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Synthesis of a Non-Heme Template for Attaching Four Peptides: An Approach to Artificial Iron(II)-Containing Peroxidases

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We are developing all-synthetic model cofactor−protein complexes in order to define the parameters controlling non-natural cofactor activity. The long-term objective is to establish the theoretical and practical basis for designing novel enzymes. A non-heme pentadentate ligand (N4Py) is being developed as a template for the site-specific attachment of a designed four-helix bundle. Previously, we attached two unprotected peptides via CH_2Cl handles to N4Py. In the presence of hydrogen peroxide, the iron(II) complex of this ligand (**2a**) generates an $Fe^{III}OOH$ intermediate (**3a**) that can oxidize a wide variety of organic compounds. Here, we describe the synthesis of **27**, a N4Py derivative in which four three-carbon spacers have been introduced, and show that four copies of an unprotected, single-cysteine peptide can be coupled via a thioether linkage to the ligand. In addition, a divergent synthesis route to tetrabromide ligand **1b** has also been developed, providing the opportunity to prepare alternative pentadentate ligands efficiently by four cross-coupling reactions on a single molecule. Also, two of the four bromides of **1b** can be selectively addressed by magnesium−bromide exchange.

Introduction

With an increasing understanding of the principles governing protein structure and function, together with initiatives over the past decade in the de novo design of proteins, simple, stable proteins that fold as predicted and exhibiting rudimentary catalytic activity can now be designed with some success.[1] One approach to aid the folding of designed proteins is template-assembled synthetic proteins (TASP),[2] where the peptide chains are attached to a template that helps direct the folding of the protein. Cavitands,[3] (cyclic) peptides,[4] carbohydrates,[5] dendrimers,[6] and a cyclotribenzylene macrocycle[7] are some examples of applied non-metal-binding templates. Metal-ion-assisted self-assembly processes of peptides[8] and chelating molecules, such as porphyrins,[4e,9] have been employed to prepare de novo designed metalloproteins. For instance, the incorporation of a heme group,

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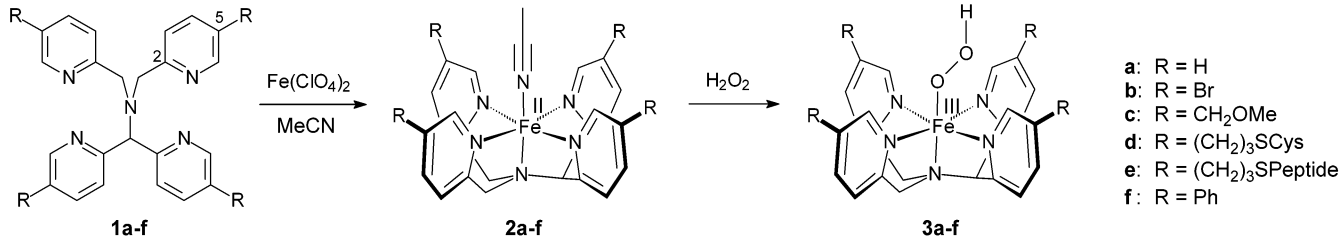
SCHEME 1



1a-f **2a-f** **3a-f**

either by covalent attachment or by coordination of the metallo−heme unit to peptide-bound histidine, can introduce functionality into a protein structure.[10]

Here, we address the design and preparation of a first-generation non-heme iron complex for incorporation into a four-helix bundle, a simple protein consisting of four α-helices. We ultimately aim to construct a functional non-heme peroxidase mimic in which the catalytic cofactor is integrated into a designed four-helix bundle. The simplicity of such catalyst−protein complexes will permit the factors controlling protein-mediated modulation of cofactor activity to be defined. Eventually, the size, shape, and hydrophobicity of the active site and the substrate binding interactions in the four-helix bundle will be altered in order to tune selective substrate oxidation.

The template chosen for peptide attachment is the neutral pentadentate ligand N4Py (**1a**) (Scheme 1),[11] which consists of four pyridine rings anchored via one-carbon bridges to a central nitrogen atom.[12] In the presence of hydrogen peroxide, its iron(II) complex N4PyFe **2a** generates a short-lived $Fe^{III}OOH$ intermediate (**3a**)[13] that can oxidize a wide variety of organic compounds.[14] This non-heme iron system has successfully been transformed into an effective DNA cleaving agent that mimics iron bleomycin.[15]

The geometry of the N4PyFe complex appears to complement the spacial arrangement of the four peptide chains that constitute the core of the four-helix bundle (Figure 1). In a preliminary study, two peptide chains were attached to a disubstituted N4Py derivative by means of cysteine thiol-mediated nucleophilic substitution on $-CH_2Cl$ groups.[16] The corresponding iron(II) complex retained its catalytic oxidation activity with hydrogen peroxide in water.[16] These results indicated that the incorporation of the N4PyFe catalyst into a designed four-helix bundle environment is feasible.

