Published in final edited form as:

J Am Chem Soc. 2009 February 25; 131(7): 2460–2461. doi:10.1021/ja809781r.

A Stable Hyponitrite-Bridged Iron Porphyrin Complex

Nan Xu, Adam L. O. Campbell, Douglas R. Powel, Jana Khandogin*, and George B. Richter-Addo*

Department of Chemistry and Biochemistry, University of Oklahoma, 620 Parrington Oval, Norman, OK 73019

Abstract

The coupling of two nitric oxide (NO) molecules in heme active sites is an important contributor to the conversion of NO to nitrous oxide (N₂O) by heme-containing enzymes. Several formulations for the presumed heme-Fe{N₂O₂}ⁿ⁻ intermediates have been proposed previously, however, no crystal structures of heme-Fe{N₂O₂}ⁿ⁻ systems have been reported to date. We report the first isolation and characterization of a stable bimetallic hyponitrite iron porphyrin, [(OEP)Fe]₂(μ-N₂O₂), prepared from the reaction of the [(OEP)Fe]₂(μ-O) with hyponitrous acid. Density functional calculations were performed on the model compound [(porphine)Fe]₂(μ-N₂O₂) to characterize its electronic structure and properties.

Heme-assisted coupling of nitric oxide (NO) to form Fe{N₂O₂}ⁿ⁻ intermediates plays an important part in the reduction of NO to N₂O.^{1,2} In transition-metal chemistry, such metal {N₂O₂}ⁿ⁻ moieties can be generated from attack of NO on a metal-NO group,^{3,4} from metal-induced coupling of NO,⁵ or from transfer of an intact (N₂O₂)ⁿ⁻ from a diazeniumdiolate to a metal.⁶ It is interesting to note that only a small number of metal{N₂O₂}ⁿ⁻-containing compounds have been structurally characterized, and the {N₂O₂} binding modes determined to date are sketched below as structures **A** (M = Pt, Ni),^{5,6} **B** (M = M' = Co),⁷ **C** (M = Ru)⁸ and **D** (M = Co).⁹

Enzymes such as the NO reductase (NOR) from *Paracoccus denitrificans*, cyt *ba₃* and *caa₃* from *Thermus thermophilus*, and cyt *cbb₃* from *Paracoccus stutzeri* catalyze the conversion of NO to N₂O using bimetallic active sites.^{1,2} For these biological systems, intermediates resembling structure **B** have been proposed, where the metal{N₂O₂} moiety is *N*-bound to heme Fe (M), and where M' (non-heme Fe or Cu) may contact both O atoms.^{1,10} Collman *et al.* have proposed, using results from the reaction of a diferrous synthetic model of NOR with NO, that a *trans* bis-nitrosyl intermediate forms at the active site of NOR followed by NO coupling (structure **E**) to give N₂O and a bis-ferric product.¹¹ Resonance Raman spectroscopy

has been employed to assign a HONNO-bridged Fe-Cu species (i.e., a protonated analogue of **E**) during NO reduction by *T. thermophilus* cyt *ba*₃.¹² To the best of our knowledge, however, no adduct of heme and an N₂O₂ moiety has been structurally characterized.

The title compound was obtained from the reaction of [(OEP)Fe]₂(μ-O) in anhydrous toluene with an in situ-generated ether solution of hyponitrous acid (Supporting Information) (eq 1). Workup gave the product [(OEP)Fe]₂(μ-N₂O₂) (**1**) as dark purple microcrystals in 52% isolated yield.

The IR spectrum of **1** reveals a band at 982 cm⁻¹ assigned to ν_{as} of the N-O group (ν_{as}(¹⁵NO) 973 cm⁻¹). This band is lower than that reported for H₂N₂O₂ at 1014/1003 cm⁻¹ (ν_{as}(¹⁵NO) 991/980 cm⁻¹).¹³ The ν_s(NO) and ν(NN) bands were not observed in the IR spectrum, presumably due to the symmetry of **1**. The EPR spectrum of **1** as a CH₂Cl₂/toluene glass (77 K) shows *g* = 5.74 and 2.03; similar data were obtained for high-spin (H.S.) ferric hydroxo porphyrins.¹⁴

