tions is the same. However, off axis ($\theta_0 > 0$), they are substantially different. To make a true omnidirectional dielectric mirror, one wants to have a broad reflection band that survives for all angles and for both polarizations. The *p*-polarized band was thought to always have a hole or "window" at Brewster's angle, θ^B, where *p*-polarized light propagates from an n_1 to an n_2 layer, and from an n_2 to an n_1 layer without any reflection (*3*). Once light gets into the multilayer film at θ^B— regardless of the frequency—it will propagate unimpeded; there is a hole in the reflection band at that angle. It is somewhat ironic that even Joannopoulos himself once implied that it was impossible to make a one-dimensional (1D) omnidirectional reflector in his own book with Meade and Winn (*4*). Their proof actually showed that there is no omnidirectional photonic band gap in an infinite 1D, periodic, layered structure. However, all physical 1D structures are finite, and this offers the way out.

The real reflection coefficient R_{12} at a n_1 to n_2 interface is identically zero for Brewster's condition, $\theta_1^B = \arctan n_2/n_1$. Now in the figure, we see that the overall incident angle θ_0 has a full domain $[0, \pi/2]$. However, the corresponding range of the refracted angle θ_1 is restricted by Snell's law ($n_0 \sin \theta_0 = n_1 \sin \theta_1$) to the interval $[0, \theta_1^{max}]$, where $\theta_1^{max} \equiv \arcsin n_0/n_1$. If $\theta_1^{max} < \theta_1^B$, then incident light from the outside can never couple to the Brewster window. That is the trick, and it is easy to do by making n_0/n_1 sufficiently large. Now we make sure that n_1 and n_2 are just right so that we run out of angle θ_0 before we run out of bandwidth, and voila, we have the omnidirectional reflector of Fink *et al.* (*1*).

An omnidirectional dielectric reflector—unlike metal—is nearly totally lossless and can be engineered to have extremely high reflectivities. Applications in the optical wavelength range are as solar and thermoelectric power sources and laser microcavities. In the microwave regime, the structure would find uses as an antenna substrate or in high-power microwave sources—where lossy metals just will not do.

References

1. Y. Fink *et al.*, *Science* **282**, 1679 (1998).
2. J. L. Jewel *et al.*, *IEEE J. Quantum Electron.* **27**, 1332 (1991).
3. P. Yeh, *Optical Waves in Layered Media* (Wiley, NY, 1988).
4. J. D. Joannopoulos, R. D. Meade, J. N. Winn, *Photonic Crystals* (Princeton Univ. Press, Princeton, NJ, 1995), chap. 4.

Biological Hydrogen Production: Not So Elementary

Michael W. W. Adams and Edward I. Stiefel

Hydrogenase is the name given to the family of enzymes that catalyze what is seemingly one of the simplest possible chemical reactions, the interconversion of the smallest molecule, hydrogen gas (H_2), and its elementary particle constituents, two protons and two electrons:

$$2H^+ + 2 \leftrightarrows H_2 \qquad (1)$$

Representatives of most prokaryotic genera, as well as a few eukaryotes, metabolize hydrogen gas and contain hydrogenase (*1*). The enzyme was discovered in the 1930s, its requirement for iron was established in the 1950s (*2*), and, in the 1980s, many, but not all, hydrogenases were shown to contain nickel as well as iron (*3*). Nickel-iron varieties are usually found in microorganisms that consume hydrogen, whereas those that typically produce hydrogen contain iron-only enzymes. The nature of the catalytic sites in hydrogenases has been subject to intense study and conjecture (*4*). On page 1853 of this issue, Peters *et al.* report their use of x-ray crystallography to provide the first structural glimpse of the iron-only hydrogenase from the hydrogen-producing, anaerobic bacterium *Clostridium pasteurianum* (*5*). The resolved array of five iron-sulfur clusters includes the very special "H cluster," which almost certainly is the catalytic site. With an unprecedented nuclearity of six, the H cluster contains two strikingly organometallic iron atoms, with metal-carbon bonds supplied by diatomic ligands, thought to be carbon monoxide (CO) or cyanide (CN^-) (or both).