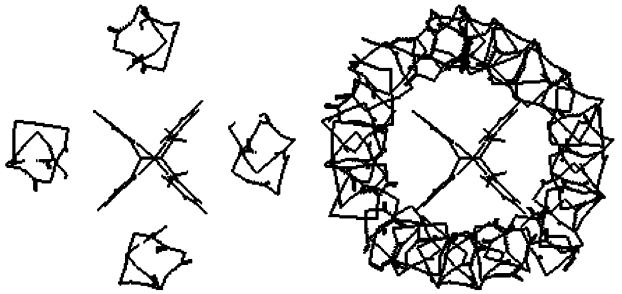


FIGURE 1. Crystal structure of N4PyFe **2a** superimposed on the crystal structure of GCN4-p1-LI, a homo-tetrameric coiled-coil (side chains are omitted for clarity). Left: a 7-residue, 10 Å slice of GCN4-p1-LI,[17] showing N4PyFe interdigitating between the helices of the bundle; right: a 47 Å slice, showing the full-length coiled-coil.

The challenge addressed in this paper is to modify the N4Py ligand to host four peptide chains. This essentially entails two key issues. The first is the development of a new synthesis route to tetrafunctionalized N4Py ligands. The second is a stability issue: the implementation of the previously developed methodology (the use of $-CH_2Cl$ groups for peptide attachment) resulted in the intrinsically unstable ligand **26** (Scheme 5). A stable ligand was obtained by extending the linker to a three-carbon spacer to provide template **27**. As both **26** and **27** were prepared via the same synthesis route, their syntheses will be discussed simultaneously.

Results and Discussion

Convergent Synthesis Strategy. From visual inspection of molecular models of **2a**, it was deduced that the linkers for peptide binding should be one methylene group long (C1-spacer) and attached at the 5-positions of the pyridine rings. Consequently, the 2-positions are to be utilized to construct the N4Py core (Scheme 1). In the final iron(II) complex, the linkers at the 5-position will thus direct the peptide chains away from the iron center. According to the models, this architecture provides an unstrained linkage between the folded four-helix bundle and the cofactor. A convergent synthesis strategy was chosen wherein the required spacer and functionality for the target template are incorporated into the building blocks before the core of the ligand itself is constructed.

Several procedures are known for the preparation of 2,5-disubstituted pyridine synthons for the construction of functionalized N4Py derivatives.[16,18−20] In a departure from these methods, tetrasubstituted N4Py derivatives

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(11) Abbreviations used: N4Py, *N*-[di(2-pyridinyl)methyl]-*N*,*N*-bis-(2-pyridinylmethyl)amine; N4PyFe, the iron(II) complex of N4Py.

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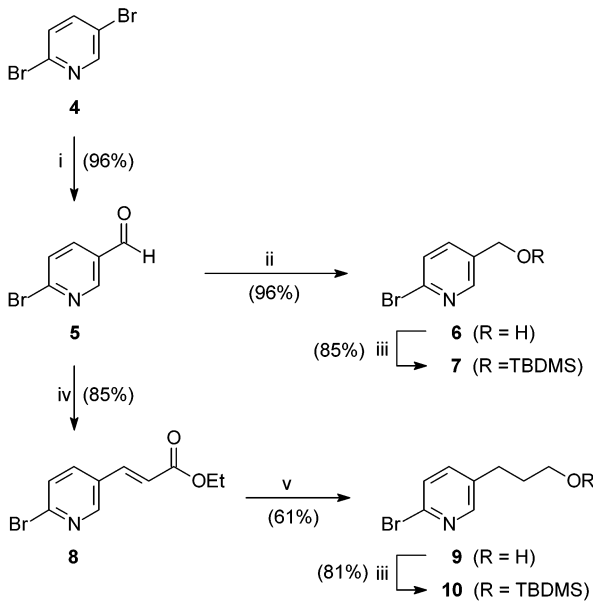
SCHEME 2[a]



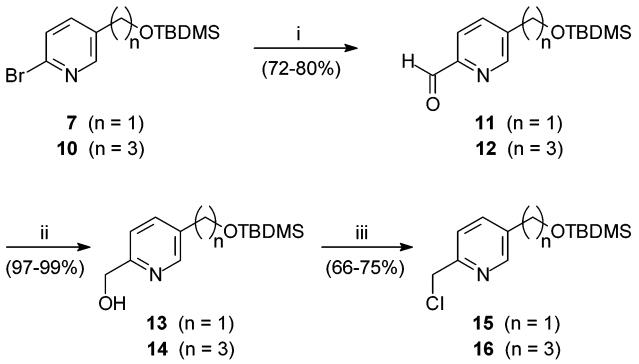
[a] Conditions: (i) (1) *n*-BuLi, Et_2O, −80 °C, 1 h, (2) DMF, −80 °C, 1 h, (3) H_3O^+; (ii) $NaBH_4$, MeOH, 0 °C to rt, 15 min; (iii) TBDMSCl, TEA, DMF, 15 min; (iv) $(EtO)_2P(O)CH_2CO_2Et$, NaH, THF/Et_2O, 0 °C to rt, 1 h; (v) $LiAlH_4$, THF, 0 °C to rt, 75 min.

23 and **24** were constructed starting from commercially available 2,5-dibromopyridine (**4**). This enables a versatile and controlled introduction of various functionalities at the desired positions of the pyridine ring (vide infra) and consequently facilitates the preparation of a wide range of tetrafunctionalized N4Py derivatives.

Ligand Synthesis. For the synthesis of the N4Py ligand **23**, with a C1-spacer, pyridine derivative **7** was required. The C1-spacer in building block **7** was introduced by preparing aldehyde **5** from 2,5-dibromopyridine (**4**) by a regioselective lithiation[21] with *n*-BuLi in diethyl ether at −80 °C, followed by quenching with *N*,*N*-dimethylformamide (DMF) (Scheme 2).

