A dendritic iron porphyrin as a novel haemoprotein mimic: effects of the dendrimer cage on dioxygen-binding activity

Dong-Lin Jiang and Takuzo Aida*

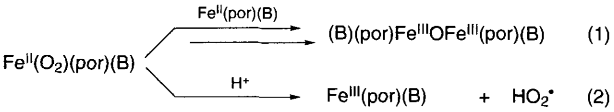
Department of Chemistry and Biotechnology, Graduate School of Engineering, The University of Tokyo, Hongo, Bunkyo-ku, Tokyo, 113, Japan

An iron(II) porphyrin 1-methylimidazole (mim) complex covalently encapsulated within a large aryl ether dendritic cage, $Fe^{II}(mim)_2L^5$, displays reversible dioxygen-binding activity, in which the dioxygen adduct survives over a period of months even in the presence of water, and exhibits a half-life of 50 h upon exposure to carbon monoxide.

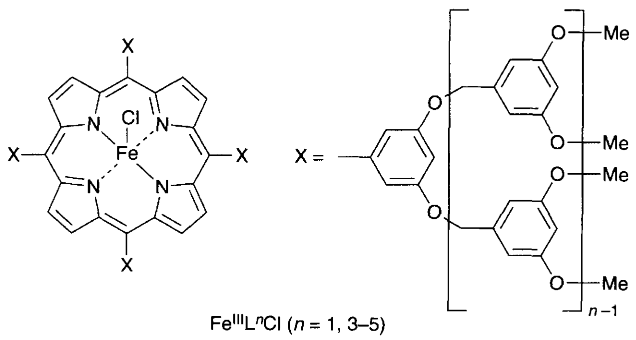
In mimicking biological functions of haemoproteins, one of the attractive targets is to realize the reversible dioxygen-binding activities of haemoglobin and myoglobin. However, (1) μ-oxo dimer formation and/or (2) H^+ (H_2O)-driven autoxidation (Scheme 1) obstruct the reversibility of this process. In order to thwart μ-oxo dimer formation, a variety of sterically hindered iron porphyrins have been synthesized,[1] some of which form stable dioxygen adducts under anhydrous conditions.[2] On the other hand, in aqueous media, successful examples have been limited to only an iron picket-fence porphyrin embedded within a synthetic bilayer membrane.[3] Herein we report an iron porphyrin covalently encapsulated within a large aryl ether dendrimer cage as the first monomolecular model of dioxygen-carrying haemoproteins.

Dendrimers are globular-shaped, hyper-branched macromolecules of nanoscopic size.[4] Recently, we have synthesized the first dendritic porphyrins (H_2L^n, n = 1–5; the number of aryloxy layers),[5–7] in which the porphyrin functionality is attached to four aryl ether dendritic arrays.[8] ^1H NMR studies showed an egg-like structural resemblance to higher-generation H_2L^n (n = 4, 5),[6] namely, a non-constrained interior environment encapsulated by a stiff exterior shell. For the preparation of $Fe^{III}ClL^n$ (n = 1, 3–5) (Fig. 1), the free-base H_2L^n and $FeBr_2$ (2–10 equiv.) were refluxed in thf for 18 h, and the reaction mixture was chromatographed on silica gel with $CHCl_3$ as eluent, where the second band collected was poured into methanolic HCl (0.1 mol dm^{-3}). $Fe^{III}ClL^n$ (n = 1, 3–5), thus obtained as precipitates, were reduced with $Na_2S_2O_4$ (1.5 equiv.) in toluene–water under argon, and the separated organic layer was washed with deaerated water, dried over Na_2SO_4, and evaporated to dryness, leaving $Fe^{II}L^n$ in 95% (n = 1)–65% (n = 5) yields based on H_2L^n.

