The Vibrational Spectrum of H_2O_3

A. Engdahl and B. Nelander

This report presents positive infrared spectroscopic identification of H_2O_3, a higher oxide of hydrogen of importance for the understanding of the chain formation ability of atomic oxygen and a possible intermediate in hydrogen oxygen radical chemistry. All fundamental vibrations of H_2O_3, isolated in an argon matrix, have been observed. In addition, several bands of HDO_3, D_2O_3, and $H_2{}^{16}O_2{}^{18}O$ have been measured. One particular mode, the antisymetric O–O stretch at 776 cm^{-1}, should be observable even in the presence of high water concentrations.

The ability of sulfur atoms to form long chains terminated by hydrogens or alkyl groups is well known. Calculations have suggested that oxygen may form similar chain compounds (*1*). Hydrogen peroxide (HOOH) has been known since 1818. Giguère (*2*) and others (*3*) obtained the first evidence for the existence of HOOOH from the vibration spectra of condensed mixtures of H_2O, HOOH, and O_2, which had passed through an electrical discharge. Nuclear magnetic resonance (NMR) investigations have shown that HOOOH is formed when ozone (O_3) reacts with isopropyl alcohol (*4*). Very recently, HOOOH has been implicated as an intermediate in the antibody-catalyzed addition of singlet molecular oxygen to water (*5*). NMR results (*6*), mass spectral data (*7*), and ab initio calculations (*8–10*) indicate that HOOOH is more stable than was previously expected and could even form when OH and HOO react at low temperatures and total pressures that are not too low [for a discussion see (*11–13*)]. Vibration spectral data would make it possible to verify the presence of HOOOH in a gaseous mixture. Unfortunately, the data of Giguère were obtained in a strongly perturbing medium. We have recently found that we can obtain significant concentrations of HOOOH in argon matrices by the photolysis of the ozone–hydrogen peroxide complex. In these experiments, we have been able to observe all fundamentals of HOOOH and several fundamentals of HOOOD, DOOOD, and $H^{18}O^{16}O^{16}OH$.

The experiments were carried out with conventional matrix isolation equipment (*14*). Spectra were recorded on a Bruker 113v FTIR spectrometer. A liquid He–cooled germanium bolometer was used for the far-infrared spectra (*15*). The handling of hydrogen peroxide and the preparation of DOOD have been described in (*16*). O_3 was prepared as in (*14*). The quadrupled output from a Continuum NY 81-20C YAG laser was used for the irradiations.

When O_3 and HOOH are co-deposited in argon matrices, a fraction of the molecules form a complex, which is revealed by the appearance of new bands that are close to but separated from those of the parent molecules (*17*). Less than 5 min of irradiation with 266-nm radiation completely eliminates this complex. At the same time, strong bands grow in the water fundamental regions. In addition, a set of relatively weak but sharp and easily observed bands grow together and are eventually eliminated by a prolonged irradiation. When O_3 is irradiated at 266 nm, the dominant products are O (^1D) and O_2 ($^1\Delta$). O(^1D) is known to insert into bonds, as exemplified by the formation of HOCl when the HCl ozone complex is irradiated (*18*). In order to identify the new compound, we performed experiments with DOOD and HOOD, which clearly show that the new compound contains two H atoms (Fig. 1). This is also evident from the appearance of one HOO bend from HOOD–O_3 but two HOO bends with HOOH–O_3 (Table 1). The presence of two torsion bands and two OO stretches shows that the new compound contains two OO bonds and thus identifies the new compound as H_2O_3. In order to find the position of the inserting O atom, we performed an experiment with $^{18}O_3$ and H_2O_2. The results are compared in Table 1 with calculated isotope shifts for $H^{18}O^{16}O^{16}OH$ and for $H^{16}O^{18}O^{16}OH$. [A diagonal, symmetry-adapted force field, including an interaction constant between the symmetric O–O stretch and the OOO bend, was fitted to the observed H_2O_3 spectrum and used to predict the effects of ^{18}O substitution. The most stable structure

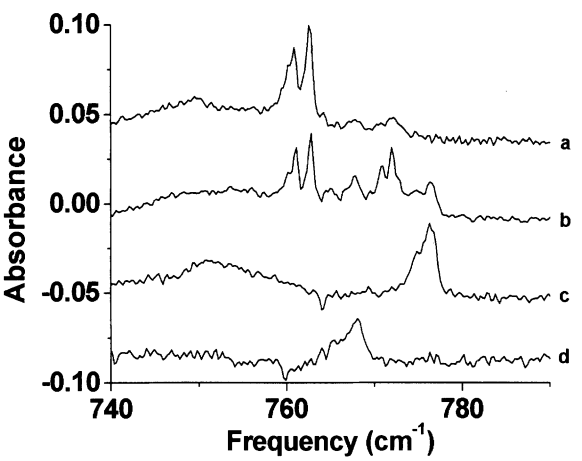

Fig. 1. The antisymmetric OO stretch of HOOOH in solid argon. (**a**) DOOOD. (**b**) A mixture of HOOOH, HOOOD, and DOOOD. (**c**) HOOOH. (**d**) $H^{18}O^{16}O^{16}OH$. The curves have been offset vertically for clarity.

