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On the coupled oxidation-reduction mechanism of molecular nitrogen fixation

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A new mechanism for the catalytic reduction of N_2 was proposed. According to the mechanism, reduction is preceded by the oxidation step with the formation of N_2O. The mechanism allows the participation of weaker reducing agents than those in purely reductive processes. Probable individual steps are considered, in particular, the oxygen atom transfer from the superoxide radical anion $O_2^{-\cdot}$ in a cyclic complex containing the N_2 molecule in the coordination sphere of a metal. The proposed mechanism can explain N_2 reduction involving recently discovered nitrogenase in which $O_2^{-\cdot}$ acts as an electron donor and N_2 reduction in purely chemical systems including the air nitrogen and relatively weak reducing agents.

Key words: nitrogen fixation, oxidative activation, nitrous oxide, nitrogenase.

The well studied biological fixation of dinitrogen and its known chemical analogs involving transition metal complexes are performed as purely reductive reactions.[1] In these processes, the N_2 molecule is activated in the composition of a polynuclear complex, and its further reduction by an external electron donor occurs as a multielectron process with the simultaneous cleavage of both π-bonds resulting in a hydrazine derivative and then the N—N bond cleavage to form ammonium (involving protons of a medium).

Thus, the natural process and model chemical systems avoid the thermodynamic limitations for the successive cleavage of each of three bonds in the N_2 molecule (in one- or two-electron reductive reactions). These limitations to be surmounted require very strong reducing agents. However, sufficiently strong reducing agents are used even for multielectron reduction in natural systems, and hydrogen evolution is coupled with the reduction of N_2. ATP is hydrolyzed simultaneously with the electron transfer (two ATP molecules per each transferred electron), which enhances, evidently, the reductive properties of the electron donor. The reaction equation can be written as follows:

$$N_2 + 8\ e + 8\ H^+ + 16\ ATP \rightarrow$$
$$\rightarrow 2\ NH_3 + H_2 + 16\ ADP + 16\ P.$$

The N_2 molecule is anaerobically reduced; dioxygen inhibits the process decomposing the catalyst.

In addition, the coupled redox process of N_2 fixation is possible in which the N_2 molecule primarily reacts with the oxygen donor to give N_2O, and the latter is reduced then to afford NH_3 and H_2O. Although the N—N bond lengths in the N_2O and N_2 molecules differ slightly, the dissociation energy of this bond in the N_2O molecule (114 kcal mol^{-1})[2] is twice as low as that of the N_2 molecule (225 kcal mol^{-1}). This allows much weaker reducing agents to be used.

It is substantial that in this process usual two-electron reducing agents, *e.g.*, donors of H^-, are thermodynamically capable of reducing the N_2O molecule to NH_3 and H_2O with the intermediate formation of such molecules as $H_2N—NO$. This conclusion follows from the calculation of the energy (see below) in the two-electron reduction

$$N_2O + H_2 \rightarrow H_2N_2O.$$

Note that in the early period of studying biological nitrogen fixation when the oxidative and reductive activation mechanisms were considered as equiprobable[3] some authors also assumed a possibility of ammonia formation through intermediate hydrogen-oxygen compounds of nitrogen.

It cannot be excluded that this is precisely the coupled mechanism which takes place for the preparation of nitrogen-containing organic compounds from the pyrocatechol derivative and N_2 on the silica gel surface containing Ti and Mn admixtures. In this reaction observed previously,[4] the air oxygen turned out to be a necessary participant: the reaction does not occur with pure N_2.* Taking into account that molecular nitrogen readily reacts with both the singlet O atom (to form N_2O)[7] and carbenes[8] (to form diazo compounds), we may assume that a metal oxo complex participates in the elementary step of the O atom transfer. The reverse

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* The formation of N_2O by the oxidation of N_2 by hydrogen peroxide has been reported previously.[5] The catalytic synthesis of HNO_3 from air and water was also observed.[6]

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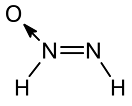
transfer of the O atom from the N_2O molecule to the metal center is well known and occurs, in the case of the ruthenium complex, in a solution at room temperature.[9] This example shows that when the bond energy of the O atom with the metal in the oxo complex does not exceed the O—N bond energy in the N_2O molecule (39 kcal mol^{-1}), which is, most likely, quite possible for oxo complexes of high-valence metals, then we can expect the formation of N_2O with a moderate activation energy. The mechanism of N_2 oxidation proposed by us is known for related molecules with the strong triple bond: acetonitrile (with the formation of acetonitrile *N*-oxide)[10] and CO (with the formation of CO_2).[11]

