Model Reactions of Molybdo-Reductase. A Novel and Highly Efficient Reduction of Nitrobenzene to Aniline Catalyzed by a Molybdenum-Mediated Oxygen Atom Transfer Reaction

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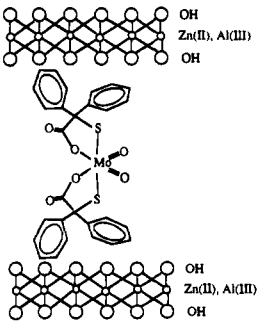
Nitroreductases, such as xanthine dehydrogenase, quinone reductase, and the nitroreductases of enteric bacteria, capable of reducing nitroaromatic compounds, are well known.[1] The enzymatic reduction of the nitrogenous substrate is believed to proceed by oxygen atom transfer from the nitrogen center to the $Mo^{IV}O$ center of the enzyme.[2] The $Mo^{VI}O_2$ center thus formed is then re-reduced by one of a variety of physiological reducing agents by coupled electron−proton transfers.[3,4] Generally, the physiological reductant does not react directly with the Mo center but rather reduces another prosthetic group in the molybdoenzyme, which then transfers the electrons to the Mo center one at a time.[4]

Mechanistic models for $Mo^{VI}O_2/Mo^{IV}O$ interconversion have been tested by studying the reactivity of three synthetic dioxomolybdenum(VI) complexes sterically hindered from dimerization:[5] (i) $MoO_2(t\text{-BuLNS}_2)$[6] and $MoO_2(LNS_2)$[7] [LNS_2 = 2,6-bis(2,2-diphenyl-2-mercaptoethyl)pyridine(2−)] with bidentate and tridentate pyridinethiolate ligands, respectively, and (ii) $LMoO_2X$ [L = hydrotris(3,5-dimethyl-1-pyrazolyl)borate, X = PhS^-, NCS^-].[8] These complexes, or their oxomolybdenum(IV) derivatives, have been shown to be capable of cleanly catalyzing the transfer of an oxygen atom from a suitable donor (dialkyl *S*-oxide, amine *N*-oxide, or nitrate) to an acceptor (trialkylphosphine or aromatic thiol). In the case of nitrate ions, an oxo transfer reaction involving the $Mo^{IV}O(LNS_2)/Mo^{VI}O_2(LNS_2)$ complexes allows the reduction of nitrate to nitrite.[9]

Other features of enzyme behavior such as the generation of Mo(V) centers are neatly accounted for by coupled electron−proton transfer reactions. The formation of Mo(V) centers was first observed in a very early modeling experiment,[10] showing that molybdate ions react with 1,2-ethanedithiol or thioglycolic acid to produce unidentified EPR active Mo(V) species in equilibrium with inactive Mo(V) dimers. These observations have now been interpreted in terms of coupled electron−proton transfer reactions of the type shown by eqs 1 and 2.

Chart 1



$$\{Mo^{VI}O_2[O_2CC(S)Ph_2]_2\}^{2-} + 2RSH \rightarrow \{Mo^{IV}O[O_2CC(S)Ph_2]_2\}^{2-} + RS{-}SR + H_2O \quad (1)$$

$$\{Mo^{VI}O_2[O_2CC(S)Ph_2]_2\}^{2-} + \{Mo^{IV}O[O_2CC(S)Ph_2]_2\}^{2-} + 2H^+ \rightarrow 2\{Mo^{V}O[O_2CC(S)Ph_2]_2\}^{-} + H_2O \quad (2)$$

Our previous work[11] on the sterically bulky bis(2-sulfanyl-2,2-diphenylethanoxycarbonyl)dioxomolybdate(VI) complex $[MoO_2(O_2CC(S)Ph_2)_2]^{2-}$ (**I**) has demonstrated that the $Mo^{VI}O_2/Mo^{V}O$ reduction consists of a proton-assisted process, which is quantitative even in the presence of excess thiol and/or moderate amounts of water. This feature, which is reminiscent of enzymatic systems, implies, however, an important limitation to the catalytic activity of **I**. Such a limitation stems from the well-known difficulty of reoxidizing $Mo^{V}O$ to $Mo^{VI}O_2$, preventing the use of common oxidants such as dialkyl *S*-oxide or amine *N*-oxide in the design of catalytic cycles.

Herein we show that thiols are viable electron donors in the **I**-mediated catalytic reduction of nitrobenzene to aniline. Oxidation of thiophenol to disulfide releases water and produces a reduced $Mo^{IV}O$ form of **I** (eq 1). The participation of this reduced $Mo^{IV}O$ species in the comproportionation reaction 2 is blocked by intercalation of the anionic complex **I** within a hydrotalcite-like Zn(II)−Al(III) layered double-hydroxide (LDH) host. Especially significant from the standpoint of enzyme-catalyzed reactions is the ability of this reduced $Mo^{IV}O$ species to react with nitrobenzene, returning to the oxidized $Mo^{VI}O_2$ parent complex.