Table 1. Ab initio–calculated (MP4 6-31G**), observed (obs), and calculated (calc) fundamentals of HOOOH, DOOOD, HOOOD, $H^{18}O^{16}O^{16}OH$ (as ^{18}O), and $H^{16}O^{18}O^{16}OH$ (sym ^{18}O) in wavenumbers (cm^{-1}). The modes are ν_1, symmetric OH stretch; ν_2, symmetric HOO bend; ν_3, symmetric OO stretch; ν_4, OOO bend; ν_5, symmetric torsional rotation; ν_6, antisymmetric OH stretch; ν_7, antisymmetric HOO bend; ν_8, antisymmetric OO stretch; and ν_9, antisymmetric torsional rotation.

Mode	Jackels (*8*)	H_2O_3 (obs)	H_2O_3 (calc)	D_2O_3 (obs)	HDO_3 (obs)	$H_2O_2{}^{18}O$ as ^{18}O		Sym ^{18}O (calc)
						obs	calc	
ν_1	3785	3529.6	3529.6	2610.4	2610.4	3520.3	3517.8	3529.6
ν_2	1383	1347.4	1347.6			1344.3	1341.9	1347.6
ν_3	868	821.0	820.4		814.6		813.5	790.0
ν_4	520	509.1	509.2				495.8	508.9
ν_5	364	346.4	345.9	273.5		346.0	345.5	345.5
ν_6	3781	3529.6	3529.6	2610.4	3529.6	3529.6	3529.6	3529.6
ν_7	1387	1359.1	1359.2	1007.3	1349.9	1357.0	1356.2	1355.0
ν_8	783	776.3	776.1	762.6	772.0	768.0	767.2	751.1
ν_9	417	387.0	387.0	301.6	369.2	386.6	385.7	385.5

Chemical Physics, Chemical Center, Post Office Box 124, S-22100 Lund, Sweden.

of (*8*) was used.] The O atom is inserted into the OH bond of HOOH. Comparison with an ab initio calculation gives agreement that is as good as can be expected, considering that the calculated frequencies are harmonic frequencies, whereas the measured ones are the $1 \leftarrow 0$ transitions. Some of the H_2O_3 bands appear as doublets (Fig. 1). We do not know if this is due to the presence of two different trapping sites or to tunneling between the two torsion minima, as has been observed for HOOH (*19*).

The strongest bands of H_2O_3 are the torsions, the OH stretches, and the antisymmetric HOO bends. All of these bands are found in regions where the water absorption is very strong, which will make them difficult to observe. The antisymmetric OO stretch, which we observed at 776 cm^{-1}, is approximately half as strong as the OH stretches, but because it is found in a region that is relatively free from atmospheric absorptions, it may be used to detect the presence of H_2O_3.

References and Notes

1. D. J. McKay, J. S. Wright, *J. Am. Chem. Soc.* **120**, 1003 (1998).
2. J. L. Arnau, P. A. Giguère, *J. Chem. Phys.* **60**, 270 (1974).
3. M. Horváth, L. Bilitzky, J Hüttner, *Ozone* (Elsevier, Amsterdam, Netherlands, 1985), p. 47.
4. B. Plesnicar, J. Cerkovnik, T. Tekavec, J. Koller, *Chem. Eur. J.* **6**, 809 (2000).
5. P. Wentworth Jr. *et al.*, *Science* **293**, 1806 (2001).
6. J. Cerkovnik, B. Plesnicar, *J. Am. Chem. Soc.* **115**, 12169 (1993).
7. M. Speranza, *Inorg. Chem.* **35**, 6140 (1996).
8. C. F. Jackels, *J. Chem. Phys.* **99**, 5768 (1993).
9. M. A. Vincent, I. A. Hillier, *J. Phys. Chem.* **99**, 3109 (1995).
10. T. H. Lay, J. W. Bozzelli, *J. Phys. Chem.* **101**, 9505 (1997).
11. W. B. DeMore, *J. Phys. Chem.* **86**, 121 (1982).
12. D. W. Toohey, J. G. Anderson, *J. Phys. Chem.* **93**, 1049 (1989).
13. C. Gonzalez *et al.*, *J. Phys. Chem.* **95**, 6784 (1991).
14. B. Nelander, A. Engdahl, T. Svensson, *Chem. Phys. Lett.* **332**, 403 (2000).
15. A. Engdahl, B. Nelander, *J. Chem. Phys.* **91**, 6604 (1989).
16. ———, G. Karlström, *J. Phys. Chem.* **A105**, 8393 (2001).
17. A. Engdahl, B. Nelander, in preparation.
18. I. Schwager, A. Arkell, *J. Am. Chem. Soc.* **89**, 6006 (1967).
19. M. Pettersson, S. Touminen, M. Räsänen, *J. Phys. Chem.* **A101**, 1166 (1997).