Although the 2-position in **4** is more electron-deficient than the 5-position, mono-metalation under these conditions occurs selectively at the latter position,[21] providing a powerful tool for regioselective functionalization. Aldehyde **5** was subsequently reduced with $NaBH_4$ in MeOH to furnish alcohol **6**, which was protected with *tert*-butyldimethylsilyl chloride (TBDMSCl) in DMF in the presence of triethylamine (TEA) to provide the essential building block **7** in 78% yield from **4**.

SCHEME 3[a]



[a] Conditions: (i) (1) *n*-BuLi, THF, −80 °C, 40 min, (2) DMF, −80 °C, 1 h; (3) H_2O; (ii) $NaBH_4$, MeOH, 0 °C to rt, 15 min; (iii) $SOCl_2$, TEA, DCM, rt, 18 h.

As noted above, molecular modeling indicates that a single methylene group as spacers would provide optimal unstrained linkage between the folded bundle and the ligand. However, due to stability problems with the ligand containing C1-spacers (vide infra), it was necessary to also investigate the synthesis of the ligand with C3-spacers. To obtain key building block **10** for the construction of the N4Py ligand **24**, with a C3-spacer, aldehyde **5** was converted to the α,β-unsaturated ester **8** by a Horner−Emmons reaction using triethyl phosphonoacetate (Scheme 2). Subsequent reduction by $LiAlH_4$ to the saturated alcohol **9** was followed by a silylation to afford the TBDMS ether **10** in 81% yield after purification.

Depending on the spacer length in the final ligand, the required 2-picolyl chlorides **15** and **16** can be obtained from building blocks **7** and **10**, respectively (Scheme 3). To this end, these building blocks were lithiated with *n*-BuLi in THF at −80 °C and quenched with DMF to provide the aldehydes **11** and **12** (80% and 72% yield, respectively). Alternatively, the bromo−metal exchange can be accomplished by the use of *i*-PrMgBr in THF at room temperature,[22] which furnishes **11** and **12** in similar yields. These aldehydes can in principle be reduced in situ to the picolyl alcohols **13** and **14**. However, as both aldehydes are also required for the synthesis of bis(2-pyridinyl)methylamine **21** and **22** (Scheme 4), these reductions were carried out in a separate reaction step. Finally, by reaction with thionyl chloride in the presence of TEA alcohols **13** and **14** were quantitatively converted to the corresponding 2-picolyl chlorides **15** and **16**, and isolated in 66% and 75% yield, respectively, after purification. Flash column chromatography of these compounds proved to be crucial to minimize decomposition of the product during purification.[23]

The key step in the synthesis of tetrasubstituted N4Py ligands is the coupling of two functionalized pyridine rings via a methylene bridge, preceding the formation of the required bis(2-pyridinyl)methylamines **21** and **22**. This coupling reaction was carried out by allowing lithiated **7** or **10** to react with the 2-formylpyridine

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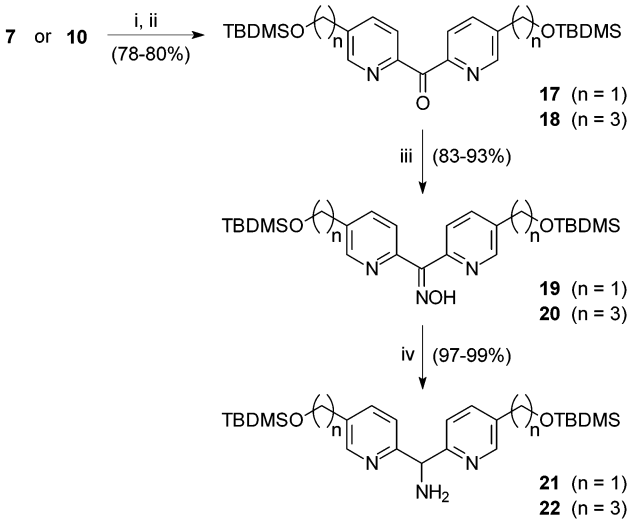
(23) The pure picolyl chlorides **15** and **16** are reasonably stable at room temperature and can be stored under nitrogen at −20 °C for many months.

SCHEME 4[a]



[a] Conditions: (i) (1) *n*-BuLi, THF, −80 °C, 40 min, (2) **11** or **12**, −80 °C, 1 h, (3) H_2O; (ii) MnO_2, $CHCl_3$, reflux, 18 h; (iii) $NH_2OH{\cdot}HCl$, TEA, EtOH, reflux, 1 h; (iv) Zn, NH_4OAc, NH_4OH, EtOH, reflux, 3 h.