Although the reduction of N_2O to NH_3 and H_2O is thermodynamically facilitated as compared to the corresponding reactions of molecular nitrogen, it can meet certain difficulties. It is necessary to prevent the formation of water and N_2, which usually occurs when N_2O acts as a two-electron oxidant. Published data are contradictory. For example, it has previously[12] been established that N_2O is reduced almost quantitatively to NH_3 and H_2O in acidic solutions of $TiCl_3$. However, later the authors[13] observed only N_2 and H_2O under similar conditions. Perhaps, to prevent the primary formation of N_2 and H_2O, one has to coordinate N_2O on a polynuclear complex in which the eight-electron reduction of N_2O to NH_3 and H_2O is facilitated.

With the purpose for analyzing the reactivity of N_2O in more detail, we theoretically studied the model reactions involving N_2O by the density functional method in the B3LYP version using the GAUSSIAN-98 program.[14] For isomers of N_2H_2O (**1—3**), calculations were performed in the 6-311++G(d,p) basis set. For other cases, the total energy was calculated in the same basis set using the geometry of stationary points, and the zero-point energies were calculated at the B3LYP/G-31G level. Comparison of our geometric parameters of the H_2N_2O (**3**) molecule (Table 1) with previously published data[15] calculated at the MP2/6-31G* level shows that the density functional method possesses a sufficient accuracy.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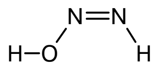
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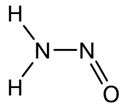
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Among probable isomers of H_2N_2O, structure **3** turned out to be most stable. The thermal effect of nitrosylamide formation from N_2O and H_2 calculated taking into account the contribution of the zero-point energy turned out to be close to zero, unlike the negative thermal effect of the hydrogenation of N_2 to *trans*-diimide (−50 kcal mol^{-1}) and hydrazine (−12 kcal mol^{-1}).[2]

The activation energy of the thermoneutral reaction of the O atom transfer

$$N_2O + N_2 = N_2 + ON_2$$

is 63 kcal mol^{-1} and exceeds much the bond energy of N—O. For the thermodynamically favorable oxidation of CO

$$N_2O + CO = N_2 + CO_2$$

the energy barrier also remains high (47 kcal mol^{-1}). At the same time, on going to the cationic species N_2O^+, the barrier of the O^+ transfer decreases to 24 kcal mol^{-1}, despite the increase in the energy of the N—O^+ bond to 50 kcal mol^{-1}.*

The negatively charged N_2O^- species can exist in two equilibrium structures with very close energies. In one of the structures, the N—O bond is considerably loosened, *viz.*, to 2.3 Å. However, the nonlinear geometry (the N—N—O angle is equal to 120°) and noticeable charge (−0.14 e) on the far N atom, as well as the N_2—O^- bond energy (10 kcal mol^{-1}), indicate that the structure of N_2O^- cannot be explained by purely polarization N_2—O^- interactions, which are highest for the orientation of the longitudinal polarizability of N_2 along the field. The O^- species possesses the π-donating orbitals, which are necessary, as known, for the formation of nitrogen complexes of transition metals. However, in addition to the π-donating properties, O^- possesses the noticeable σ-donating properties, which are highest in the case of the orientation of the singly occupied p_σ-orbital along the N—N axis. The latter is unfavorable for the formation of such complexes. It is most likely that this is the reason for which the N_2 molecules deviate from the linear coordination, which is optimum for the simultaneous transfer of the electron density to the N_2 molecule *via* two π-channels. Thus, O^- can be considered as an isolobal analog of the transition metal atom. This viewpoint is confirmed by the fact that interaction with the second N_2 molecule does not lead to the O^- transfer but the $[N_2ON_2]^-$ complex is formed. In the latter, the bond energy of the second N_2 molecule (8 kcal mol^{-1}) and geometric characteristics of its coordination are close to the corresponding values for the first N_2 molecule.

