4: Conjugate **4** was synthesized on a solid-phase peptide synthesizer. Standard fmoc-protected amino acids (fmoc = 9-fluorenylmethoxycarbonyl) were used for the synthesis. Conjugate **4** was purified by HPLC and analyzed with ^{31}P NMR spectroscopy and mass spectrometry (Figure 1). ^{31}P NMR (121 MHz, D_2O): $\delta = -137.91$ (s). ^{31}P NMR (121 MHz, proton coupled, D_2O) $\delta = -137.90$ (t, $J_{P,H} = 190.2$ Hz). LR-MS (FAB) m/z calcd for $[M^+ + H]$: 1591.9; found: 1592.0.

5: To a solution of **4** (0.5 mg) in ethanol (400 μL) and DMF (100 μL) were added 0.1N HCl (25 μL) and 37% aqueous formaldehyde (25 μL), and the reaction mixture was stirred at room temperature (25 °C) for 5 min. The formation of P_2S_2-D-Lys^6-LHRH hydroxymethylphosphonium chloride was confirmed by the ^{31}P NMR signal at $\delta = 31.39$(s). The P_2S_2-D-Lys^6-LHRH hydroxymethylphosphonium chloride was converted into **5** by the addition of 1M aqueous sodium bicarbonate (30 μL) in near quantitative yields as demonstrated by the ^{31}P NMR chemical shift at $\delta = -24.23$.

Received: December 9, 1998
Revised version: February 9, 1999 [Z12764IE]
German version: *Angew. Chem.* **1999**, *111*, 2152 – 2155

Keywords: bioorganic chemistry · peptide conjugates · phosphanes

[1] K. Severin, R. Bergs, W. Beck, *Angew. Chem.* **1998**, *110*, 1722 – 1743; *Angew. Chem. Int. Ed.* **1998**, *37*, 1634 – 1654.

[2] a) B. K. Keppler, C. Friesen, H. Vongerichten, E. Vogel in *Metal Complexes in Cancer Chemotherapy* (Ed.: B. K. Keppler), VCH, Weinheim, **1993**, pp. 297 – 323; b) P. Köpf-Maier, H. Köpf in *Metal Compounds in Cancer Therapy* (Ed.: S. P. Fricker), Chapman & Hall, London, **1994**, pp. 109 – 146; c) C. Christodoulou, D. Ferry, D. Fyfe, A. Young, J. Doran, G. Sass, A. Eliopoulos, T. Sheehan, D. J. Kerr, *Proc. 88th Annu. Meeting Am. Assoc. Cancer Res.* **1997**, *38*, 222; d) S. G. Ward, R. C. Taylor in *Metal-based Antitumor Drugs* (Ed.: M. F. Gielen), Freund, London, **1988**, pp. 1 – 54.

[3] a) P. E. Garrou, *Chem. Rev.* **1985**, *85*, 171 – 185; b) *Transition-Metal Complexes of Phosphorus, Arsenic, and Antimony Ligands* (Ed.: C. A. McAuliffe), Wiley, New York, **1973**; c) C. A. McAuliffe, W. Levason, *Phosphane, Arsenine, and Stibine Complexes of Transition Elements*, Elsevier, New York, **1979**; d) W. Levason, C. A. McAuliffe, *Acc. Chem. Res.* **1978**, *11*, 363 – 368.

[4] a) K. Drauz, A. Kleeman, J. Martens, *Angew. Chem.* **1982**, *94*, 590 – 613; *Angew. Chem. Int. Ed. Engl.* **1982**, *21*, 584 – 608; b) A. Mori, H. Abe, S. Inoue, *Appl. Organomet. Chem.* **1995**, *9*, 189 – 197; c) P. Kvintovics, B. R. James, B. Hail, *J. Chem. Soc. Chem. Commun.* **1986**, 1810 – 1811; d) H. Brunner, B. Reiter, G. Riepl, *Chem. Ber.* **1984**, *117*, 1130 – 1354.

[5] a) B. L. Iverson, S. A. Iverson, V. A. Roberts, E. D. Getzoff, J. A. Tainer, S. J. Benkovic, R. A. Lerner, *Science* **1990**, *249*, 659 – 662; b) L. A. Regan, *Rev. Biophys. Biomol. Struct.* **1993**, *22*, 257 – 281; c) J. T. Kellis Jr., F. H. Arnold, *BioTechnology* **1991**, *9*, 994 – 995.