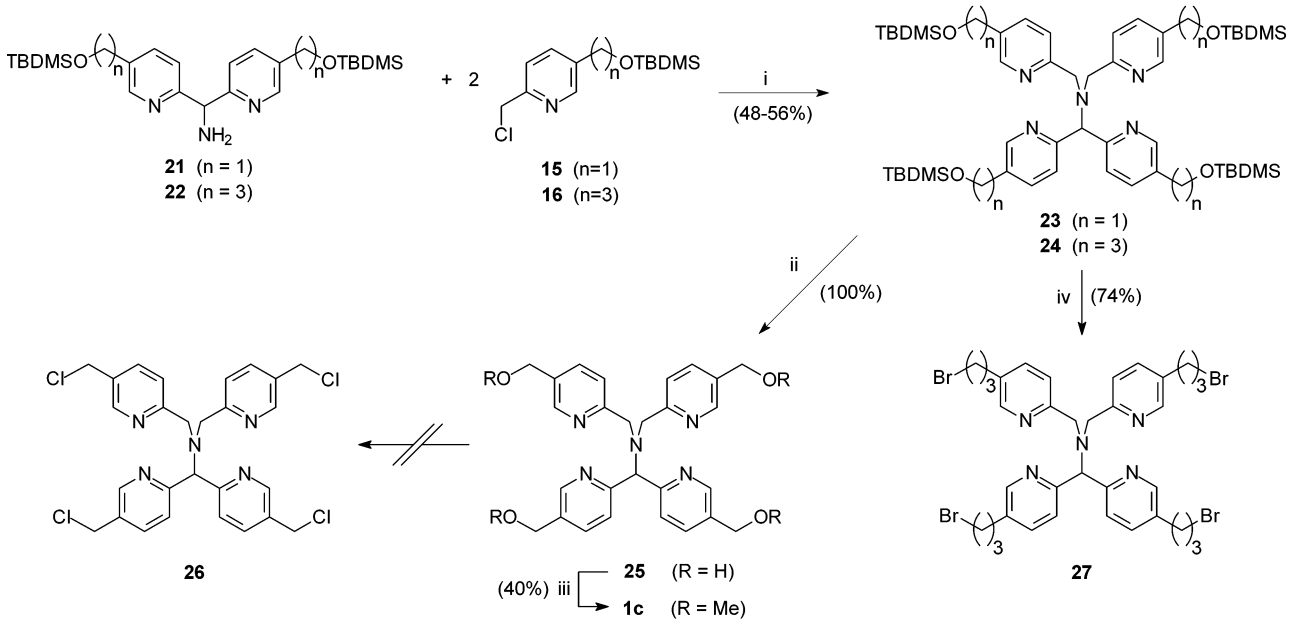
derivatives **11** or **12**, respectively (Scheme 4). The crude intermediate bis(2-pyridinyl)methanols were oxidized by manganese dioxide to furnish bis(2-pyridinyl) ketones **17** and **18** in 78% and 80% yield, respectively, after purification by column chromatography. Next, **17** and **18** were converted to the bis(2-pyridinyl) ketoximes **19** and **20** in 83% and 93% yield, respectively, by a reaction with hydroxylammonium chloride in the presence of TEA. These oximes were then converted to bis(2-pyridinyl)-methylamines **21** and **22** in near-quantitative yields by a mild reduction using zinc powder under basic conditions.[24]

The dialkylation of bis(2-pyridyl)methylamines **21** and **22** with the 2-picolyl chlorides **15** and **16**, respectively, in the presence of cesium carbonate furnished the tetrafunctionalized N4Py derivatives **23** and **24** (Scheme 5). Although similar results were obtained for diisopropylethylamine (DIEA) as the base in acetonitrile, it was found that the use of cesium carbonate resulted in a cleaner reaction. Compounds **23** and **24** were obtained only in moderate yields, partially due to incomplete dialkylation of amines **21** and **22** (even in the presence of an excess of the picolyl chloride derivative) and difficult separation of the mono- and dialkylated products by column chromatography.

To obtain the tetrachloride N4Py derivative **26**, the silyl protecting groups of **23** were first removed by dilute hydrochloric acid in ethanol. Various strategies were investigated to convert **25** to the tetrachloride N4Py derivative **26**, either to be isolated as such or to be converted in situ to a sulfide by reaction with benzyl mercaptan. Tetra-alcohol **25** was treated with (a) thionyl chloride in the presence of TEA in CH_2Cl_2 and pyridine as cosolvent; (b) methanesulfonyl chloride, lithium chloride, and collidine in DMF;[25] or (c) triphenylphosphine and carbon tetrachloride in acetonitrile with or without pyridine as base or cosolvent. These conditions were attempted at various temperatures and with different orders of addition of the reagents, but in all cases the N4Py derivative decomposed and no characterizable products were obtained. Tetrachloride **26** was detected by electrospray ionization mass analysis as its HCl salt from the reaction of **25** with thionyl chloride in DMF at 0 °C.[26] However, this compound decomposed immediately when water or a base was added.

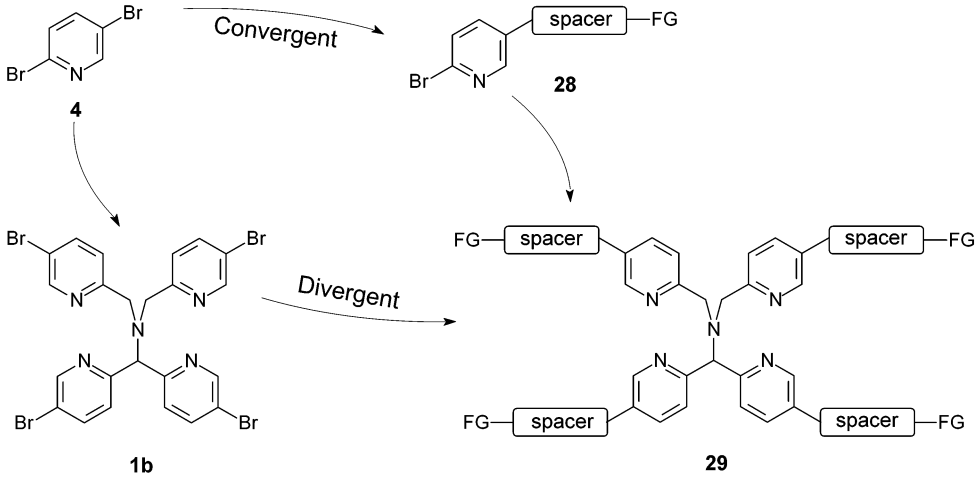
Replacement of the four hydroxyl groups in **25** by (chloride) leaving groups apparently leads to the formation of an intrinsically unstable compound. The observed instability is probably inherent to the structure of **26**. Although at this moment the exact mechanism of decomposition can only be speculated upon, it might be triggered by hydrogen chloride elimination, which is