Hydrogen gas, a mainstay of the chemical industry (*6*) and much discussed potential fuel (*7*), is central to the life forms that inhabit virtually all anaerobic environments, including lake sediments, deep-sea hydrothermal vent sites, and the human intestine (*1*, *8*, *9*). Hydrogen is produced by fermentative organisms, such as clostridia, which degrade (oxidize) sugars and other organic matter, through pyruvate, to carbon dioxide and acetate (see figure). To sustain this process, oxidized electron carriers, such as the protein ferredoxin, must be regenerated so that the next sugar molecule can be degraded. With the humble proton as the electron acceptor, ferredoxin is oxidized through the production of hydrogen gas by hydrogenase.

Hydrogen seldom escapes anaerobic ecosystems. In fact, in humans and various animals, hydrogen in the breath is used to monitor intestinal malfunction (*8*). Depending on the environment, various types of hydrogen-consuming anaerobes obtain energy by using the electrons from hydrogen to produce methane or acetate from carbon dioxide, sulfide from sulfate, ferrous from ferric iron, or nitrogen and nitrite from nitrate. Hydrogen is also used to reduce carbon dioxide to cellular building blocks and, in some cases, to reduce nitrogen to ammonia. Any hydrogen that reaches aerobic environments is consumed by bacteria that obtain energy using the reaction of hydrogen and oxygen to produce water (*1*). Indeed, metabolic hydrogen interdependence may have driven the evolution of the original eukaryotic cell (*10*).

The hydrogen-evolving hydrogenase of clostridia, first purified almost 30 years ago (*11*), is now revealed as a mushroom-shaped molecule of 60-kD mass (*5*). Its single subunit contains the five distinct iron-sulfur clusters, covalently attached to the protein, that had been previously identified by spectroscopic and biochemical studies (*4*). Four of these are "conventional," one Fe_2S_2 cluster and three Fe_4S_4 clusters. Their spatial arrangement (see figure) implicates them in interprotein and intraprotein electron transfer to the fifth center, the H cluster.

Hydrogenase receives electrons from pyruvate oxidation (catalyzed by an enzyme that contains at least two Fe_4S_4 clusters) through ferredoxin (which contains two Fe_4S_4 clusters). Indeed, electrons from pyruvate oxidation may wend their way through at least seven iron-sulfur clusters before they reach yet another iron-sulfur center, the H cluster. This multitude of iron-sulfur centers brings to mind the proposals of Wächtershäuser for an iron sulfide–based primordial metabolism as the prelude to cellular life (*12*).

The complexity of the molecular machinery that has evolved in hydrogenase

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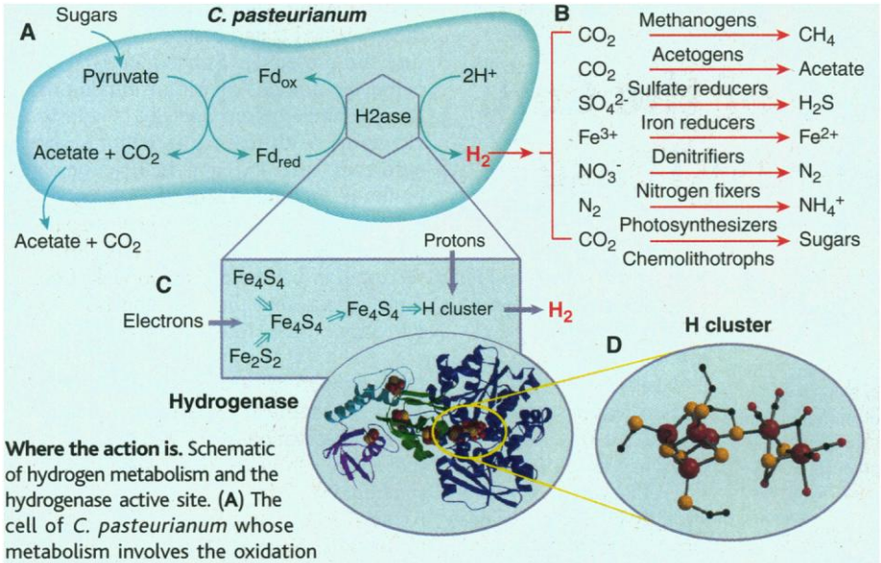