The dioxygen-binding activities of the $Fe^{II}L^n$ (n = 1, 3–5) family were investigated in anhydrous or water-saturated toluene (4×10^{-3} dm^3) $\{[Fe^{II}L^n]_0 = 2 \times 10^{-5}$ mol dm$^{-3}\}$ at 23 °C in the presence of 30 equiv. 1-methylimidazole (mim).† The reaction was monitored by means of electronic absorption spectroscopy, using a quartz cell of 1 mm pathlength connected to a round bottomed flask (1×10^{-2} dm^3) with a three-way stopcock and a gas inlet regulator.‡ As expected from previous work,[1,9] the smallest complex, $Fe^{II}(min)_2L^1$ without any steric protection around the active site was instantly and irreversibly oxidized, upon introduction of O_2, to give a μ-oxo dimer (422, 575 nm).[1] On the other hand, the larger $Fe^{II}(mim)_2L^n$ members of the family (n = 3–5) did not show any sign of μ-oxo dimer formation, and exhibited reversible dioxygen-binding profiles. An example is shown by the largest member, $Fe^{II}(O_2)(mim)L^5$ (Fig. 2) in anhydrous toluene, where the characteristic absorbances at 423.5 and 548.5 nm did not decrease at all throughout observation over a period of 2 months in an atmospheric pressure of O_2. When O_2 was flushed out from the system by bubbling of N_2, the dioxygen adduct changed back into the deoxygenated form. Such an oxygenation–deoxygenation cycle could be repeated many times without irreversible oxidation. Interestingly, reversible dioxygen uptake of $Fe^{II}(mim)_2L^5$ was also observed in water-saturated toluene $\{[H_2O]_0 : [Fe^{II}(mim)_2L^5]_0 = 1000 : 1\}$,[10] where the dioxygen adduct still showed an extremely long lifetime (Fig. 3, with 95%



Scheme 1 Irreversible oxidation pathways for iron porphyrin–dioxygen adducts; por = porphyrinate, B = base



$Fe^{III}L^nCl$ (n = 1, 3–5)

Fig. 1 Schematic representations of $Fe^{III}L^nCl$ (n = 1, 3–5)

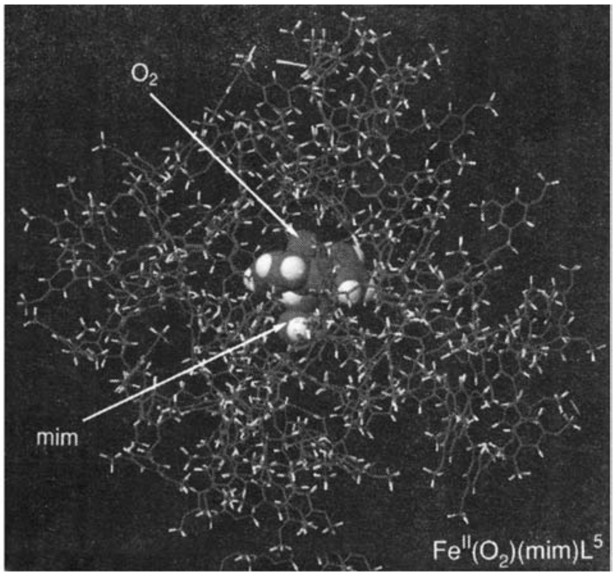


Fig. 2 Structure of $Fe^{II}(O_2)(mim)L^5$

survival after 2 months) and a rate constant of decay (k_{decay}) of 7.4×10^{-13} s^{-1}. In contrast, smaller $Fe^{II}(O_2)(mim)L^n$ (n = 3, 4) showed rapid spectral changes as a result of irreversible autoxidation,[11] where the half-lives of the dioxygen adducts were 1.5 ($k_{decay} = 1.3 \times 10^{-4}$ s^{-1}) and 6 h (3.2×10^{-5} s^{-1}), respectively. Therefore, the large dendrimer cage of $Fe^{II}(O_2)(mim)L^5$ not only inhibits bimolecular oxidation but also provides a hydrophobic barrier around the active site preventing entry of water molecules.