SCHEME 5[a]



[a] Conditions: (i) Cs_2CO_3, MeCN, reflux, 48 h; (ii) H_3O^+, EtOH, 15 min; (iii) (1) NaH, DMF, 0 °C, 30 min, (2) MeI, 0 °C to rt, 2 h; (iv) Ph_3PBr_2, CH_2Cl_2, rt, 18 h.

SCHEME 6. Convergent vs Divergent Synthesis Routes of Tetrasubstituted N4Py Derivatives



attributed to the double benzylic, tertiary hydrogen in **26** and the reactive 'benzylic' leaving groups.[27,28] In evidence that it is not the structure of **25** that is unstable, this N4Py derivative was successfully converted into the tetramethoxy derivative **1c** by first deprotonating the four hydroxyl groups of **25** and subsequently quenching with methyl iodide (Scheme 5). The rather low 40% yield in this case is due to loss of product during purification by column chromatography.

In view of the seemingly unsolvable problems associated with the instability of the tetrachloride **26**, we decided to increase the N4Py ligand spacer length. A dimethylene (C2) linker was rejected, because HCl elimination to form vinyl-pyridine derivatives can obviously occur. The synthesis of the C3-spacer ligand **27** is outlined in Schemes 2−5 for $n = 3$. To enhance the leaving group ability at the nonbenzylic position, the silyl ether groups in **24** were converted directly into bromides by a reaction with triphenylphosphine dibromide (Scheme 5).[29,30] As a result, the tetrafunctionalized N4Py derivative **27** was obtained in 12 steps from 2,5-dibromopyridine (**4**) in an overall yield of 5%.

Divergent Synthesis Strategy. The problems of instability encountered with **26** and the consequent need to prepare the more stable **27** suggested that a divergent synthetic route, permitting the modification of the N4Py ligand after its construction, would facilitate a versatile preparation of alternative ligand systems. A potential N4Py framework, tetrabromide N4Py derivative **1b**, that could enable a variety of tetrafunctionalized N4Py derivatives **29** to be generated via divergent synthesis is presented in Scheme 6. Again starting from 2,5-dibromopyridine (**4**), tetrabromo N4Py **1b** can be prepared by initially using the bromide at the 2-position of the pyridine ring to construct the core of the N4Py scaffold. The remaining bromides in **1b** are then available for further functionalization by, for instance, metal-catalyzed cross-coupling methodologies or via bromide−metal exchange reactions.

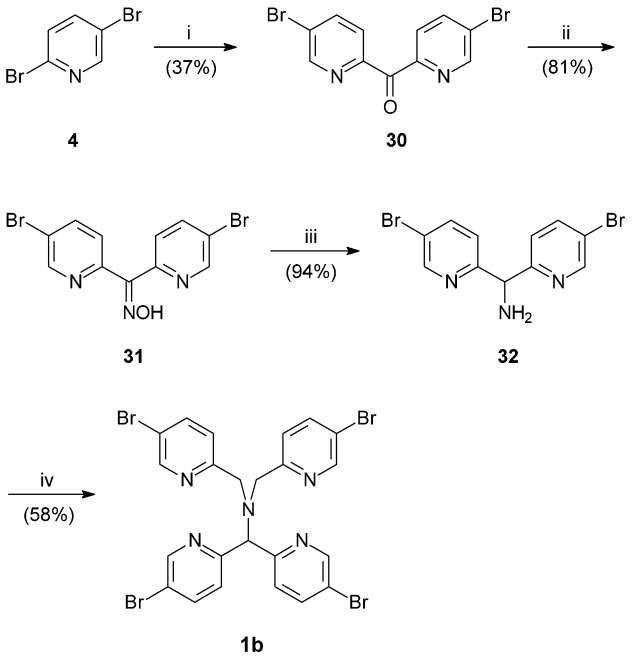
As indicated above, mono-metalation of **4** by *n*-BuLi occurs preferentially at the 5-position in diethyl ether at −80 °C.[31] However, at low concentrations in noncoordinating solvents such as toluene or dichloromethane, lithiation occurs preferentially at the 2-position rather than the 5-position.[21b] This reversed regioselectivity enabled the synthesis of bispyridinyl ketone **30**, which was achieved by quenching the in situ prepared 5-bromo-2-lithiopyridine in toluene at −80 °C with 0.5 equiv of diethyl carbonate (Scheme 7). The low yield is partially due to incomplete regioselectivity under these conditions. Subsequent conversion of the ketone **30** into the bispyridinyl ketoxime **31** by reaction with hydroxylamine, and a highly efficient reduction of the ketoxime **31** using zinc in acetic acid at room temperature provided bispyridinyl methylamine **32** in 94% yield.

