

Formation of the Dinuclear Iron–Nitrosyl Complex $[\text{Fe}_2(\text{SMe})_2(\text{NO})_4]$ by Incorporation of SMe Groups from Methionine in Reactions with Iron(II) Salts and Nitrite

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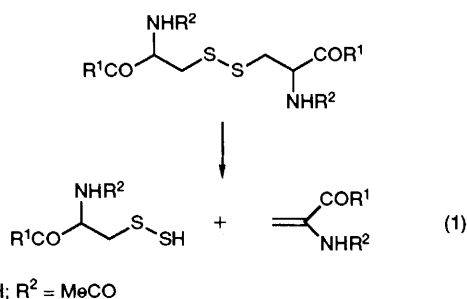
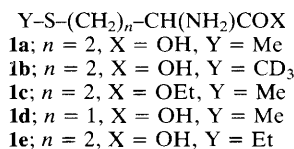
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The dinuclear iron–nitrosyl complex $[\text{Fe}_2(\text{SMe})_2(\text{NO})_4]$ has been isolated from reactions of iron(II) salts and sodium nitrite with methionine, under reaction conditions relevant to food processing.

Sodium nitrite is a long-established food preservative, valuable particularly for preventing the growth of *Clostridium botulinum* in preserved foodstuffs. The isolation of the iron–sulfur nitrosyl complex $[\text{Fe}_2(\text{SMe})_2(\text{NO})_4]$ from plant material (of uncertain taxonomic identity) after storage in water of moderately high nitrite content,¹ and the confirma-

tion² of this observation using parsley, *Petroselinum crispum*, has led to speculation³ on the biosynthetic origin of this complex, known to be a promoter of the tumorigenic properties of both *N*-nitrosamines and polycyclic aromatic hydrocarbons.^{1d,4,5}

The iron–sulfur clusters of redox proteins and their



synthetic analogues both react readily with nitrite to yield iron-nitrosyl complexes.^{6,7} For the synthetic clusters, the primary product of the reaction with nitrite is $[\text{Fe}(\text{NO})_2(\text{SH})_2]^-$, which in a self-assembly reaction forms the tetranuclear product $[\text{Fe}_4\text{S}_3(\text{NO})_7]^-$, isolable in yields of ca. 40%.⁷ The anion $[\text{Fe}_4\text{S}_3(\text{NO})_7]^-$ itself reacts with electrophilic alkylating or arylating agents to give the dinuclear complexes $[\text{Fe}_2(\text{SR})_2(\text{NO})_4]$;⁸ hence reaction of nitrite with such preformed iron-sulfur clusters has been suggested as the initial step in the biological formation of $[\text{Fe}_2(\text{SMe})_2(\text{NO})_4]$.³ However, we have subsequently demonstrated⁹ that preformed clusters are not necessary for the formation of $[\text{Fe}_4\text{S}_3(\text{NO})_7]^-$: in the presence of iron(II) salts, nitrite will effect the incorporation into $[\text{Fe}_4\text{S}_3(\text{NO})_7]^-$ of sulfur from a wide range of amino acids and their derivatives.

We now report direct formation of $[\text{Fe}_2(\text{SMe})_2(\text{NO})_4]$ by incorporation of the intact SMe fragment of methionine: in the presence of iron(II) sulfate and sodium nitrite, methionine yields $[\text{Fe}_2(\text{SMe})_2(\text{NO})_4]$, isolable by chromatography on silica, under a range of experimental conditions relevant to food processing.[†] The product was identified by a combination of Fourier transform IR,¹⁰ ¹H (including ASIS) and ¹³C NMR, and ¹⁵N NMR on the product $[\text{Fe}_2(\text{SMe})_2(^{15}\text{NO})_4]$ isolated from a reaction using 99% enriched $\text{Na}[^{15}\text{NO}_2]$,¹¹ and by mass spectrometry of material isolated both from normal methionine, **1a** $[\text{Fe}_2(\text{SMe})_2(\text{NO})_4]$ and from $[\text{Me-}^2\text{H}_3]$ -methionine **1b**, $[\text{Fe}_2(\text{SCD}_3)_2(\text{NO})_4]$.[‡] That the sulfur is derived from methionine, rather than from reduction of sulfate,¹² was confirmed by control experiments using iron(II) chloride instead of the sulfate: the yields of $[\text{Fe}_2(\text{SMe})_2(\text{NO})_4]$ were unchanged. Comparable yields of $[\text{Fe}_2(\text{SMe})_2(\text{NO})_4]$ were obtained from similar reactions of iron(II) salts, sodium nitrite and either methionine ethyl ester **1c** or *S*-methylcysteine **1d**.