Another interesting aspect is the low gas permeability through a large dendrimer framework. When O_2 was introduced into an anhydrous toluene solution (4×10^{-3} dm^3) of the highest-generation complex $Fe^{II}(mim)_2L^5$ (2×10^{-5} mol dm^{-3}), complete dioxygen binding required 12 min [Fig. 4(*a*)]. Bubbling N_2 into this system resulted in complete deoxygenation after 180 min. In sharp contrast, lower-generation $Fe^{II}(mim)_2L^3$ required only 2 and 30 min for complete oxygenation and deoxygenation, respectively. These observations indicate that even small gaseous molecules such as O_2 and N_2 find it difficult to permeate through the tightly packed dendrimer framework. Along such lines, even more interesting is the unusually high resistivity of $Fe^{II}(O_2)(mim)L^5$ towards carbonylation in a carbon monoxide atmosphere: upon introduction of CO into an anhydrous toluene solution, the dioxygen adduct was transformed only very slowly into the corresponding CO adduct (426, 548 nm)[12] at a rate constant (k_{CO}) of 3.9×10^{-6} s^{-1}, the half-life of the dioxygen adduct being as high as 50 h [Fig. 4(*b*)]. This is again in sharp contrast with lower-generation $Fe^{II}(O_2)(mim)L^3$ which was completely carbonylated within 30 min ($k_{CO} = 3.9 \times 10^{-4}$ s^{-1}).

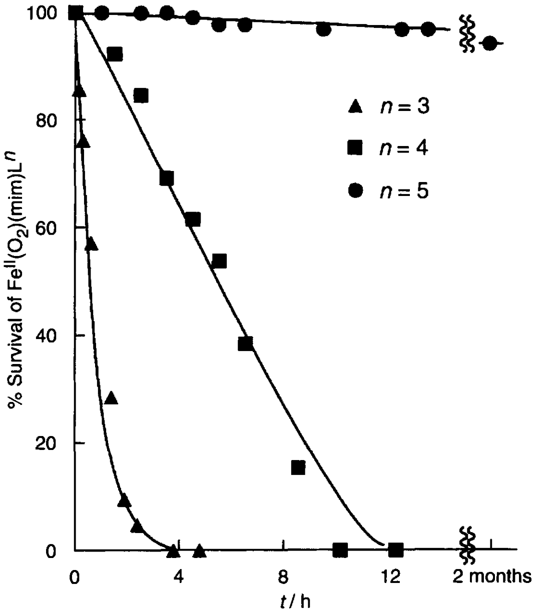


Fig. 3 % Survival of $Fe^{II}(O_2)(mim)L^n$ (n = 3–5), as determined from spectral changes in the Q-band region, in water-saturated toluene (4×10^{-3} dm^3) at 23 °C in an atmospheric pressure of O_2; $[Fe^{II}L^n]_0 = 2 \times 10^{-5}$ mol dm^{-3}, $[Fe^{II}L^n]_0 : [mim]_0 : [H_2O]_0$ = 1 : 30 : 1000

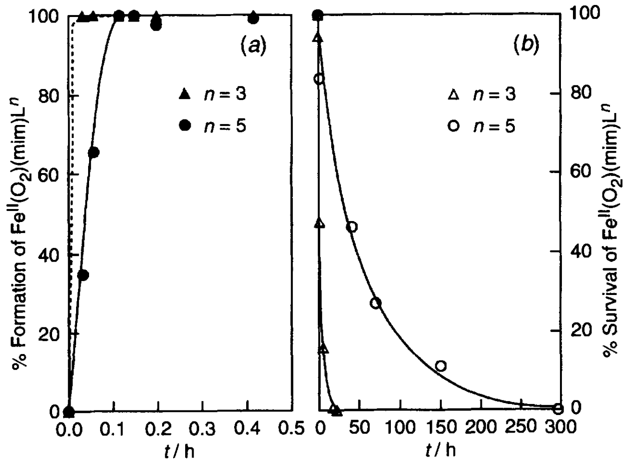


Fig. 4 Reactions of (*a*) $Fe(mim)_2L^n$ (n = 3, 5) with O_2 and (*b*) $Fe^{II}(O_2)(mim)L^n$ (n = 3, 5) with CO in anhydrous toluene (4×10^{-3} dm^3) under an atmospheric presure at 23 °C; $[Fe^{II}L^n]_0 = 2 \times 10^{-5}$ mol dm^{-3}, $[mim]_0 = 6 \times 10^{-4}$ mol dm^{-3} : time–conversion relationships, as determined from the spectral changes at the Soret region, upon bubbling of O_2 and CO, respectively, at a flow rate of 2.5×10^{-2} dm^3 min^{-1}