In a recent study,[12] we described an ion-exchange method to replace nitrate with complex **I** in LDH. Elemental analysis of the solid product supports its formulation as $[Zn_{3-x}Al_x(OH)_6]^{x+}[(NO_3)_{x-y}\mathbf{I}_{y/2}]^{x-}\cdot H_2O$ ($x = 0.75$). Data from powder X-ray diffraction, IR, diffuse reflectance, and thermogravimetric analyses confirmed intercalation of **I** within a 13-Å-high gallery (see Chart 1).[13] Intercalation is also corroborated by the similarity of the Mo 3d X-ray photoelectron spectra for both **I** and **I**-LDH, which show two well-resolved peaks of very similar overall shape and apparent binding energy (spectra and a table of the energies are given in the supplementary material).

Intercalation of molybdenum complexes into zeolite cages or LDH layers inhibit comproportionation reactions by keeping the oxidized and reduced species apart, just as proteins do in enzymes.[14,15] Not surprisingly then, **I**−LDH catalyzes the

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oxidation of thiophenol (PhSH) by dioxygen or dimethyl sulfoxide, which is consistent with I−LDH being the active oxidant and a Mo^{IV} O−LDH species the active reductant.[12]

The heterogeneous catalytic activity of I−LDH for the reduction of nitrobenzene ($PhNO_2$) by PhSH was investigated under strictly anaerobic conditions in the 35−75 °C temperature range. In a typical experiment, 200 mg of solid I−LDH (0.065 mmol of intercalated complex I) and 200 mg of solid amberlite IR-120(H) were stirred in 20 mL of ethanol containing 650 mg of $PhNO_2$ (5.3 mmol) and a variable excess of PhSH (90−225 mmol). Amberlite acts as a proton source that presumably promotes the catalytic protonation of an oxo ligand, increasing considerably the rate of the thiol oxidation reaction.[11] Aliquots of 100 μL were monitored for reactants (PhSH and $PhNO_2$) and products (PhS−SPh and $PhNH_2$) by gas chromatography (GC) and mass spectrometry. The complete reduction of all $PhNO_2$ occurred in 4 h at 75 °C, while no reaction was detected in the absence of I−LDH. Aniline was found as the only reduction product of $PhNO_2$.[16] After the reaction, I−LDH was filtered and reused with no loss of catalytic activity. The identity of its XPS spectrum with that obtained before the reaction confirms the catalytic behavior of the intercalated complex I.

While the detailed mechanism of this catalytic reaction is not yet certain, the following observations severely limit the possibilities. GC analysis indicates that 6 mol of PhSH are consumed for each mole of aniline produced. The rate of aniline formation is a complex function of both $PhNO_2$ and PhSH concentration, and it is also linearly dependent on the amount of I−LDH catalyst used. In a separate homogeneous reaction without I−LDH catalyst, nitrosobenzene (PhNO) reacts instantaneously with PhSH, yielding 2 mol of PhS−SPh/mol of $PhNH_2$ (eq 7). Taking these features together, we propose a mechanism for this catalytic reaction in terms of eqs 3−7 (reactants and products in bold type). This mechanism parallels that generally accepted for most enzymatic reductions of nitroaromatic compounds, with nitroso and hydroxylamino species as intermediates.[17]

$$\{Mo^{VI}O_2[O_2CC(S)Ph_2]_2\}^{2-} + \mathbf{Ph\text{-}SH} \xrightarrow{k_1} \{Mo^{V}O(OH)[O_2CC(S)Ph_2]_2\}^{2-} + \text{Ph-S}\cdot \quad (3)$$

$$\{Mo^{V}O(OH)[O_2CC(S)Ph_2]_2\}^{2-} + \mathbf{Ph\text{-}SH} \xrightarrow{fast} \{Mo^{IV}O[O_2CC(S)Ph_2]_2\}^{2-} + \text{Ph-S}\cdot + \mathbf{H_2O} \quad (4)$$

$$2\ \text{Ph-S}\cdot \xrightarrow{fast} \mathbf{Ph\text{-}S\text{-}S\text{-}Ph} \quad (5)$$

$$\{Mo^{IV}O[O_2CC(S)Ph_2]_2\}^{2-} + \mathbf{PhNO_2} \xrightarrow{k_2} \{Mo^{VI}O_2[O_2CC(S)Ph_2]_2\}^{2-} + \text{PhNO} \quad (6)$$

$$\mathbf{PhNO} \xrightarrow[2e^-/2H^+]{Fast} \mathbf{PhNHOH} \xrightarrow[2e^-/2H^+]{Fast} \mathbf{PhNH_2} + \mathbf{H_2O} \quad (7)$$

$$\mathbf{2\ PhSH} \qquad\qquad \mathbf{PhSSPh} \quad \mathbf{2\ PhSH} \qquad\qquad \mathbf{PhSSPh}$$