Where the action is. Schematic of hydrogen metabolism and the hydrogenase active site. **(A)** The cell of *C. pasteurianum* whose metabolism involves the oxidation of sugars and evolution of hydrogen by the iron-only hydrogenase designated as a hexagon. **(B)** The range of organisms that use hydrogen as a reductant and use the nickel-iron uptake hydrogenase. **(C)** Schematic of the iron-only hydrogenase enzyme showing paths for electron and proton transfer converging at the H center. **(D)** Schematic of the H center showing the six-iron cluster with a two-iron subcluster bound to five CO or CN^- ligands.

does not reflect the deceptive simplicity of the reaction that it catalyzes. Electrons are not "free," as implied in Eq. 1, as they shuttle between metal clusters. Similarly, protons are not "free" and available to the hydrogen-producing catalytic site. The hydrogenase structure reveals a potential pathway, involving polar protein residues, for the binding and transport of protons (*5*). From their hydrated form in solvent water, protons "glide" through the protein to the vicinity of the H cluster, where they likely rendezvous with electrons to evolve hydrogen (*4*).

The H cluster is perhaps the most remarkable feature of this hydrogenase. The precise definition of this cluster has been the subject of numerous spectroscopic investigations (*4*). Although six Fe atoms had been proposed for the H cluster, their unanticipated arrangement, now made clearer, involves a thiocubane Fe_4S_4 cluster bridged through the sulfur of a cysteine residue to a biologically unprecedented dinuclear iron subcluster. This two-iron unit has distinct organometallic character with each six-coordinate octahedral iron having two terminal CO or CN^- ligands (which are not distinguishable crystallographically). The two irons are bridged by two sulfur ligands (which are themselves connected by an as yet unknown moiety) and one CO/CN bridge. The structure of the two-iron subcluster is distinctly reminiscent of the well-studied series of organometallic complexes based on $Fe_2S_2(CO)_6$ (*13*). The presence of all the CO (or CN^-) ligands on one side of the di-iron site is a common feature of organometallic complexes (*14*).

The structure of the H cluster is new to the bioinorganic world. However, the closely related moiety $Fe(CO)(CN)_2$ had been identified as a component of the dinuclear nickel-iron site in the other major class of hydrogenase (*15*). This too came as a great surprise, extending the realm of overlap between organometallic chemistry and bioinorganic chemistry. Hence, the two families of metal-containing hydrogenase, separated for almost 20 years at the biochemical and genetic levels (*3*), are now reunited in terms of iron chemistry (*5*, *15*) and probably for very good reason. The ligation of iron by CO or CN^- ligands likely facilitates the binding of hydrogen gas or hydride (H^-) ligands, which is favored by the low oxidation states or spin states (or both) that these strong-field ligands stabilize. A site to donate and to accept protons is also required to satisfy the heterolytic mechanism of hydrogen production (from H^- and H^+) by hydrogenase (*4*), and a noncoordinated cysteine in the active site region is favored for this role in the present study (*5*).

It is ironic that the chemical reactivity necessary to reduce protons and form the H–H bond is conferred upon iron by two of the most toxic chemicals known to aerobic organisms, carbon monoxide and cyanide. How such compounds are synthesized (or accumulated) and assembled into the active site remains an open question. Nevertheless, their presence as key components of the active sites in both classes of metal-containing hydrogenase reveals that a large set of simple diatomic molecules, including H_2, N_2, NO, CO, CN^-, O_2^-, and O_2, all play key roles in biology as metabolites, substrates, signals, and toxins and, now, additionally, as components of active-site structures.

The hydrogen-evolving iron site reveals nature as opportunistic in exploiting organometallic, as well as coordination chemistry in the activation of small molecules by its metalloenzymes. The rapid evolution of hydrogen catalyzed by hydrogenase, over 10^6 turnovers per second per site at 30°C for the clostridial enzyme (*4*), is a capability that remains the envy of catalytic chemists, who wonder whether the mechanisms underlying nature's hydrogen metabolism, now known with greater definition, can be mimicked and exploited in chemical systems.