[6] a) T. Janáky, A. Juhász, Z. Rékási, P. Serfözö, J. Pinski, L. Bokser, G. Srkalovic, S. Milovanovic, T. W. Redding, G. Halmos, A. Nagy, A. V. Schally, *Proc. Natl. Acad. Sci. USA* **1992**, *89*, 972 – 976; b) J. Pinski, A. V. Schally, T. Yano, K. Szepeshazi, G. Halmos, K. Groot, A. M. Comaru-Schally, S. Randulovic, A. Nagy, *Prostate (NY)* **1993**, *23*, 165 – 178.

[7] M. A. DeRosch, J. W. Brodack, G. D. Grummon, M. E. Marmion, D. L. Nosco, K. F. Deutsch, E. Deutsch, *J. Nucl. Med.* **1992**, *33*, 850; b) A. M. Forster, A. E. Storey, K. R. Nagel, F. S. Brooker, B. Edwards, H. K. Gill, J. D. Kelly, M. McPartlin, *J. Nucl. Med.* **1992**, *33*, 850.

[8] For recent reviews and papers in radiolabeled biomolecules see: a) S. R. Karra, R. Schibli, H. Gali, K. V. Katti, T. J. Hoffman, C. Higginbotham, G. L. Sieckman, W. A. Volkert, *Bioconjugate Chem.* **1999**, *10*, 254 – 260; b) W. C. Eckelman, R. E. Gibson, *Nuclear Imaging in Drug Discovery, Development and Approval* (Eds.: H. D. Burns, R. F. Gibson, R. F. Dannals, P. K. S. E. Siegl), Birkhäuser, Basel, **1993**, pp. 113 – 134; c) A. J. Fischman, J. W. Babich, J. W. Strauss, *J. Nucl. Med.* **1993**, *34*, 2253 – 2263; d) A. R. Fritzberg, L. M. Gustavson, M. D. Hylarides, J. M. Reno, *Chemical and Structural Approaches to Rational Drug Design* (Eds.: D. B. Weiner, W. V. Williams), CRC Press, Boca Raton, FL, **1994**, pp. 125 – 158; e) J. C. Reubi, *J. Nucl. Med.* **1995**, *36*, 1825 – 1835; f) W. H. Brakker, R. Albert, C. Bruns, *Life Sci.* **1991**, *49*, 1583 – 1591.

[9] a) S. R. Gilbertson, G. Chen, M. Mcloughlin, *J. Am. Chem. Soc.* **1994**, *116*, 4481 – 4482; b) S. R. Gilbertson, X. Wang, G. S. Hoge, C. A. Klug, J. Schaefer, *Organometallics* **1996**, *15*, 4678 – 4680.

[10] a) K. V. Katti, H. Gali, D. E. Berning, C. J. Smith, *Acc. Chem. Res.* **1999**, *32*, 9 – 17; b) D. E. Berning, K. V. Katti, C. L. Barns, W. A. Volkert, *J. Am. Chem. Soc.* **1999**, *121*, 1658 – 1664; c) C. J. Smith, V. S. Reddy, K. V. Katti, *J. Chem. Soc. Dalton Trans.* **1998**, 1365 – 1370; d) D. E. Berning, K. V. Katti, C. L. Barnes, W. A. Volkert, *Chem. Ber.* **1997**, *130*, 907 – 911; e) D. E. Berning, K. V. Katti, C. L Barnes, W. A. Volkert, A. R. Ketring, *Inorg. Chem.* **1997**, *36*, 2765 – 2769; f) C. J. Smith, V. S. Reddy, K. V. Katti, L. J. Barbour, *Inorg. Chem.* **1997**, *36*, 1786 – 1791; g) C. J. Smith, K. V. Katti, W. A. Volkert, L. J. Barbour, *Inorg. Chem.* **1997**, *36*, 3928 – 3935; h) C. J. Smith, K. V. Katti, C. Higginbotham, W. A. Volkert, *J. Labelled Compd. Radiopharm.* **1997**, *40(S1)*, 444 – 446; i) V. S. Reddy, K. V. Katti, C. L. Barnes, *J. Chem. Soc. Dalton Trans.* **1996**, 1301 – 1304; j) C. J. Smith, V. S. Reddy, K. V. Katti, *Chem. Commun.* **1996**, 2557 – 2558; k) V. S. Reddy, K. V. Katti, D. E. Berning, W. A. Volkert, A. R. Ketring, C. L. Barnes, *Inorg. Chem.* **1996**, *35*, 1753 – 1757; l) V. S. Reddy, K. V. Katti, W. A. Volkert, *J. Chem. Soc. Dalton Trans.* **1996**, 4459 – 4462; m) K. V. Katti, *Curr. Sci.* **1995**, *70*, 219 – 225; n) K. V. Katti, V. S. Reddy, P. R. Singh, *Chem. Soc. Rev.* **1995**, *24*, 97 – 107; o) V. S. Reddy, K. V. Katti, C. L. Barnes, *Inorg. Chim. Acta* **1995**, *240*, 367 – 370.