The X-ray crystal structure of **1** reveals that the N₂O₂ ligand is bound to each Fe via the η¹-O binding mode and that the ONNO moiety is *trans* (Figure 1). The N–N bond length of 1.250 (3) Å suggests double bond character (*c.f.*, 1.256(2) Å in crystalline Na₂N₂O₂).¹⁵ The distance between the two Fe centers is 6.7 Å, and is longer than the 4.4 Å distance between the Fe and Cu centers in *T. thermophilus* cyt *ba*₃ that exhibits NOR activity.¹⁶ The magnitude of the Fe–N(por) bond lengths in **1** and the apical displacement of Fe from the 4N-atom porphyrin mean plane (ΔFe = 0.40 Å) are indicative of H.S. ferric Fe centers. It follows that the N₂O₂ ligand in **1** must have significant hyponitrite (i.e., dianionic) character.

To characterize the electronic structure and properties of **1**, we performed calculations using density functional theory (DFT) on the model compound [(porphine)Fe]₂(η¹,η¹,μ-N₂O₂) (**1**-calc) in the *trans* and *cis* configurations. The DFT calculations were based on the B3LYP exchange-correlation functional and a triple-zeta polarization basis set. In both *trans* and *cis* configurations, the H.S. species is calculated to be lower in energy relative to the intermediate- (I.S.) and low-spin (L.S.) variants by 8 and 19 kcal/mol, respectively (Table S2). The calculated geometry of the *trans* H.S. species most closely reproduces the crystal structural data (Table 1). Curiously, the different spin states of the *trans* isomer are calculated to be ~2 kcal/mol higher in energy than the respective *cis* variants. This small difference lies within the typical error of 3–5 kcal/mol estimated for B3LYP calculations of transition-metal complexes. Thus, the DFT calculations are consistent with the EPR and structural data in establishing the identity of a stable H.S. *trans* species. The small energy gap between the *trans* and *cis* configurations suggests that isolation of the *cis* derivative of **1** might be attainable.

To probe the charge distribution in **1**, we used multipole-derived population analysis for the *trans* H.S. form of **1**-calc. The atomic charges on the Fe, O and N atoms are shown in Figure 2. To further understand the charge distribution in the FeONNOFe moiety, we used the Nalewajski-Mrozek scheme to calculate the bond orders. Accordingly, the bond orders for Fe–O, O–N, and N–N in 1-calc *trans*-H.S. (Figure 2) are 0.79, 1.25, and 1.84, respectively, which supports the notion of an N=N double bond in the hyponitrite bridge.

The Kohn-Sham orbitals of the *trans* H.S. species of **1**-calc are expected to be similar to those of square pyramidal metal complexes, for which the highest occupied orbital and the lowest unoccupied orbital are the result of anti-bonding interactions between dz² as well as d(x²-y²) orbitals of the metal and the symmetrized combinations of the ligand σ orbitals. The frontier spin orbitals from the unrestricted openshell calculation are shown in Figure 3. The highest

occupied α spin orbital exhibits more porphine character while the lowest unoccupied spin β orbital displays more metal character. The N atoms of the hyponitrite bridge form a bonding interaction in both highest occupied spin orbitals.

Protonation of a toluene solution of **1** at 0 °C using HCl results in the formation of N₂O and (OEP)FeCl (eq 2). An IR

spectrum of the headspace reveals new bands at 2236/2213 and 1298/1266 cm⁻¹ assigned to ν_{as} and ν_{s} of N₂O, respectively. The use of the ¹⁵N-labeled **1** (labeled at hyponitrite) shifts the ν_{as} bands to 2167/2144 cm⁻¹; the corresponding ν_{s} bands were not observed due to its occurrence outside the detection window.

We hypothesized that the protonation reaction is initiated by H⁺ attack on one of the O atoms of the hyponitrite bridge followed by O–N bond cleavage to give N₂O. This is supported by the pronounced negative charge on the hyponitrite O atom in *trans*-H.S. **1**-calc obtained from the DFT calculation. Further studies are underway to uncover the mechanism of the protonation reaction.