This result shows that the main difficulty for the multielectron reduction of N_2O to NH_3 is the one-electron transfer, which provides a possibility for the real cleavage of the N—O bond, and conditions for the also thermodynamically favorable reduction of N_2O to N_2 and H_2O are thus created. Therefore, we may assume that the N—N bond must necessarily be involved in the primary processes of deep N_2O reduction. The most probable pathways are the mono- or polydentate

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* A similar increase in the reactivity with increasing the strength of the cleaved bond has previously[16] been observed for thermoneutral reactions of the transfer of O and O^+ to the H_2O molecule from H_2OO and H_2OO^+, respectively.

Table 1. Bond lengths (*d*), bond angles (ω), and energy parameters of the calculated structures[a]

System[b]	Term (symmetry)	Bond	d/Å	Bond angle	ω/deg	$-E^c$	ZPE^d
						a.u.	
1	$^1A'$ (C_s)	N—N	1.267	N—N—O	126	185.8791	0.0344
		N—H(1)	1.041	N—N—H(1)	106		
		N—H(2)	1.023	N—N—H(2)	119		
		N—O	1.223				
2	$^1A'$ (C_s)	N—N	1.232	N—N—O	112	185.9002	0.0335
		N—H	1.025	N—N—H	106		
		N—O	1.362	N—O—H	106		
		O—H	0.979				
3	$^1A'$ (C_s)	N—N	1.333	N—N—O	114	185.9035	0.0323
		N—H(1)	1.007	N—N—H(1)	117		
		N—H(2)	1.018	N—N—H(2)	119		
		N—O	1.213				
N_2O +	$^1\Sigma^+$	N—N	1.126			185.8929	0.0212
+ H_2	$^1\Sigma^+$	N—O	1.183				
		H—H	0.744				
N_2—O—N_2 (TS)	^1A (C_2)	N—N	1.123	N—N—O	164	294.1714	0.0142
		N—O	1.722	N—O—N	170		
N_2—O—CO (TS)	$^1A'$ (C_s)	N—N	1.132	N—N—O	159	297.9879	0.0141
		N—O	1.522	N—O—C	151		
		O—C	1.790	O—C—O	136		
		C—O	1.163				
N_2—O^+—N_2 (TS)	^2A (C_2)	N—N	1.119	N—N—O	178	293.7573	0.0131
		N—O	1.768	N—O—N	153		
N_2O^+	$^2\Pi$ ($C_{\infty v}$)	N—N	1.150	N—N—O	180	184.2373	0.0082
		N—O	1.251				
N_2O^-	$^2A'$ (C_s)	N—N	1.149	N—N—O	115	184.7103	0.0058
		N—O	2.236				
	$^2A'$ (C_s)	N—N	1.219	N—N—O	133	184.7191	0.0070
		N—O	1.369				
N_2O^- (TS)	$^2A'$ (C_s)	N—N	1.156	N—N—O	129	184.7062	0.0055
		N—O	1.880				
$(N_2)O^-(N_2)$	^2B (C_2)	N—N	1.137	N—N—O	114	294.2730	0.0123
		N—O	2.345	N—O—N	168		
HNO	$^1A'$ (C_s)	H—N	1.170	H—N—O	127	130.4912	0.0104
		N—O	1.250				
NOH	$^3A''$ (C_s)	H—O	0.986	H—O—N	112	130.4811	0.0130
		O—N	1.372				
	$^1A'$ (C_s)	H—O	1.012	H—O—N	114	130.4485	0.0127
		O—N	1.302				
cis-HONNOH	$^1A'$ (C_s)	H(1)—O(1)	0.994	O(1)—N—N	119	261.1120	0.0351
		H(1)—O(2)	1.902	N—N—O(2)	112		
		O(1)—N	1.420	O(1)—H(1)—O(2)	115		
		N—N	1.247				
		N—O(2)	1.502				
		O(2)—H(2)	0.977				
		O(2)—H(1)	1.902				
cis-HONNOH	$^1A_1$ (C_{2v})	H—O	0.982	H—O—N	102	261.1127	0.0356
		O—N	1.459	O—N—N	116		
		N—N	1.238	N—N—O—H	180		
cis-HONNOH	$^1A_1$ (C_{2v})	H—O	1.005	H—O—N	107	261.1023	0.0375
		O—N	1.396	O—N—N	110		
		N—N	1.299	N—N—O—H	0		
trans-HONNOH	^1A_g (C_{2h})	H—O	0.994	H—O—N	108	261.1080	0.0351
		O—N	1.416	O—N—N	111		
		N—N	1.257	N—N—O—H	0		