[11] a) J. L. Cabioch, J. M. Denis, *J. Organomet. Chem.* **1989**, *377*, 227 – 233; b) E. C. Ashby, J. Prather, *J. Am. Chem. Soc.* **1966**, *88*, 729.

[12] a) M. J. S. Dewar, E. G. Zoebisch, E. F. Healy, J. J. P. Stewart, *Quantum Chemistry Programme*, Exchange Package No. 455, 1985; b) H. Gali, K. V. Katti, unpublished results.

[13] D. E. C. Corbridge, *Phosphorus-An Outline of its Chemistry, Biochemistry and Technology*, Elsevier, New York, **1990**.

[14] A. F. Wagner, E. Walton, G. E. Boxer, M. P. Pruss, F. W. Holly, K. Folkers, *J. Am. Chem. Soc.* **1956**, *78*, 5079 – 5081.

Modeling a Nitrogenase Key Reaction: The N_2-Dependent HD Formation by D_2/H^+ Exchange**

Dieter Sellmann* and Anja Fürsattel

Dedicated to Professor Helmut Werner on the occasion of his 65th birthday

Biological N_2 fixation is one of the fundamental natural synthetic processes and is catalyzed by FeMo, FeV, or FeFe nitrogenases.[1] X-ray structure analyses have revealed the molecular structure of FeMo nitrogenase and its active centers, in particular the structure of the FeMo cofactors (FeMoco).[2] However, the intimate molecular mechanism of biological N_2 reduction and the concomitant "obligatory dihydrogen evolution" (OHE) has remained a mystery. The OHE is an integral part of enzymatic N_2 reduction and cannot

[*] Prof. Dr. D. Sellmann, Dipl.-Chem. A. Fürsattel
Institut für Anorganische Chemie der Universität Erlangen-Nürnberg
Egerlandstrasse 1, D-91058 Erlangen (Germany)
Fax: (+49) 9131-852-7367
E-mail: sellmann@anorganik.chemie.uni-erlangen.de

[**] Transition Metal Complexes with Sulfur Ligands. Part 137. This work was supported by the Deutsche Forschungsgemeinschaft and the Fonds der Chemischen Industrie. Part 136: D. Sellmann, J. Utz, F. W. Heinemann, *Inorg. Chem.* submitted.

be suppressed even under high pressures of N_2. Equation (1) gives the (probably) limiting stoichiometry for FeMo nitrogenase.[1, 2] The key feature of this OHE is the N_2-dependent formation of HD in the presence of D_2 [Eq. (2)], first

$$N_2 + 8H^+ + 8e^- \longrightarrow 2NH_3 + H_2 \quad (1)$$

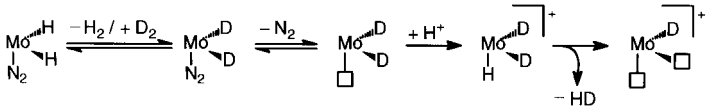
$$N_2 + 8H^+ + 8e^- \xrightarrow{D_2} 2NH_3 + (H_2, HD) \quad (2)$$

observed back in 1960.[3] The HD must result from D_2 and protons derived from H_2O. The dependence on N_2 is stringent, and N_2 cannot be replaced by any other nitrogenase substrate, for example C_2H_2, N_3^-, or N_2O. Electron balance studies prove that one electron is required per each molecule HD formed.[4]

The stringent N_2 dependence of HD formation proves the intimate coupling of OHE and N_2 reduction, and it compellingly demands the occurrence of a reduction intermediate that only is able to form with N_2. Elucidation of its molecular mechanism can therefore be expected to shed light also on the mechanistic details of N_2 reduction and the function of the FeMo cofactors.