The required picolyl chloride derivative **36** can be obtained from 5-bromo-2-picoline[18] (**33**) by oxidation with *m*-CPBA to the *N*-oxide **34** in 98% yield (Scheme 8). Subsequent reaction of **34** with trifluoroacetic acid anhydride (TFAA), and hydrolysis of the resulting acetate furnished 5-bromo-2-(hydroxymethyl)pyridine (**35**) in 81% yield. Finally, this picolyl alcohol was converted with thionyl chloride to **36** in 97% yield.

Analogously to the previously described preparation of N4Py derivatives, the dialkylation of amine **32** with 2 equiv of picolyl chloride **36** in the presence of base enabled the construction of tetrabromo N4Py **1b** (Scheme

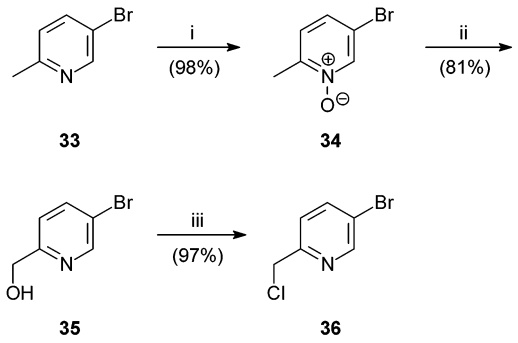
(24) Niemers, E.; Hiltmann, R. *Synthesis* **1976**, 593−595.
(25) Collington, E. W.; Meyers, A. I. *J. Org. Chem.* **1971**, *36*, 3044−3045.
(26) The molecular ion was observed at *m/z* 560.1 $[M + H]^+$ for the main isotope peak, which corresponded to the expected mass of 559.1 $[M]^+$, and the detected isotope distribution pattern matched the calculated arrangement.
(27) A dichloride N4Py derivative was previously prepared as described in ref 16. The two benzylic chlorides were located in the upper half of the N4Py scaffold (see Scheme 5 for compound **26**) and did not prevent its formation. However, in the tetrachloride N4Py derivative **26** the double benzylic proton in the bottom half of the N4Py ligand is now flanked by two pyridine groups that each contain a benzylic chloride moiety.
(28) Attempts to replace the double benzylic proton by a methyl group have been unsuccessful. Lithiation of **23** at −80 °C in THF in the presence of *N*,*N*,*N*′,*N*′-tetramethylethylenediamine (TMEDA) followed by quenching with methyl iodide resulted repeatedly only in the recovery of **23** and not to the desired product. These conditions were successful when applied to N4Py (ref 20).
(29) Despite the more reactive bromine atoms, **27** can be stored under nitrogen at −20 °C for at least 2 months and at room temperature for at least 2 days.
(30) The electrospray mass analysis of **27** ($[M + H]^+$ at *m/z* 848.0) corresponded to the calculated isotope distribution pattern.
(31) Parham, W. E.; Piccirilli, R. M. *J. Org. Chem.* **1977**, *42*, 257−260.

SCHEME 7[a]



[a] Conditions: (i) (1) *n*-BuLi, toluene, −80 °C, 2 h, (2) $(EtO)_2CO$, −80 °C to rt; (ii) $NH_2OH{\cdot}HCl$, py, EtOH, reflux, 1.5 h; (iii) Zn, AcOH, rt, 30 min; (iv) **36**, DIEA, MeCN, reflux, 48 h.

SCHEME 8[a]

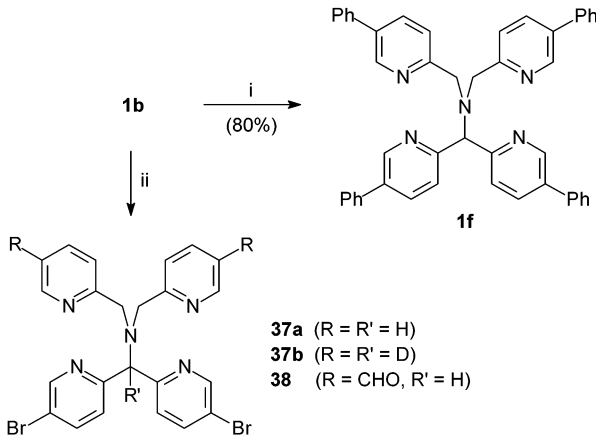


[a] Conditions: (i) *m*-CPBA, CH_2Cl_2, rt, 18 h; (ii) (1) TFAA, (2) $NaHCO_3$ (aq); (iii) $SOCl_2$, CH_2Cl_2, 0 °C to rt, 18 h.

7). The best results were obtained by heating at reflux for 2 days in the presence of a large excess of DIEA and a minimal amount of acetonitrile to dissolve the reactants. After purification of the crude product by short path flash column chromatography, traces of monoalkylated product could be removed by precipitation of the product in diethyl ether to afford **1b** in 58% yield (16% yield from **4**).