(*to be continued*)

Table 1 (*continued*)

System[b]	Term (symmetry)	Bond	d/Å	Bond angle	ω/deg	$-E^c$	ZPE^d
						a.u.	
cis-HONN(H)O	$^1A'$ (C_s)	H—O(1)	1.006	O(1)—N—N	110	261.1023	0.0375
		O(1)—N	1.396	O(2)—N—N	126		
		H—N	1.020	H—N—N	115		
		O(2)—N	1.309				
		N—N	1.299				
trans-O(H)NN(H)O	^1A_g (C_{2h})	H—N	1.026	H—N—N	113	261.0739	0.0377
		O—N	1.296	O—N—N	124		
		N—N	1.316				
cis-O(H)NN(H)O	$^1A_1$ (C_{2v})	H—N	1.026	H—N—N	114	261.0692	0.0376
		O—N	1.284	O—N—N	124		
		N—N	1.326				
cis-$HONNO^-$	$^1A'$ (C_s)	H—O(1)	1.015	O(1)—N—N	111	260.5703	0.0228
		H—O(2)	1.736	N—N—O(2)	118		
		O(1)—N	1.521				
		N—N	1.289				
		N—O(2)	1.359				
trans-$HONNO^-$	$^1A'$ (C_s)	H—O(1)	0.995	O(1)—N—N	105	260.5544	0.0223
		O(1)—N	1.522	N—N—O(2)	117		
		N—N	1.300				
		O(2)—N	1.325				
cis-$N_2O_2^{2-}$	$^1A_1$ (C_{2v})	N—N	1.318	O—N—N	122	259.7730	0.0108
		N—O	1.411				
$N_2O_2^-$	$^2A''$ (C_s)	N—N	1.136	O—N—N	93	259.9580	0.0095
		N—O	2.575	O—O—N	87		
		O—O	1.371				
$MgO_2(N_2)$	$^1A'$ (C_s)	Mg—O	1.929	O—O—N	111	460.0004	0.0118
		O—O	1.417	O—N—N	111		
		O—N	2.117	O—Mg—N	94		
		N—N	1.150				
		Mg—N	2.213				
$MgOON_2$ (TS)	$^1A'$ (C_s)	Mg—O	1.830	O—O—N	107	459.9587	0.0125
		O—O	2.259	O—N—N	92		
		Mg—N	2.150	O—Mg—N	105		
		N=N	1.204				
		N—O	1.250				
$Mg(O)N_2O$	$^1A'$ (C_s)	Mg—O	1.780	O—Mg—N	177	459.9860	0.0125
		Mg—N	2.094				
		N=N	1.155				
		N—O	1.213				
$Mg(ON_2O)$	$^1A_1$ (C_{2v})	Mg—O	1.897	O—N—N	120	460.0294	0.0133
		N=N	1.274				
		N—O	1.456				
MgO + + N_2O	$^1\Sigma^+$	Mg—O	1.774			459.9749	0.0130

[a] The total energy and zero-point energy (in parentheses) of the molecules are the following (a.u.): N_2O ($^1\Sigma^+$) −184.7147 (0.0112), N_2 ($^1\Sigma_g^+$) −109.5583 (0.0054), CO ($^1\Sigma^+$) −113.3473 (0.0047), H_2O ($^1A_1$) −76.4579 (0.0205), NO^- ($^3\Sigma^-$) −129.9248 (0.0030), NO ($^2\Pi$) −129.9295 (0.0042), O_2 ($^3\Sigma_g^-$) −150.3673 (0.0032), O_2^- ($^2\Pi_g$) −150.3889 (0.0024).

[b] TS is the transition state.

[c] Total energy.