Two mechanisms have been postulated in order to explain the N_2-dependent HD formation. Both the "trihydride" and the "diazene" mechanism suggest that nitrogenase (as isolated in the dithionite reduced state) must first be reduced before HD formation can occur, explaining that NH_3, H_2 as well as HD formation are electron-requiring processes.

The "trihydride" mechanism is based on the kinetic Lowe–Thorneley scheme of nitrogenase and model reactions of dinitrogen(phosphane)molybdenum complexes such as $[Mo(N_2)_2(PR_3)_4]$.[5] Successive electronations and protonations transfer nitrogenase into a stage in which the Mo center of FeMoco binds N_2 and hydride ligands (Scheme 1). Exchange



Scheme 1. "Trihydride" mechanism of the N_2-dependent HD formation.

of the hydride ligands with D_2, loss of N_2, and protonation of the resulting vacant coordination site (□) gives a $[Mo(H)(D)_2]$ species which liberates HD. A direct interaction between the N_2 and hydrogen/deuterium ligands occurs at no stage. In the decisive step of HD formation, N_2 does not bind to the Mo center, and N_2 essentially acts as "stand-in" ligand only. This raises the question why N_2 is indispensable for HD formation and cannot be replaced by other nitrogenase substrates known to function likewise as two-electron ligands, for example, N_3^- or N_2O.

The "diazene" mechanism avoids this problem by proposing N_2H_2 as enzyme-bound pivotal N_2 reduction intermediate formed by Equation (3).[3, 4] D_2 attack upon the N_2H_2 intermediate is postulated to result in formation of HD and enzyme-bound N_2 according to Equation (4). Equation (4)

$$N_2 + 2H^+ + 2e^- \longrightarrow N_2H_2 \quad (3)$$

$$\text{Enzyme}(N_2H_2) + D_2 \longrightarrow \text{Enzyme}(N_2) + 2HD \quad (4)$$

sums up to a "D_2-catalyzed N_2H_2 decomposition".[4] The "diazene" mechanism is supported by the electron balance ($1e^-$ per HD), but raises the question why N_2 should first get reduced in order to be subsequently reoxidized by D_2(!). The intimate molecular mechanism remained speculative because a chemical equivalent could not be found that catalyzed the H/D exchange between molecular D_2 and N_2H_2 whose protons ultimately had to come from water. Here we want to report the first example for such a reaction.

The ruthenium complex $[(\mu\text{-}N_2H_2)\{Ru(PCy_3)('S_4')\}_2]$ (**1a**) was synthesized by the method described for the homologous $[(\mu\text{-}N_2H_2)\{Fe(PPr_3)('S_4')\}_2]$ ($H_2'S_4' = 1,2$-bis(2-sulfanylphenylthio)ethane).[6] N_2H_2 was generated from $K_2N_2(CO_2)_2$ and acetic acid and trapped by $[Ru(dmso)(PCy_3)('S_4')]$ in THF. Acetolysis of $K_2N_2(CO_2)_2$ with CH_3COOD yielded the deuterated derivative $[(\mu\text{-}N_2D_2)\{Ru(PCy_3)('S_4')\}_2]$ (**1b**).[7] The spectroscopic properties of **1a**/**1b** (see Figure 2a and 3a) and X-ray structure determinations of analogous $[(\mu\text{-}N_2H_2)\{Ru(PR_3)('S_4')\}_2]$ complexes (R = Ph, *i*Pr)[8] support the structure indicated in Figure 1 for for **1a**. A *trans*-diazene

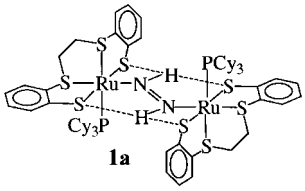


Figure 1. Schematic representation of the structure of **1a**; only one of the two diastereomeres existing in solution at room temperature is shown.

ligand bridges two enantiomeric $[Ru(PCy_3)('S_4')]$ fragments and gives rise to two (unsymmetrical) bifurcated $N{-}H\cdots(S)_2$ bridges. Complex **1a**, like most complexes of this type, forms two diastereomers in solution. Both are centrosymmetric and each diastereomer gives rise to one ^1H NMR diazene signal (Figure 2a). The two "hydrogen bridge diastereomers" differ only in the positioning of the $N{-}H\cdots(S)_2$ bridges.[8b]

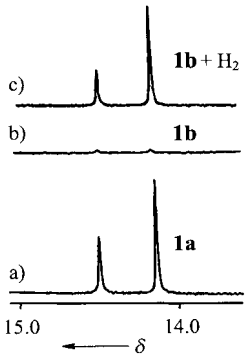


Figure 2. Diazene region of the ^1H NMR spectra (CD_2Cl_2) of a) **1a**, b) **1b**, and c) **1b** after complete reaction with H_2.