[†] Autoclave at 118 °C for 20 min; isolated, purified, yield of $[\text{Fe}_2(\text{SMe})_2(\text{NO})_4]$ ca. 4% (based on equimolar methionine and iron(II) as limiting components, with nitrite in 20% molar excess); microwave 90 °C for 1 h, 650 W, trace only; reflux under N_2 for 2 h; isolated, purified yield ca. 10%, calculated as previously.

[‡] Compound **1a** yielded $[\text{Fe}_2(\text{SMe})_2(\text{NO})_4]$: IR (tetrahydrofuran (thf)), $\nu(\text{NO})/\text{cm}^{-1}$ 1776s, 1751s (lit.,¹⁰ 1776s, 1751s); δ_{H} (CDCl_3), 2.83 (lit.,^{11a} 2.83); δ_{H} ($^2\text{H}_8$ toluene), 2.17, 2.23 (lit.,^{11d} 2.16, 2.23); δ_{C} (CDCl_3) 27.5(q) [lit.,^{11b} 27.5(q)]; m/z , 326 M^+ , 296 $(\text{M} - \text{NO})^+$, 266 $(\text{M} - 2\text{NO})^+$, 236 $(\text{M} - 3\text{NO})^+$, 206 $(\text{M} - 4\text{NO})^+$, 191 $(\text{Fe}_2\text{S}_2\text{Me})^+$, 176 $(\text{Fe}_2\text{S}_2)^+$.

Compound **1b** yielded $[\text{Fe}_2(\text{SCD}_3)_2(\text{NO})_4]$: IR (thf), $\nu(\text{NO})/\text{cm}^{-1}$ 1776s, 1751s; m/z , 332, 302, 272, 242, 212, 194, 176.

Compound **1a** in the presence of $\text{Na}[^{15}\text{NO}_2]$ yielded $[\text{Fe}_2(\text{SMe})_2(^{15}\text{NO})_4]$: IR (thf), $\nu(^{15}\text{NO})/\text{cm}^{-1}$ 1739s, 1714s [lit.: (Nujol),^{11b} 1740s, 1716s]; δ_{N} ($^2\text{H}_6$ acetone) 26.1 (d, J 3.2 Hz), 32.7(s), 38.0(d, J 3.2 Hz) [lit., (CD_2Cl_2)^{11c} 23.1 (d, J 2.8 Hz), 30.5(s), 36.2(d, J 2.8 Hz)]; m/z , 330, 299, 268, 237, 206, 191, 176.

Although preformed iron-sulfur clusters are clearly not required for the formation of $[\text{Fe}_2(\text{SR})_2(\text{NO})_4]$ complexes, nevertheless we have observed that the tetranuclear anion $[\text{Fe}_4\text{S}_3(\text{NO})_7]^-$ reacts with methionine to provide $[\text{Fe}_2(\text{SMe})_2(\text{NO})_4]$, although in this case the source of the sulfur in the dinuclear product is not yet proven.

The formation of tetranuclear $[\text{Fe}_4\text{S}_3(\text{NO})_7]^-$ from cysteine and its derivatives⁹ and of dinuclear $[\text{Fe}_2(\text{SMe})_2(\text{NO})_4]$ from derivatives of methionine or *S*-methylcysteine requires cleavage of carbon-sulfur bonds. Ghadimi and Hill have recently shown¹³ that such C-S bond cleavage in cysteine derivatives is extremely easy under conditions similar to those employed here [see eqn. (1)]. A similar cleavage of methionine derivatives would yield MeSH, long known¹⁴ to provide $[\text{Fe}_2(\text{SMe})_2(\text{NO})_4]$ in the presence of iron(II) and either nitrite or NO.

It is of interest that in reactions of iron(II) salts with nitrite and ethionine **1e**, where two different C-S bond cleavage reactions are possible, the sole iron complex identified by IR, ¹H, ¹³C and ¹⁵N NMR, and mass spectrometry was the neutral dinuclear $[\text{Fe}_2(\text{SEt})_2(\text{NO})_4]$.[§]