[d] Zero-point energy.

coordination of N_2O at one or both N atoms or the direct cleavage of the N—N bond in the hydrated form of N_2O, *viz.*, hyponitrous acid HON=NOH or its derivatives. A possibility of the first route is indicated by the found[17] cleavage of the N—N bond in the N_2O molecule upon its interaction with a tricoordinate Mo^{III} complex. The consideration of the second route is based on the influence of the heterolytic dissociation of the OH groups in the $H_2N_2O_2$ molecule on the strength of the N—N bond. If the decomposition

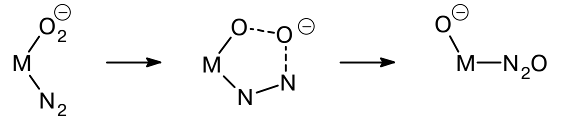
$$H_2N_2O_2 \rightarrow 2\ HNO$$

leads to energy expense of 50 kcal mol^{-1} (reverse reaction of HNO dimerization is the main channel of N_2O

formation by H_2NOH oxidation), the dissociation energy of $HONNO^-$ to HNO and NO^- is already 20 kcal mol^{-1}, and the $N_2O_2^{2-}$ dianion is a metastable species: its decomposition to 2 NO^- is thermodynamically favorable (+30 kcal mol^{-1}). Solvation must stabilize $N_2O_2^{2-}$. However, we can expect that, under appropriate conditions, *e.g.*, in the presence of sites with a high affinity to NO^-, the reaction of the hydrated and ionized forms of N_2O results in the N—N bond cleavage.

The radical anion $O_2^{-\cdot}$ is an oxidant but simultaneously possesses reductive properties (the redox potential of $O_2^{-\cdot}/O_2$ is equal**[18]** to −0.56 V). Therefore, the coordination of $O_2^{-\cdot}$ can result in the enhancement of the donating properties of the central metal atom and, correspondingly, the enhancement of its affinity to form complexes with N_2. In this case, compounds of the $LM(O_2^-)(N_2)$ type should preferentially be obtained because the coordination of the second $O_2^{-\cdot}$ molecule instead of N_2 is less probable due to the Coulomb repulsion. It is reasonable to question about the inner-sphere oxidation of N_2 to N_2O (Scheme 1).

Scheme 1



This reaction is facilitated by the high M—O bond energy. A decrease in the activation barrier of N_2O formation is also possible due to the negative charge on the $O_2^{-\cdot}$ ligand. As an example, we examined the reaction

$$Mg(O_2)(N_2) \rightarrow MgO(N_2O)$$

involving Mg^+ as a coordination site. The system in the initial state represents the MgO_2 peroxo complex with weak Mg—N_2 interaction, the Mg—N distance being 2.21 Å. The O—N distance (2.12 Å) is shorter than that in the $(O_2^-)(N_2)$ complex (2.58 Å). The transition state is higher in energy by 25.6 kcal mol^{-1} and has the late character: the O—O and O—N bond lengths are 2.26 and 1.25 Å, respectively. N_2O is linearly coordinated in the molecule of reaction product: the Mg—N distance equals 2.09 Å, and the Mg—N_2O bond energy is 7 kcal mol^{-1}.

Our study shows grounds to consider the redox process of N_2 fixation to occur without strong oxidants and reducing agents. Probably, this reaction occurs only under conditions of the enhanced organization of the active site. As known, the purely biological mechanism of N_2 fixation also requires a complicated organized active site, Fe—Mo cofactor, to be involved. Perhaps, a wide abundance of reactions of this type is related to their early appearance, still before the epoch of oxygen formation on the Earth due to photosynthesis. In addition, it is more difficult to perform the coupled process because it needs N_2 to be oxidized in the reductive environment.

Nitrogenase of the new type recently discovered**[19]** in *Streptomyces thermoautotrophicus* bacteria attracts attention in connection with the problem discussed. Neither O_2 nor H_2O_2 impede its functioning. Even *vice versa*, O_2 is needed to be involved to transform N_2 into NH_3 because $O_2^{-\cdot}$ formed in the special protein, which is bound in a complex with dinitrogenase, is the electron donor for N_2 according to the authors's**[19]** opinion. Judging from all properties, requirements to the strength of the reducing agent for N_2 in the new nitrogenase are noticeably weaker compared to the anaerobic nitrogenase because a polynuclear complex is not likely used for the activation of N_2, and an approximately halved number of ATP molecules is consumed for the transfer of one electron.