Under standard conditions (1 bar, 25 °C) treatment of **1a** with molecular D_2 gave **1b** and HD [Eq. (5a)]. Higher pressures resulted in accordingly faster turnovers. Likewise, **1b** reacted with H_2 to give **1a** and HD [Eq. (5b)]. (Absolutely

$$\mathbf{1a} + 2D_2 \xrightarrow{CH_2Cl_2} \mathbf{1b} + 2HD \quad (5a)$$

$$\mathbf{1b} + 2H_2 \xrightarrow{CD_2Cl_2} \mathbf{1a} + 2HD \quad (5b)$$

anhydrous conditions were observed and gases were dried with Na/K alloy in order to exclude any interference with the reactions according to Equation (6) (below)). Formation of

 © WILEY-VCH Verlag GmbH, D-69451 Weinheim, 1999 1433-7851/99/3813-2024 $ 17.50+.50/0 *Angew. Chem. Int. Ed.* **1999**, *38*, No. 13/14

either **1a** or **1b** was monitored by their characteristic $^1H/^2D$ NMR diazene signals in the region 14–15 ppm (shown in Figure 2b, c for reaction (5b)). Formation of HD was confirmed by mass spectrometry and ^1H NMR spectroscopy. In the reaction of **1b** with H_2 (35 bar) in a NMR pressure tube, in addition to the signal for **1a**, a 1:1:1 triplet ($^1J(H,D) = 42.51$ Hz) was detected at $\delta = 4.57$ for the resulting HD (Figure 3b). Because the Equations (1), (2), and (3), imply

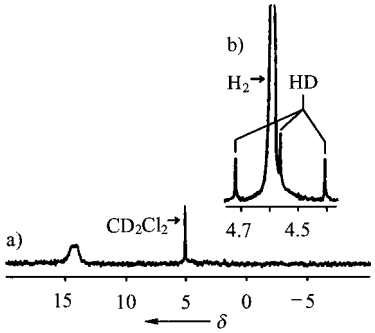


Figure 3. a) ^2H NMR spectrum of **1b** in CH_2Cl_2; b) H_2/HD region of the ^1H NMR spectrum of **1b** in CD_2Cl_2 after reaction with 35 bar of H_2.

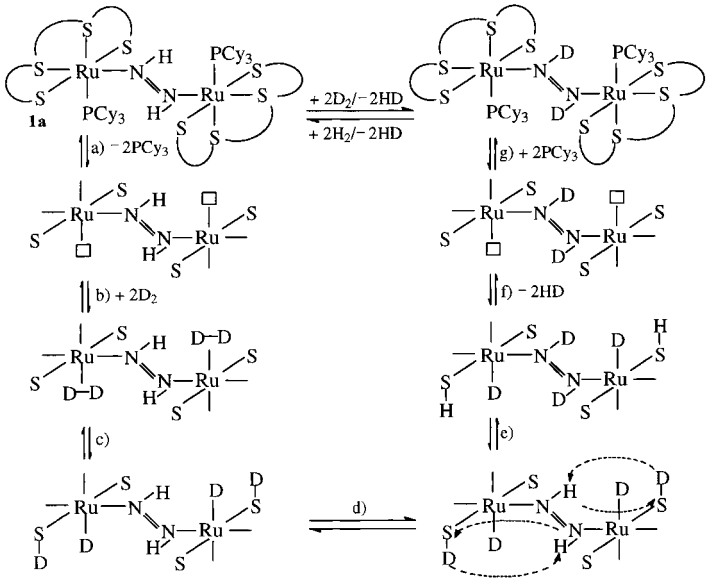
that the protons of reductively formed N_2H_2 must derive from H_2O, also the H^+/D^+ exchange of **1a**/**1b** was probed according to Equation (6). As in the case of the isoelectronic Fe complex

$$\mathbf{1a} + D_2O \overset{CD_2Cl_2}{\rightleftharpoons} \mathbf{1b} + H_2O \qquad (6)$$

$[(\mu\text{-}N_2H_2)\{Fe(PPr_3)('S_4')\}_2]$[9] a reversible H^+/D^+ exchange was observed. It is noteworthy that this H^+/D^+ exchange took place about seven times slower than the D_2/H^+ exchange according to Equation (5). Combining reactions (5) and (6) thus proves that HD formation results from D_2 exchange with N_2H_2 whose protons can derive from water.