In summary, we have prepared and characterized the first isolable hyponitrite iron porphyrin complex, and describe the first established hyponitrite O-bonding mode to a heme model.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgment

We are grateful to the National Institutes of Health for funding (GM076640; GBR-A).

References

1. Moenne-Loccoz P. Nat. Prod. Rep 2007;24:610–620. [PubMed: 17534533]
2. Yeung N, Lu Y. Chem. Biodiversity 2008;5:1437–1454.
3. Franz KJ, Lippard SJ. J. Am. Chem. Soc 1999;121:10504–10512.
4. Schneider JL, Carrier SM, Ruggiero CE, Young VG, Tolman WB. J. Am. Chem. Soc 1998;120:11408–11418.
5. Arulsamy N, Bohle DS, Imonigie JA, Moore RC. Polyhedron 2007;26:4737–4745.
6. Arulsamy N, Bohle DS, Imonigie JA, Levine S. Angew. Chem. Int. Ed 2002;41:2371–2373.
7. Villalba MEC, Navaza A, Guida JA, Varetto EL, Aymonino PJ. Inorg. Chim. Acta 2006;359:707–712.
8. Bottcher H-C, Graf M, Mereiter K, Kirchner K. Organometallics 2004;23:1269–1273.
9. Bau R, Sabherwal IH, Burg AB. J. Am. Chem. Soc 1971;93:4926–4928.
10. Blomberg LM, Blomberg MRA, Siegbahn PEM. Biochim. Biophys. Acta 2006;1757:31–46. [PubMed: 16375849]
11. Collman JP, Dey A, Yang Y, Decreau RA, Ohta T, Solomon EI. J. Am. Chem. Soc 2008;130:16498–16499. [PubMed: 19049449]
12. Varotsis C, Ohta T, Kitagawa T, Soulimane T, Pinakoulaki E. Angew. Chem. Int. Ed 2007;46:2210–2214.
13. McGraw GE, Bernitt DL, Hisatsune IC. Spectrochim Acta 1967;23A:25–34.
14. Cheng R-U, Latos-Grazynski L, Balch AL. Inorg. Chem 1982;21:2412–2418.

15. Arulsamy N, Bohle DS, Imonigie JA, Sagan ES. *Inorg. Chem* 1999;38:2716–2725.
16. Soulimane T, Buse G, Bourenkov GP, Bartunik HD, Huber R, Than ME. *Embo J* 2000;19:1766–1776. [PubMed: 10775261]

Figure 1.

Molecular structure of $[(\text{OEP})\text{Fe}]_2(\mu\text{-ONNO})$ (**1**). Hydrogen atoms and the dichloromethane solvates have been omitted for clarity. Selected bond lengths ($\text{\AA}$) and angles ($^\circ$): $\text{Fe-O} = 1.889(2)$, $\text{O-N} = 1.375(2)$, $\text{N-N} = 1.250(3)$, $\text{Fe-N}(\text{por}) = 2.049(2)\text{--}2.064(2)$, $\angle\text{FeON} = 118.56(12)$, $\angle\text{NNO} = 108.5(2)$.

Figure 2.
Calculated atomic charges and bond orders (*bo*) for the FeONNOFe moiety.

Figure 3. Frontier spin orbitals for high-spin 1-calc. HOSO and LUSO denote the highest occupied and the lowest unoccupied spin orbitals, respectively.

Table 1
Selected crystal data for **1**, and calculated geometry data (in Å and °) for [(porphine)Fe]₂(η¹,η¹,μ-N₂O₂)(**1**-calc).

	Fe-O	O-N	N-N	∠NNO	ΔFe
1 (crystal; <i>trans</i>)	1.889	1.375	1.250	108.5	0.402
1 -calc					
<i>trans</i> -H.S.	1.909	1.376	1.269	108.9	0.482
<i>trans</i> -I.S.	1.951	1.364	1.280	109.3	0.272
<i>trans</i> -L.S.	1.831	1.398	1.266	107.4	0.249
<i>cis</i> -H.S.	1.899	1.390	1.254	117.0	0.485
<i>cis</i> -I.S.	1.945	1.379	1.262	117.9	0.279
<i>cis</i> -L.S.	1.831	1.403	1.258	116.5	0.232