References

1. H. Gest, *Bacteriol. Rev.* **18**, 43 (1954); C. T. Gray and H. Gest, *Science* **148**, 186 (1965); L. E. Mortenson and J.-S. Chen, in *Microbial Iron Metabolism*, J. B. Neilands, Ed. (Academic Press, New York, 1974), pp. 232–282; M. W. W. Adams, L. E. Mortenson, J.-S. Chen, *Biochim. Biophys. Acta* **594**, 105 (1980).
2. H. D. Peck, A. San Pietro, H. Gest, *Proc. Natl. Acad. Sci. U.S.A.* **42**, 13 (1956).
3. E. G. Graf and R. K. Thauer, *FEBS Lett.* **136**, 165 (1981); J. LeGall *et al.*, *Biochem. Biophys. Res. Commun.* **106**, 610 (1982); R. Cammack, D. Patil, R. Aguirre, E. C. Hatchikian, *FEBS Lett.* **142**, 289 (1982); C. G. Friedrich, K. Schneider, B. Friedrich, *J. Bacteriol.* **152**, 42 (1982).
4. M. W. W. Adams, *Biochim. Biophys. Acta* **1020**, 115 (1990); S. P. J. Albracht, *ibid.* **1188**, 167 (1994).
5. J. W. Peters, W. N. Lanzolotta, B. J. Lemon, L. C. Seefeldt, *Science* **282**, 1853 (1998).
6. P. J. Chenier, *Survey of Industrial Chemistry* (VCH, New York, ed. 2, 1992), pp. 60–66.
7. J. Benemann, *Nature Biotechnol.* **14**, 1101 (1996); C. E. Thomas, I. F. Kuhn, B. D. James, F. D. Lomax, G. N. Baum, *Int. J. Hydrogen Energy* **23**, 507 (1998).
8. E. Bruun, J. N. Meyer, J. J. Rumessen, E. Gudmandhoyer, *Gut* **37**, 256 (1995); A. Minocha and S. Rashid, *Dig. Dis. Sci.* **42**, 672 (1997); J. H. Hoekstra, *Eur. J. Pediatr.* **154**, 362 (1995); F. Casellas *et al.*, *Pancreas* **16**, 481 (1998).
9. H. W. Jannasch and M. J. Mottl, *Science* **229**, 717 (1985).
10. W. Martin and M. Muller, *Nature* **392**, 37 (1998).
11. G. Nakos and L. E. Mortenson, *Biochim. Biophys. Acta* **227**, 576 (1971); J.-S. Chen and L. E. Mortenson, *ibid.* **371**, 283 (1974).
12. C. Huber and G. Wächtershäuser, *Science* **276**, 245 (1997); G. Wächtershäuser, *Prog. Biophys. Mol. Biol.* **58**, 85 (1992); in *Thermophiles: The Keys to Evolution and the Origin of Life?*, J. Wiegel and M. W. W. Adams, Eds. (Taylor and Francis, London, 1998), pp. 47–58.
13. D. Seyferth and R. S. Henderson, *J. Am. Chem. Soc.* **101**, 508 (1979); T. D. Weatherill, T. B. Rauchfuss, R. A. Scott, *Inorg. Chem.* **25**, 1466 (1986); F. T. Al-Ani, D. L. Hughes, C. J. Pickett, *J. Organomet. Chem.* **307**, c31 (1986); J. Chen, C. L. Day, R. A. Jacobson, R. J. Angelici, *ibid.* **522**, 21 (1996); D. E. Barber, R. F. Bryan, M. Sabat, K. S. Bose, B. Averill, *Inorg. Chem.* **35**, 4635 (1996); P. F. Brandt, D. A. Lesch, P. R. Stafford, T. B. Rauchfuss, *Inorg. Synth.* **31**, 112 (1997).
14. R. H. Crabtree, *The Organometallic Chemistry of the Transition Metals* (Wiley Interscience, New York, ed. 2, 1994).
15. A. Volbeda *et al.*, *Nature* **373**, 580 (1995); R. P. Happe, W. Roseboom, A. J. Pierik, S. P. J. Albracht, K. A. Bagley, *ibid.* **385**, 126 (1997); A. Volbeda *et al.*, *J. Am. Chem. Soc.* **118**, 12989 (1996); A. L. de Lacey *et al.*, *ibid.* **119**, 7181 (1997).