Insight into the D_2/H^+ exchange mechanism of reaction (5a) results from previous findings: 1) The complexes $[Rh(H_2O)(PCy_3)('S_4')]BF_4$ and $[Ru(dmso)(PCy_3)('S_4')]$, which have labile H_2O and dmso ligands and are closely related to **1**, catalyze the heterolytic cleavage of H_2 (or D_2) via $[M(\eta^2\text{-}H_2)]$ and $[M(H)(SH)]$ hydride–thiol intermediates. They also catalyze the scrambling of hydride ligands and thiol protons.[10] 2) The N_2H_2 complex **1a** exchanges its PCy_3 for $PiPr_3$ ligands under retention of the $[\mu\text{-}N_2H_2\{Ru('S_4')\}_2]$ entity. This indicates that, through dissociation of PCy_3, **1a** can provide vacant Ru sites for the addition and heterolytic cleavage of H_2.[7b, 10]

Combination of these results and Equations (5) suggests the mechanism given in Scheme 2 for the NH/D_2 or ND/H_2 exchange of **1**. Essential core atoms of **1** are the Ru centers, thiolate donors, and diazene atoms. Dissociation of PCy_3 (step a) yields vacant sites (□) to which D_2 adds (step b) that is cleaved heterolytically into D^- and D^+ by the concerted action of the Lewis acidic Ru centers and Brønsted basic thiolate donors (step c). Intramolecular scrambling of the acidic diazene protons and thiol deuterons (steps d and e) gives the N_2D_2 species, which releases HD (step f). Readdition of PCy_3 yields **1b** (step g).



Scheme 2. D_2/NH exchange of **1a**. The arrows in the lower right formula should only be considered to indicate intramolecular H/D exchange which is possible following D_2 heterolysis; it should not be considered to imply simultaneous H/D migration.

These results thus support a "diazene" mechanism that explains the N_2 dependence of nitrogenase-catalyzed HD formation. They need, however, further discussion, because Equations (5) evidently contrast with Equation (4). Equations (5) show that H_2 or D_2 attack upon bound diazene does not decompose N_2H_2 to give N_2. They rather suggest that the N_2-dependent HD formation takes place on the diazene level. In other words: The diazene reduction stage stays preserved in the course of HD formation and does not switch back to the N_2 level. This necessitates an important conclusion with regard to the overall mechanism of the N_2-dependent HD formation. Reaction (5a), rewritten as Equation (7), utilizes only one half of the D_2 for HD formation, binds the other half in the diazene, and does *not* require electrons. In order to utilize also the diazene-bound deuterium and to make the HD formation catalytic, the reaction according to Equation (8) must take place. Adding Equation (7) to Equation (8) gives Equation (9). Equation (9) fulfills the experimentally established stoichiometry of nitrogensase-catalyzed N_2-dependent HD formation (1e$^-$ per HD).

$$N_2H_2[M]_2 + 2D_2 \longrightarrow N_2D_2[M]_2 + 2HD \qquad (7)$$

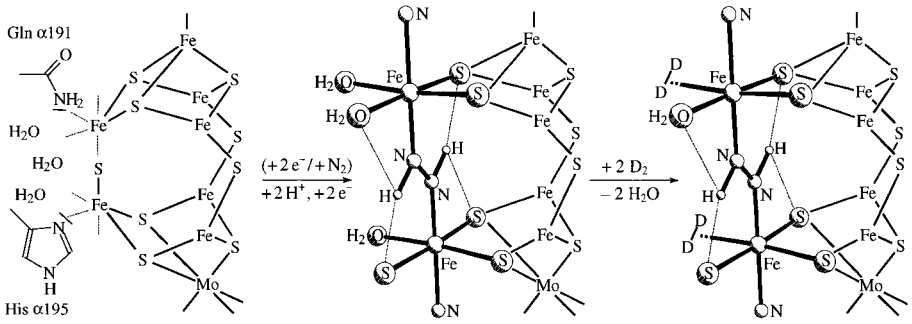
$$N_2D_2[M]_2 + 4H^+ + 4e^- \longrightarrow N_2H_2[M]_2 + 2HD \qquad (8)$$

