oxy-siloxy carbene-carbynes (cf. refs 20 and 21) parallels the reported stability of bis(dialkylamino)carbynes.[6,8,22] We have thus far been unable to obtain the isoelectronic bis(siloxy)carbynes.[6,23] Coupling of unsubstituted "bis-carbyne" fragments has been reported to occur spontaneously.[24,25]

Some interesting questions remain to be answered for the present system. Addition of the aluminum reagent to **3** has brought the carbon atoms of the carbene-carbyne ligands of **4** closer together. Would a stronger electrophile (e.g., R_3Si^+) result in the distance closing to that required for acetylene formation (Scheme I, **1** to **1b**)? Does the extent of the interaction of the electrophile determine the position of the equilibrium illustrated in Scheme I (**1a** ⇌ **1b**), or does C-C bond formation require addition of a nucleophile to the metal to drive the carbene-carbyne ligands together in a seven-coordinate complex to accomplish coupling (**1a** to **2**)?[5] Experiments are in progress to address these issues.

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Supplementary Material Available: Tables of rate data and reaction order plots, listings of full characterization and experimental details for **3** and **4**, complete ORTEP diagrams of **3** and **4**, and tables of positional and thermal parameters (24 pages). Ordering information is given on any current masthead page.

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Nitric Oxide-Triggered Heme-Mediated Hydrolysis: A Possible Model for Biological Reactions of NO

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The recent discoveries of wide-ranging physiological effects of nitric oxide,[1-12] in most cases associated with the enzyme guanylate cyclase,[13] have stimulated increased interest in the chemistry and biology of nitric oxide.[14-17] The UV-vis spectrum of guanylate cyclase, which contains a rather loosely bound five-coordinated protoheme,[13] changes from one which resembles ferrous myoglobin to a species with a Soret band at 399 nm upon addition of nitric oxide.[13,18] This change is attributed to the loss of the proximal base (eq 1). The corresponding reaction of myoglobin gives a six-coordinate complex (λ_{max} = 419 nm).[19] The spectroscopic change in guanylate cyclase is accompanied by an activation of this enzyme, resulting in the conversion of GTP to cyclic GMP.[3-13]

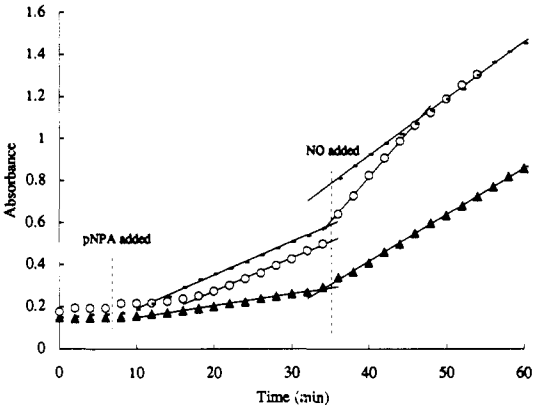
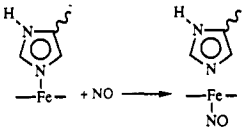
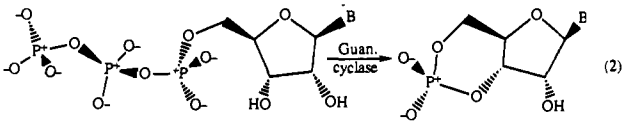


Figure 1. Absorbance of *p*-nitrophenolate (405 nm) in the aqueous phase (3 mL) stirred with dichloromethane solutions: (-) MCPH-CO (5 μmol) in dichloromethane (0.3 mL); (▲) 1-MeIm-Hm-CO heme (5 μmol) in 0.3 mL of dichloromethane; (○) 1-MeIm-Hm-CO (10 μmol) in dichloromethane (0.3 mL). The substrate (pNPA, 25 μmol in 50 μL of dichloromethane) and NO gas (2 mL) were added at the points indicated.



(1)



(2)

The process in eq 1 suggests two possible mechanisms for catalysis of the reaction. Either the released imidazole acts as a general or nucleophilic catalyst[20] or the breaking of the iron-imidazole bond results in a change to another enzyme conformation which favors GTP binding and possibly aids in the catalytic steps. The first mechanism is particularly attractive if the catalytic site is near the heme, as has been suggested by Ignarro et al.[21] Examples of heme-induced conformational change are numerous; a possible model system for this type of mechanism was recently suggested[22] in which peptide helices were stabilized by binding a metal to two properly juxtaposed imidazoles.

We present here a model for the first mechanism and demonstrate NO-triggered catalysis of a hydrolysis reaction. We have

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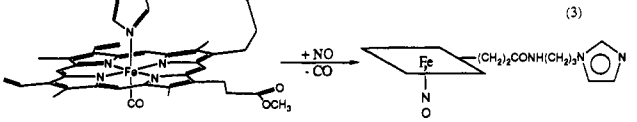
Table I. Rates of *p*-Nitrophenyl Acetate Hydrolysis Catalyzed by 1-Methylimidazole Before and After NO Addition

PHDME⁺Cl⁻ (μmol)	1-MeImid (μmol)	rate of ester hydrolysis before NO adn (ng/s)[a]	rate of ester hydrolysis after NO adn (ng/s)[a]
10	10	8.0	21.7
5	5	2.9	11.3
0	5		12.1[b]
5[c]	0	2.0[d]	2.2[d]
5[e]	5[e]	8.2	13.8

