Chem. Phys. Lett.* **1989**, *154*, 285-291.
(10) Roberts, J. E.; Cline, J. F.; Lum, V.; Freeman, H. C.; Gray, H. B.; Peisach, J.; Reinhammer, B.; Hofmann, B. M. *J. Am. Chem. Soc.* **1984**, *106*, 5324-5330.

0002-7863/91/1513-5911$02.50/0