$$4H^+ + 4e^- + 2D_2 \overset{N_2H_2[M]_2}{\longrightarrow} 4HD \qquad (9)$$

These considerations show that even on the diazene level two distinguishable pathways of HD formation have to be taken into account. However, in nitrogenase the H^+/e^- flow must be assumed to continue when the diazene stage has been reached. In order to achieve reaction (8), a scheme can be suggested which is quite similar to Scheme 2. As additional elementary reaction, it contains a H^+ reduction for which chemical precedents at iron–sulfur centers are known.[11]

With regard to the FeMo cofactors, the results reported here are plausibly compatible with and support the recently

proposed concept of FeMoco functioning.[12] According to this concept reduction of nitrogenase for turnover leads to opening the FeMoco cluster. One Fe-S-Fe bridge dissociates and vicinal amino acid donors (from Gln α191 and His α195) and H_2O molecules add such that two unique five-coordinate low-spin Fe^{II} centers result. The (variable) space between these Fe centers can accommodate N_2 as well as its reduction products N_2H_2, N_2H_4, and two NH_3. Scheme 3 depicts the N_2H_2 stage. Like PCy_3 dissociation in **1**, dissociation of the H_2O ligands in Scheme 3 can generate vacant sites at the two unique Fe centers rendering possible D_2 addition and D_2/H^+ exchange. It is pointed out that the labile H_2O binding sites also permit addition of CO or other *non-competitive* N_2 reduction inhibitors.



Scheme 3. Opening the FeMoco when changing from the resting (**I**) to the turnover state (**II**), and catalysis of the D_2/H^+ exchange (**III**).

Experimental Section

Unless noted otherwise, all manipulations were carried out in absolute solvents under nitrogen at room temperature. Reactant gases were dried with Na/K alloy. Reaction rates were estimated by means of the NH/CH intensity ratio of diazene protons and aromatic protons of the 'S_4' ligands. Smaller line widths and greater sensitivity make ^1H NMR spectra better suited for this purpose than ^2H NMR spectra, which, however, were also recorded.

1a, b: Acetic acid (14 mL, 2.80 mmol, 0.2 M in H_2O) was added dropwise to a yellow-green suspension of $K_2N_2C_2O_4$ (560 mg, 2.88 mmol) and [Ru(dmso)-(PCy_3)('S_4')] (552 mg, 0.72 mmol) in THF (20 mL). Gas evolved and the suspension turned into a dark green solution, from which dark green microcrystals precipitated. Removal of the aqueous phase and dropwise addition of MeOH (10 mL) to the THF phase completed the crystallization. The precipitated microcrystals were separated after 1 h, washed with MeOH (10 mL), and recrystallized from CH_2Cl_2/MeOH ($-30\,^\circ C$). Compound **1a** (455 mg, 88 %) crystallizes in diastereomeric pure form at this temperature. Correct elemental analyses. ^1H NMR (269.6 MHz, CD_2Cl_2, $-30\,^\circ C$): $\delta = 14.15$ (s, 1 H, N_2H_2), 7.55 – 6.75 (m, 8 H, C_6H_4), 2.85 – 2.60 (m, 2 H, C_2H_4), 2.10 – 0.85 (m, 35 H, C_2H_4, $P(C_6H_{11})_3$); $^{13}C\{^1H\}$ NMR (67.7 MHz, CD_2Cl_2, $-30\,^\circ C$): $\delta = 158.80$ (d), 158.00, 134.20, 134.00, 132.80, 131.60, 130.80, 130.30, 128.50, 128.20, 122.30, 121.10 (C_6H_4), 44.80 (d), 38.30 (C_2H_4), 37.60, 30.00, 28.30 (d), 26.70 [$P(C_6H_{11})_3$]; $^{31}P\{^1H\}$ NMR (109.38 MHz, CD_2Cl_2, $-30\,^\circ C$): $\delta = 28.00$ (s); UV/Vis (CH_2Cl_2): λ_{max} (nm) ($\varepsilon \times 10^{-4}$) = 500 (0.67), 630 (0.40). Compound **1b**: Using CH_3COOD/D_2O and CH_3OD and recrystallization from CH_2Cl_2/MeOD ($-30\,^\circ C$) yielded **1b** in an analogous way.