[a] Concentrations were calculated based upon aqueous solution of sodium *p*-nitrophenolate at pH 7.5. [b] No nitric oxide was added. [c] PHDME⁺Cl⁻ is not reduced. [d] This value corresponds to the rate of hydrolysis of *p*-nitrophenyl acetate by OH⁻. [e] MCPH⁺Cl⁻ was used instead of PHDME⁺Cl⁻; the 5 μmol of imidazole is attached to the heme.

shown that addition of NO converts the carbonmonoxy-chelated heme to the five-coordinate iron(II)–NO complex (eq 3).[23] The



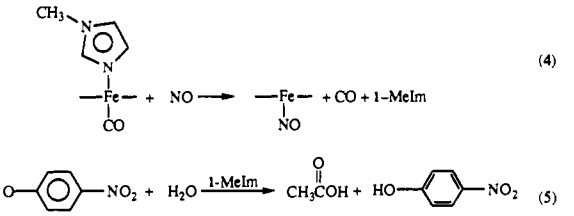
(3)

same change is observed for the 1-methylimidazole–protoheme–CO complex at low (<1 M) concentrations of 1-methylimidazole.[14] These observations suggest that the complexes might mimic the catalytic behavior of guanlyate cyclase.

We have chosen the general base-catalyzed hydrolysis of *p*-nitrophenyl acetate (pNPA)[24] to investigate the use of the model system as a trigger for such hydrolytic reactions. In order to facilitate the spectroscopic observation of *p*-nitrophenolate in the presence of optically dense heme solutions, we have carried out the reaction in a two-phase system. An aqueous buffer (3 mL at pH 7.5) was layered over a methylene chloride solution (0.3 mL) of either 5 μmol of chelated protoheme–CO (MCPH–CO) or 5 μmol of 1-methylimidazole–protoheme dimethyl ester–CO (Im-PHDME-CO). All experiments were under 1 atm of CO throughout. Reduction was accomplished with ascorbic acid. Gentle stirring of the methylene chloride layer (with a magnetic bar) was briefly interrupted at intervals to determine the absorbance of the upper layer. Figure 1 shows three absorbance versus time plots for a solution of chelated protoheme–CO: (1) in the absence of pNPA; (2) after its addition; and (3) after addition of NO gas (2 mL, 1 atm) to the aqueous phase. A similar series of plots for the 1:1 mixture of protoheme dimethyl ester and 1-methylimidazole under 1 atm of CO is also shown in Figure 1. The rates of *p*-nitrophenyl acetate hydrolysis for the control systems, in which either the heme or the 1-methylimidazole was absent, are given in Table I.

Figure 1 indicates an increase in the rate of hydrolysis upon the addition of NO which, as seen in Table I, is about the same as the rate increase upon addition of the same concentration of 1-methylimidazole. The heme alone does not alter the rate. Furthermore, the initial spectrum of the diluted heme from the reaction mixture is of imidazole–heme–CO, while the spectrum after hydrolysis corresponds to the heme–NO complex. It can be seen from Table I that the hydrolysis rate increase is proportional to the initial concentration of 1-MeIm-Hm-CO, as is expected if, upon addition of NO, all imidazole is released. Clearly, catalysis of this hydrolysis is due to released imidazole (as represented by eqs 4 and 5 for the 1-MeIm-Hm-CO).

Stimulation of imidazole catalysis of a hydrolytic reaction upon addition of NO to the chelated protoheme–CO complex represents an intriguing mechanism for biological catalysis. Although nitric oxide stimulation of guanylate cyclase might proceed by other mechanisms, our model presents a viable explanation. Continuing



(4)

(5)

discoveries of the physiological effects of NO[25,26] suggest that both base catalysis and conformational triggering mechanisms might occur in biological systems.

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A High-Potential Ferrous Complex and Its Conversion to an Alkylperoxoiron(III) Intermediate. A Lipoxygenase Model

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Lipoxygenases are mononuclear non-heme iron enzymes which catalyze the peroxidation of fatty acids containing a 1,4-diene unit.[1] Spectroscopic evidence suggests that the native soybean lipoxygenase-1 possesses a six-coordinate high-spin Fe(II)[2,3] center with histidine and carboxylate ligands and a redox potential around 0.6 V vs NHE.[4] Treatment of the Fe(II) enzyme with product peroxide converts it to the active Fe(III) enzyme, which in turn reacts with excess peroxide to form a metastable purple species with λ_{max} = 570 nm.[1,5] In our efforts to model active sites of non-heme iron proteins, we have synthesized a high-spin ferrous complex that approximates the iron coordination environment proposed for native soybean lipoxygenase and affords a transient intermediate species upon treatment with alkyl hydroperoxide.

Reaction of equimolar amounts of $Fe(ClO_4)_2 \cdot 6H_2O$, TLA,[6,7] HOBz, and Et_3N in methanol/H_2O affords $[Fe(TLA)(OBz)]ClO_4$ (**1**)[8] as a light yellow powder. The structure of the BPh_4 salt[9]

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(9) Crystal data for the BPh_4 salt ($C_{52}H_{49}BFeN_4O_2$·acetone, fw 886.72) at 176 K: yellow prism, monoclinic, space group $P2_1$(No.4); a = 13.347 (3) Å, b = 9.336 (5) Å, c = 18.760 (8) Å; β = 91.45 (3)°, V = 2337 (3) Å³, Z = 2. For 9965 unique, observed reflections with $I > 2\sigma(I)$ and 557 parameters, the current discrepancy indices are R = 0.050 and R_w = 0.054.

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