It can be assumed that the coupled new mechanism involving O_2 takes place precisely in the case of the new nitrogenase. The only fact, which does not completely agree with this, is that N_2 can be reduced without O_2 if an artificial donor dithionite is used as a reducing agent.**[19]** Perhaps, this is related to the presence of residual O_2 (it was not specially removed from the system) capable of assisting the reaction with N_2: the activity of the new nitrogenase is at least by an order of magnitude lower than the activity of the traditionally studied purely reductive enzyme. This explanation can be verified by reduction with dithionite in the presence of superoxide dismutase, which suppresses the enzyme activity during the anaerobic fixation of N_2.

Thus, based on analysis performed for the model systems, we proposed the new mechanism of nitrogen fixation, which is possibly accomplished also in the nitrogenase of the new type.**[19]**

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References

1. A. E. Shilov, *Metal Complexes in Biomimetic Chemical Reactions*, CRC Press, Boca Raton, New York, 1997, 534 pp.
2. *Molekulyarnye postoyannye neorganicheskikh soedinenii* [*Molecular Constants of Inorganic Compounds*], Ed. K. S. Krasnov, Khimiya, Leningrad, 1979, 446 pp. (in Russian).
3. V. I. Lyubimov, *Biokhimiya fiksatsii molekulyarnogo azota* [*Biochemistry of Molecular Nitrogen Fixation*], Nauka, Moscow, 1969, p. 28 (in Russian).
4. V. B. Vol'eva, I. S. Belostotskaya, A. Yu. Bundeev, N. L. Komissarova, and V. V. Ershov, *Izv. Akad. Nauk, Ser. Khim.*, 1997, 398 [*Russ. Chem. Bull.*, 1997, **46**, 385 (Engl. Transl.)].
5. T. M. Nagiev, *Usp. Khim.*, 1985, **54**, 1654 [*Russ. Chem. Rev.*, 1985, **54** (Engl. Transl.)].

6. M. M. Karavaev and V. A. Matyshak, *Khim. Prom-st´* [*Chemical Industry*], 1998, 814 (in Russian).
7. A. D. Katakis and H. Taube, *J. Chem. Phys.,* 1962, **36**, 416.
8. A. E. Shilov, A. A. Shteinman, and M. B. Tjabin, *Tetrahedron Lett.,* 1968, 4177.
9. J. T. Groves and J. S. Roman, *J. Am. Chem. Soc.*, 1995, **117**, 5594.
10. G. Bucher and J. C. Scaiano, *J. Phys. Chem.*, 1994, **98**, 12471.
11. H. Dobber, L. Gremer, O. Meyer, and R. Hurbert, *Proc. Nat. Acad. Sci. USA*, 1999, **96**, 8884.
12. M. L. Nichols and I. A. Derbigny, *J. Phys. Chem.*, 1926, **30**, 491.
13. R. G. S. Banks, R. J. Henderson, and J. M. Pratt, *J. Chem. Soc., A*, 1968, 2886.
14. M. J. Frisch, G. W. Trucks, M. Head-Gordon, P. M. W. Gill, M. W. Wong, J. B. Foresman, B. G. Johnson, H. B. Schlegel, M. A. Robb, E. S. Replogle, R. Gomperts, J. L. Andres, K. Raghavachari, J. S. Binkley, C. Gonzalez, R. L. Martin, D. J. Fox, D. J. Defrees, J. Baker, J. J. P. Stewart, and J. A. Pople, *GAUSSIAN-98*, *Revision A.6*, Gaussian, Inc., Pittsburgh (PA), 1998.
15. M. Aschi and F. Grandinetti, *Chem. Phys. Lett.*, 1997, **267**, 98.
16. G. V. Lyubimova and A. F. Shestakov, *Izv. Akad. Nauk, Ser. Khim.,* 2000, 981 [*Russ. Chem. Bull.*, *Int. Ed.*, 2000, **49**, 976].
17. D. Dobos, *Electrochemical Data*, Académiai Kiadó, Budapest, 1978.
18. C. E. Laplaza, A. L. Odom, W. M. Davis, C. C. Cummins, and J. D. Protasiewicz, *J. Am. Chem. Soc*, 1995, **117**, 4999.
19. M. Ribbe, D. Gadkari, and O. Meyer, *J. Biol. Chem.*, 1997, **272**, 26627.

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