By applying the steady state condition to both Mo(IV) and Mo(VI) concentrations, we obtain

$$-\frac{d[PhNO_2]}{dt} = \frac{k_1k_2w_0[PhSH]_0}{k_1[PhSH]_0 + k_2[PhNO_2]}[PhNO_2] \quad (8)$$

where w_0 (= $[Mo^{VI}O_2$−LDH] + $[Mo^{IV}O$−LDH]) represents the initial number of moles of I in LDH per unit of volume. Integration of eq 8 yields eq 9.

$$\ln\frac{[PhNO_2]}{[PhNO_2]_0} = -k_2w_0t + \frac{k_21}{k_16}\ln\left(\frac{[PhSH]_0}{[PhSH]_0 - 6([PhNO_2]_0 - [PhNO_2])}\right) \quad (9)$$

The reaction of $PhNO_2$ with a 20−50-fold excess of PhSH gave a $PhNO_2$ concentration *vs* time curve (see Figure 1) consistent with the foregoing mechanism expressed by eq 9. Thus, values of the coefficients k_2w_0 [(1.07 ± 0.09) × 10^{-2} min^{-1} for w_0 = 0.200 g] and k_2/k_1 could be obtained by means of a multiple linear least-squares regression.[18] All fits gave good correlations ($r > 0.999$), but the k_2/k_1 accuracy was poor because the logarithmic term on the right-hand side of eq 9 was always considerably smaller than the time term. As expected, a linear increase of k_2w_0 was obtained by increasing w_0.

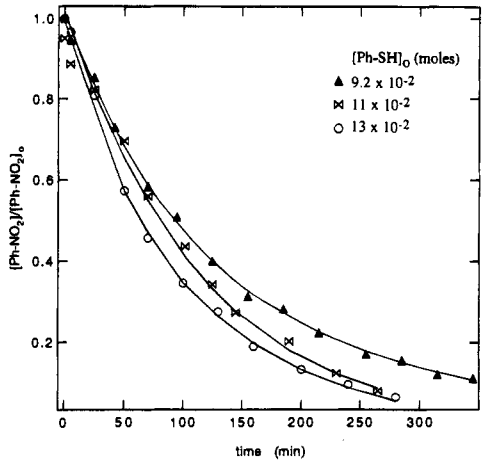

Figure 1. Experimental (symbols) and theoretical (solid line) variation of $[PhNO_2]/[PhNO_2]_0$ ratio *vs* time at different PhSH initial concentrations. T = 75 °C. Experimental data are given in the text.

Alternatively, the reciprocal initial rate of disappearance of $PhNO_2$ was found to be linearly related to $1/[PhSH]_0$ (eq 8). From the slope ($1/k_1w_0$) and origin ordinate ($1/k_2w_0[PhNO_2]_0$) of such a representation, k_2w_0 and k_2/k_1 coefficients were recalculated. The value of k_2w_0, (1.07 ± 0.12) × 10^{-2} min^{-1} for w_0 = 0.200 g, is in excellent agreement with the above-determined value using the integrated eq 9, which corroborates the stability of the I−LDH catalyst. The value of k_2/k_1, ~10^2, indicates that reduction of $PhNO_2$ (eq 6) proceeds 100-fold faster than oxidation of PhSH (eq 3).

In conclusion, the study reported here provides strong evidence that oxygen atoms can be transferred from nitroaromatic compounds to Mo(IV) species. However, a direct oxygen atom transfer reaction involving both species has not been unambiguously demonstrated by a ^{18}O-labeling experiment.

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Supplementary Material Available: Molybdenum XPS spectra for I−LDH and pure complex I with data and binding energy data (4 pages). This material is contained in many libraries on microfiche, immediately follows this article in the microfilm version of the journal, can be ordered from the ACS, and can be downloaded from the Internet; see any current masthead page for ordering information and Internet access instructions.

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(16) The reduction of nitrobenzene to aniline in the presence of a homogeneous transition metal catalyst has been widely explored since the work of l'Eplattenier *et al.*[19] using $[Ru_3(CO)_{12}]$. The recent literature[20] shows that efficient catalytic reduction using $[Ru_3(CO)_{12}]$ can be performed either *via* direct hydrogenation under water−gas shift conditions or *via* generation of hydrogen from formic acid or ammonium formate. Uncatalyzed oxidations of thiols by nitro- and nitrosobenzene in basic media have been also reported.[21] Under such conditions, only azoxy or azo nitrogen-containing products exhibiting an intermediate oxidation state are formed.

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