D_2/NH exchange of **1a** at standard pressure: NMR spectra were recorded of a solution of **1a** in CH_2Cl_2 that was stirred under D_2. The N_2D_2 signals of **1b** were observed after 48 h. H_2/ND exchange of **1b**: ^1H NMR spectra were recorded of saturated solutions of **1b** in CD_2Cl_2 which were treated with H_2 under 1 bar, 35 bar, and 120 bar. The reaction with H_2 at 35 bar was monitored by using a high-pressure NMR tube purchased from Firma Wilmad (528-PV-1, inner diameter 2.2 mm). H^+/D^+ exchange of **1b**: An approximate 70-fold excess of H_2O was added to a saturated solution of **1b** in CD_2Cl_2. NMR spectra were recorded after 25 h.

Received: February 12, 1999 [Z13026IE]
German version: *Angew. Chem.* **1999**, *111*, 2142 – 2145

Keywords: homogeneous catalysis · nitrogen fixation · nitrogenases · reaction mechanisms · S ligands

[1] a) B. K. Burgess, D. J. Lowe, *Chem. Rev.* **1996**, *96*, 2983 – 3011; b) R. R. Eady, *Chem. Rev.* **1996**, *96*, 3013 – 3030.
[2] J. B. Howard, D. C. Rees, *Chem. Rev.* **1996**, *96*, 2965 – 2982.
[3] G. E. Hoch, K. C. Schneider, R. H. Burris, *Biochim. Biophys. Acta* **1960**, *37*, 273 – 279.
[4] B. K. Burgess, S. Wherland, W. E. Newton, E. I. Stiefel, *Biochemistry* **1981**, *20*, 5140 – 5146.
[5] a) C. J. Pickett, *J. Biol. Inorg. Chem.* **1996**, *1*, 601 – 606; b) R. N. F. Thorneley, D. J. Lowe in *Molybdenum Enzymes* (Ed.: T. G. Spiro), Wiley, New York, **1985**, pp. 221 – 286; c) G. J. Leigh, *Eur. J. Biochem.* **1995**, *229*, 14 – 20; d) D. J. Lowe, R. N. F. Thorneley, *Biochem. J.* **1984**, *224*, 887 – 894.
[6] D. Sellmann, A. Hennige, *Angew. Chem.* **1997**, *108*, 270 – 271; *Angew. Chem. Int. Ed. Engl.* **1997**, *36*, 276 – 278.
[7] a) D. Sellmann, T. Gottschalk-Gaudig, F. W. Heinemann, F. Knoch, *Chem. Ber.* **1997**, *130*, 571 – 579; b) T. Gottschalk-Gaudig, Dissertation, Universität Erlangen-Nürnberg, **1997**, pp. 82 – 88.
[8] a) D. Sellmann, E. Böhlen, M. Waeber, G. Huttner, L. Zsolnai, *Angew. Chem.* **1985**, *97*, 984 – 985; *Angew. Chem. Int. Ed. Engl.* **1985**, *24*, 981 – 982; b) D. Sellmann, H. Friedrich, F. Knoch, M. Moll, *Z. Naturforsch. B* **1993**, *48*, 76 – 88.
[9] D. Sellmann, A. Hennige, F. W. Heinemann, *Inorg. Chim. Acta* **1998**, *280*, 39 – 49.
[10] a) D. Sellmann, G. H. Rackelmann, F. W. Heinemann, *Chem. Eur. J.* **1997**, *3*, 2071 – 2080; b) D. Sellmann, T. Gottschalk-Gaudig, F. W. Heinemann, *Inorg. Chem.* **1998**, *37*, 3982 – 3988, and references therein.
[11] D. Sellmann, M. Geck, M. Moll, *J. Am. Chem. Soc.* **1991**, *113*, 5259 – 5264.
[12] D. Sellmann, J. Sutter, *Acc. Chem. Res.* **1997**, *30*, 460 – 469, and references therein.

 © WILEY-VCH Verlag GmbH, D-69451 Weinheim, 1999 1433-7851/99/3813-2026 $ 17.50+.50/0