In conclusion, we have exploited the first dendrimer-based monomolecular model of haemoproteins, which affords a long-lived dioxygen adduct due to steric and hydrophobic protection of the active site. Restricted gas permeation through the dendrimer framework indicates a new potential of cascade architectures in materials science.

The present work was partly supported by the Grant-in-Aid on Priority-Area-Research on 'Photoreaction Dynamics' from the Ministry of Education, Science, Sports and Culture, Japan (No. 07228212).

Footnotes

† The complete formation of $Fe^{II}(mim)_2L^n$ (n = 3–5) at $[mim]_0/[Fe^{II}L^n]_0$ = 30 : 1 was confirmed by spectroscopic titration and S-shaped magnetic circular dichroism (MCD) bands in the Soret region, typical of low-spin six-coordinated iron(II) porphyrins.

‡ All gases were passed through a toluene bubbler before introduction to the reaction mixture in order to avoid vaporization of the solvent. The flow rate was adjusted at 2.5×10^{-2} dm^3 min^{-1} in each case.

References

1 M. Momemteau and C. A. Reed, *Chem. Rev.*, 1994, **94**, 659 and references therein.
2 J. Almong, J. E. Baldwin and J. Huff, *J. Am. Chem. Soc.*, 1975, **97**, 227; J. P. Collman, R. R. Gange, C. A. Reed, T. R. Halbert, G. Lang and W. T. Robinson, *J. Am. Chem. Soc.*, 1975, **97**, 1427; B. Ward, C.-B. Wang and C. K. Chang, *J. Am. Chem. Soc.*, 1981, **103**, 5236; J. P. Collman, J. I. Brauman, T. J. Collins, B. L. Iverson, G. Lang, R. B. Pettman, J. L. Sessler and M. A. Walters, *J. Am. Chem. Soc.*, 1983, **105**, 3038.
3 E. Tsuchida, T. Komatsu, K. Arai and H. Nishide, *J. Chem. Soc., Dalton Trans.*, 1993, 2465.
4 For reviews see: D. A. Tomalia, *Adv. Mater.*, 1994, **6**, 529; J. M. J. Fréchet, *Science*, 1994, **263**, 1710.
5 R.-H. Jin, T. Aida and S. Inoue, *J. Chem. Soc., Chem. Commun.*, 1993, 1260.
6 Y. Tomoyose, D.-L. Jiang, R.-H. Jin, T. Aida, T. Yamashita, K. Horie, E. Yashima and Y. Okamoto, *Macromolecules*, in the press.
7 R. Sadamoto, N. Tomioka and T. Aida, *J. Am. Chem. Soc.*, 1996, **118**, 3978.
8 C. J. Hawker and J. M. J. Fréchet, *J. Am. Chem. Soc.*, 1990, **112**, 7638.
9 K. Shikama, *Coord. Chem. Rev.*, 1988, **83**, 73.
10 M. C. Hoff. *Encyclopedia of Chemical Technology*, 3rd edn., ed. M. Grayson, Wiley, New York, 1984, vol. 23, p. 246.
11 K. Shigehara, K. Shinohara, Y. Sato and E. Tsuchida, *Macromolecules*, 1981, **14**, 1153.
12 M. Momenteau, B. Loock, C. Tetreau, D. Lavalette, A. Croisy, C. Schaeffer, C. Huel and J.-M. Lhoste, *J. Chem. Soc., Perkin Trans. 2*, 1987, 249.

Received, 7th March 1996; Com. 6/01622G