Functionalization of Tetrabromo N4Py 1b. For the functionalization of tetrabromo N4Py **1b** via cross-coupling reactions we have focused on the Suzuki−Miyaura reaction.[32,33] Heating **1b** for 16 h with 10 equivalents of phenylboronic acid and 12 mol % $Pd(PPh_3)_4$ as the catalyst (i.e., 3 mol % of palladium catalyst per bromide) in a mixture of toluene and aqueous sodium

SCHEME 9[a]



[a] Conditions: (i) $PhB(OH)_2$, $Pd(PPh_3)_4$, Na_2CO_3 (aq), toluene, reflux, 18 h; (ii) (1) *i*-PrMgBr, THF, rt to reflux, 1 h, (2) H_2O, D_2O, or DMF.

carbonate afforded the cross-coupled product **1f** in 80% yield (Scheme 9). ^1H NMR and mass analysis of the crude product indicated that the tetrasubstituted cross-coupled product was obtained with no detectable amounts of mono-, di-, or trisubstituted products.[34]

To test the susceptibility of tetrabromo N4Py **1b** toward halogen−metal exchange and subsequent quenching with electrophiles, **1b** was treated at room temperature with 4.5 equiv of *i*-PrMgBr as a solution in THF (Scheme 9). Mass analysis of a product sample, obtained after quenching with water, revealed the presence of only the dibromide N4Py derivative **37a**. The addition of more *i*-PrMgBr did not result in a complete magnesium−bromide exchange, even after heating at reflux temperature. When a sample was quenched with D_2O, mass analysis revealed the incorporation of three deuterium atoms with two bromine atoms still present, which suggested the formation of **37b**. Not only does this indicate that the double benzylic position in **1b** is deprotonated, but also that magnesium−bromide exchange occurs only at two of the four sites. The use of DMF as an electrophile afforded the dialdehyde derivative **38**, which has been characterized by COSY NMR experiments and mass analysis.[35] The anion at the double benzylic position apparently does not react with DMF, and is therefore reprotonated during workup. These findings indicate that the bottom half of **1b** is not susceptible toward a metal−halogen exchange, probably due to the delocalization of the negative charge at the

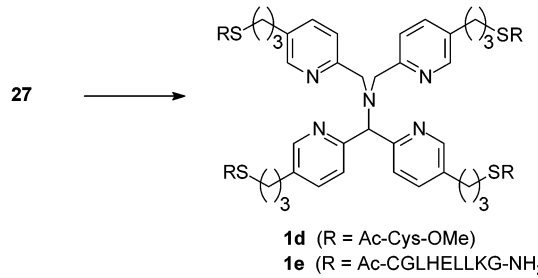
(32) (a) Miyaura, N.; Suzuki, A. *Chem. Rev.* **1995**, *95*, 2457−2483. (b) Suzuki, A. In *Metal-catalyzed Cross-coupling Reactions*; Diederich, F., Stang, P. J., Eds.; Wiley-VCH Verlag GmbH: Germany, 1998; pp 49−97. (c) Suzuki, A. *J. Organomet. Chem.* **1999**, *576*, 147−168.

(33) For bromopyridines in Suzuki reactions, see: (a) Thompson, W. J.; Gaudino, J. *J. Org. Chem.* **1984**, *49*, 5237−5243. (b) Stavenuiter, J.; Hamzink, M.; Van der Hulst, R.; Zomer, G.; Westra, G.; Kriek, E. *Heterocycles* **1987**, *26*, 2711−2716. (c) Thompson, W. J.; Jones, J. H.; Lyke, P. A.; Thies, J. E. *J. Org. Chem.* **1988**, *53*, 2052−2055. (d) Aliprantis, A. O.; Canary, J. W. *J. Am. Chem. Soc.* **1994**, *116*, 6985−6986. (e) Gong, Y.; Pauls, H. W. *Synlett* **2000**, 829−831.

(34) In test reactions carried out on several bromopyridines, it was established that THF as a solvent generally resulted in superior efficiencies over toluene. However, when toluene was replaced by THF as the solvent for the Suzuki reaction with **1b**, only a single, structurally related, but unidentified compound was obtained within a few minutes as observed by ^1H NMR, without any trace of the desired product **1f**. Presumably, a very reactive species is formed under these conditions, which results in the formation of this undesired product.

(35) The observed fragmentation with *m*/*z* 329, 254, and 122 can only be ascribed to the proposed structure of **38** and not its regioisomer.

SCHEME 10[a]



[a] Conditions: *N*-Ac-Cys-OMe or peptide *N*-Ac-CGLHELLKG, Cs_2CO_3 (aq), pH 9, DMF, rt, 18 h.

double benzylic anion over the two adjacent pyridine rings. Incidentally, the very selective dimetalation of **1b** with *i*-PrMgBr provides the opportunity to introduce two different sets of spacers at the periphery of the N4Py ligand: after quenching the dimetalated species of **1b** with an appropriate electrophile, the two remaining bromides in the resulting product can be submitted to further functionalization by cross-coupling reactions.

Peptide Coupling. Using our previously developed methodology for coupling unprotected single-cysteine peptides to the ligand via halogen displacement by the cysteine thiolate,[16] tetracysteine derivative **1d** was prepared by reaction of **27** with methyl *N*-acetylcysteine and purified by reversed-phase HPLC. Similarly, the model nonapeptide Ac-Cys-Gly-Leu-His-Glu-Leu-Leu-Lys-Gly-NH_2 was coupled quantitatively to the tetrabromide template **27** to provide **1e** (Scheme 10).[36] It should be noted that the sequence of the unprotected test peptide was chosen only for its representative content of nucleophilic groups in the amino acids side chains that might interfere with the halogen displacement, and was not designed to fold into a four-helix bundle.