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References

- (a) G. H. Wang, W. X. Zhang and W. G. Chai, *Acta Chim. Sinica*, 1980, **38**, 95; (b) G. H. Wang, W. X. Zhang and W. G. Chai, *Adv. Mass Spectrom.*, 1980, **8B**, 1369; (c) W. X. Zhang, M. S. Xu and G. H. Wang, *Cancer Res.*, 1983, **43b**, 339; (d) M. X. Li and S. J. Cheng, *Chin. Med. J.*, 1984, **97b**, 311.
- J. D. Baty, R. G. Willis, M. G. Burdon, A. R. Butler, C. Glidewell, I. L. Johnson and R. Massey, *Inorg. Chim. Acta*, 1987, **138**, 15.
- A. R. Butler, C. Glidewell and M. H. Li, *Adv. Inorg. Chem.*, 1988, **32**, 335.
- S. J. Cheng, M. Sala, M. H. Li, I. Courtois and I. Chouroulinkov, *Carcinogenesis*, 1981, **2**, 313.
- S. Lu, P. Lin, F. Lu and Y. Wang, *Zhonghua Zhonglin Zazhi*, 1985, **7**, 241; *Chem. Abstr.*, 1986, **104**, 63958e.
- D. Reddy, J. R. Lancaster, Jr. and D. P. Cornforth, *Science*, 1983, **221**, 769.
- A. R. Butler, C. Glidewell, A. R. Hyde and J. C. Walton, *Inorg. Chim. Acta*, 1985, **106**, L7.
- C. Glidewell, R. J. Lambert, M. E. Harman and M. B. Hursthouse, *J. Chem. Soc., Dalton Trans.*, 1990, 2685.
- A. R. Butler, C. Glidewell and S. M. Glidewell, *Polyhedron*, 1990, **9**, 2399.
- J. A. Crayston, C. Glidewell and R. J. Lambert, *Polyhedron*, 1990, **9**, 1741.
- (a) C. Glidewell and A. R. Hyde, *Polyhedron*, 1985, **4**, 1155; (b) C. Glidewell and I. L. Johnson, *Chem. Scr.*, 1987, **27**, 441; (c) A. R. Butler, C. Glidewell, A. R. Hyde and J. McGinnis, *Inorg. Chem.*, 1985, **24**, 2931; (d) A. R. Butler, C. Glidewell, A. R. Hyde, J. McGinnis and J. E. Seymour, *Polyhedron*, 1983, **2**, 1045.
- A. R. Butler, C. Glidewell and S. M. Glidewell, *Thermochim. Acta*, 1986, **106**, 355.
- M. Ghadimi and R. R. Hill, *J. Chem. Soc., Chem. Commun.*, 1991, 903.
- K. A. Hofmann and O. F. Wiede, *Z. Anorg. Allg. Chem.*, 1895, **9**, 295.

[§] Compound **1e** yielded $[\text{Fe}_2(\text{SEt})_2(\text{NO})_4]$: IR (thf) $\nu(\text{NO})/\text{cm}^{-1}$ 1774s, 1749s (lit.,¹⁰ 1774s, 1749s); δ_{H} (CDCl_3) 1.53(t), 1.58(t), 3.05(q), 3.07(q) [lit.,^{11d} 1.53(t), 1.58(t), 3.07(q), 3.10(q)]; δ_{C} (CDCl_3) 19.1(q), 39.5(t), 40.1(t) [lit.,^{11d} 19.1(q), 39.5(t), 40.2(t)]; m/z 354 M^+ , 324 $(\text{M} - \text{NO})^+$, 294 $(\text{M} - 2\text{NO})^+$, 266 $[\text{Fe}_2(\text{NO})_2(\text{SEt})(\text{SH})]^+$, 236 $[\text{Fe}_2(\text{NO})(\text{SEt})(\text{SH})]^+$, 208 $[\text{Fe}_2(\text{NO})_2\text{S}_2\text{H}_2]^+$, 178 $(\text{Fe}_2\text{S}_2\text{H}_2)^+$, 177 $(\text{Fe}_2\text{S}_2\text{H})^+$, 176 $(\text{Fe}_2\text{S}_2)^+$.

Compound **1e** in the presence of $\text{Na}[^{15}\text{NO}_2]$ yielded $[\text{Fe}_2(\text{SEt})_2(^{15}\text{NO})_4]$: IR (thf) $\nu(^{15}\text{NO})/\text{cm}^{-1}$ 1739s, 1714s [lit.: (Nujol)^{11b} 1740s, 1716s]; δ_{N} (CDCl_3) 20.9 (d, J 2.8 Hz), 26.5(s), 31.3 (d, J 2.8 Hz) [lit., (CD_2Cl_2)^{11c} 25.0 (d, J 3.0 Hz), 31.4(s), 36.1 (d, J 3.0 Hz)]; m/z 358, 327, 296, 268, 237, 209, 178, 177, 176.