20. Supported by the Swedish Natural Science Research Council, Forskningsrådsnämden, OK miljöstiftelse, and Carl Tryggers Stiftelse.

18 October 2001; accepted 3 December 2001

Electrode-Reducing Microorganisms That Harvest Energy from Marine Sediments

Daniel R. Bond,[1] Dawn E. Holmes,[1] Leonard M. Tender,[2] Derek R. Lovley[1]*

Energy in the form of electricity can be harvested from marine sediments by placing a graphite electrode (the anode) in the anoxic zone and connecting it to a graphite cathode in the overlying aerobic water. We report a specific enrichment of microorganisms of the family Geobacteraceae on energy-harvesting anodes, and we show that these microorganisms can conserve energy to support their growth by oxidizing organic compounds with an electrode serving as the sole electron acceptor. This finding not only provides a method for extracting energy from organic matter, but also suggests a strategy for promoting the bioremediation of organic contaminants in subsurface environments.

The organic matter stored in anoxic subsurface environments and aquatic sediments represents a large potential source of energy. Although some of this organic matter, most notably petroleum, is in a concentrated form that can readily be extracted and used, most of the organic matter cannot be converted to useful energy by current technologies. However, anaerobic microorganisms capable of using organic matter inhabit anoxic subsurface environments and continually tap into this energy reservoir.

It was recently shown that electrical current could be harvested from anoxic marine sediments by embedding an electrode (the anode) into the sediment and connecting it through electronic circuits to a similar electrode in the overlying aerobic seawater (the cathode) (*1*). Even with a simple, unmodified graphite electrode, the magnitude of current produced, ~0.01 W/m^2, was sufficient to theoretically power marine-deployed electronic instrumentation. Killing the microorganisms in the sediments inhibited current flow (*1*).

To further evaluate the potential role of microorganisms in electron transfer to the anode, we constructed sediment batteries in laboratory aquaria in a manner similar to that described in (*1*), with graphite anodes in the anoxic marine sediment and graphite cathodes in overlying aerobic water (*2*). Electrical power output from these batteries was continuous, averaging 0.016 W per square meter of electrode surface area in three independent experiments during 6 months of current harvesting. After this energy-harvesting phase, microbial communities attached to the anodes were compared to communities on identical control electrodes that had been placed in the same sediments for the same length of time but were not electrically connected to the electrode in the overlying water.

Analysis of 16*S* ribosomal DNA (rDNA) genes (*3–6*) demonstrated that there was a pronounced enrichment of microorganisms from the δ-subgroup of the Proteobacteria colonizing energy-harvesting anodes (*7*). Whereas only 17 ± 4.3% (mean ± SD, $n = 3$) of 16*S* rDNA sequences from control electrodes were in the δ-subgroup of the Proteobacteria, 71.3 ± 9.6% (mean ± SD, $n = 3$) of the 16*S* rDNA sequences from microorganisms colonizing anodes of the current-producing batteries were in this subgroup. Furthermore, 70% of the increase in δ-Proteobacterial sequences was due to a single cluster of bacteria in the family Geobacteraceae, a group of anaerobic microorganisms that can couple the oxidation of organic compounds to reduction of insoluble Fe(III) oxides (*8, 9*). The organism in pure culture most closely related to the sequences repeatedly enriched on anodes (*7*) was *Desulfuromonas acetoxidans*, a marine microorganism known to grow anaerobically by oxidizing acetate with concomitant reduction of elemental sulfur (*10*) or Fe(III) (*11*). Enumeration of *Desulfuromonas* 16*S* rDNA sequences by a most-probable-number polymerase chain reaction (MPN-PCR) technique (*12*) revealed that *Desulfuromonas* target sequences on anodes from current-generating batteries were enriched by a factor of 100 relative to those on anodes from control batteries.

To further investigate the specific enrichment of microorganisms on anodes, we inoculated the anoxic side of a two-chambered microbial battery with sediment; the seawater was periodically changed with fresh acetate-amended anoxic seawater (*13*). Replacing the medium diluted the sediment and microorganisms from the inoculum that were not growing, while resupplying acetate, the primary intermediate in the degradation of organic carbon in anoxic sediments (*14*). After 85 days, 16*S* rDNA sequences from bacteria attached to the anode were examined. All of the anode-attached bacteria detected were members of the δ-Pro-

[1]Department of Microbiology, University of Massachusetts, Amherst, MA 01003, USA. [2]Center for Bio/molecular Science and Engineering, Code 6900, Naval Research Laboratory, Washington, DC 20375, USA.

*To whom correspondence should be addressed. E-mail: dlovley@microbio.umass.edu