Synthesis and Characterization of Iron Complexes of N4Py Derivatives. By addition of iron(II) perchlorate to the pentadentate ligands **1a**−**f** in acetonitrile the corresponding iron complexes **2a**−**f** were obtained (Scheme 1). The ^{1}H NMR spectra of **2b** and **2c** in CD_3CN revealed signals in the diamagnetic range of 0−10 ppm, which is distinctive for low-spin iron(II) complexes of N4Py ligands.[12,20] The electrospray ionization mass analyses (ESI-MS) were also in accordance with the postulated composition of the complexes **2b** and **2c**.

Upon complexation of the tetrapeptide-bound N4Py ligand **1e** with iron(II) perchlorate, the metalated complex **2e** was obtained in 18% yield after preparative reversed-phase HPLC. Its chemical integrity was verified by MALDI mass analysis. The calculated mass for **2e** corresponded to the detected mass (calcd for $[M - 2(ClO_4)^- - (MeCN)]^{2+}$ *m*/*z* 6121.3 amu (monoisotopic), 6125.1 amu (average isotope composition), found 6124.0 amu). The UV−vis absorption bands of **2b**−**d** correlate well with those of the previously prepared low-spin iron(II) complexes of N4Py derivatives, with maximum absorptions typically around 380 and 460 nm (Table 2).[12,20] For **2d**, the pH appeared to have only a minor effect on its extinction coefficient (λ_{max} 452 nm, $\epsilon_{pH\ 4}$ 35 × 10^2 M^{-1} cm^{-1}, $\epsilon_{pH\ 7}$ 38 × 10^2 M^{-1} cm^{-1}, $\epsilon_{pH\ 10}$ 33 × 10^2 M^{-1} cm^{-1}) at room temperature.

TABLE 1. Selected Bond Distances for 2b and 2c[a,b]

bond (Å)		**2a**[12]	**2b**	**2c**
$Fe-N_{amine}$	N1	1.961(3)	1.965(3)	1.958(3)
$Fe-N_{py}$	N2	1.976(3)	1.965(3)	1.957(3)
	N3	1.967(3)	1.956(3)	1.960(3)
	N4	1.968(3)	1.962(3)	1.960(3)
	N5	1.975(3)	1.970(3)	1.967(3)
$Fe-N_{MeCN}$	N6	1.915(3)	1.927(4)	1.928(3)
$Fe-N_{py-mean\ plane}$		0.207(5)	0.207(1)	0.197(1)

[a] Standard deviations are given in parentheses. [b] X-ray diffraction of **2a** was performed at 130 K, **2b** at 90 K, and **2c** at 100 K.

TABLE 2. Spectroscopic Data of the Fe^{II}- and $Fe^{III}OOH$ Complexes of N4Py (1a) and Its Derivatives 1b−d in Acetone at Room Temperature[a]

	$[(L)Fe^{II}(MeCN)](ClO_4)_2$		$[(L)Fe^{III}OOH](ClO_4)_2$	
ligand (L)	λ_{max} (nm)	ϵ (× 10^2 M^{-1} cm^{-1})	λ_{max} (nm)	ϵ (× 10^2 M^{-1} cm^{-1})
1a	457 (455)	53 (48)	539 (520)	11 (7.2)
1b	464 (460)	70 (88)	553 (544)	11 (8.9)
1c	461 (457)	41 (59)	538 (522)	10 (7.8)
1d[b]	456 (n.d.)	37 (n.d.)	525 (n.d.)	3.4 (n.d.)

[a] The results in parentheses refer to acetonitrile as solvent. n.d. = not determined. [b] At 0 °C, $\lambda_{max,\mathbf{2d}}$ 457 nm, ϵ 26 × 10^2 M^{-1} cm^{-1}; $\lambda_{max,\mathbf{3d}}$ 531 nm, ϵ 3.7 × 10^2 M^{-1} cm^{-1}.

Crystals of the iron(II) complexes **2b** and **2c** suitable for X-ray analysis were obtained by the slow diffusion of ethyl acetate into their solutions in acetonitrile. The molecular structures are shown in Figure 2. Five coordination sites to the iron center are occupied by the chelating ligand and a solvent-derived acetonitrile molecule functions as an axial ligand. This mode of coordination to the metal ion is identical to that previously observed for N4PyFe **2a**.[12,13b] As can be seen in Figure 2, the bromine atoms in **2b** and the methoxymethyl groups in **2c** are directed away from the iron center in the complex. The same spatial arrangement can, therefore, also be predicted for the iron-complexes of other tetrasubstituted N4Py ligands with functional groups at the same positions, such as in **2d** and **2e**. Although the tetraphenyl N4Py derivative **1f** could be complexed successfully with iron perchlorate in acetonitrile,[37] no suitable crystals of the iron complex could be obtained for X-ray diffraction.

The iron−nitrogen bond lengths of **2b** and **2c** are in the range of 1.91−1.98 Å, which is typical in bond lengths for low-spin iron(II) centers.[38] Complexes **2b** and **2c** show only a small variation when compared to those found for N4PyFe **2a** (Table 1).[12,13b] Electronic effects alone are, in our opinion, insufficient to explain these features. Therefore, these minor differences in bond lengths in **2a**−**c** are most likely due to the differences in temperature during the X-ray analysis (see footnote in Table 1).

(36) MALDI analysis of the crude iron(II) complex of the peptide coupled N4Py ligand revealed only the tetrasubstituted product. The masses for mono-, di-, or trisubstituted peptide complexes were not observed.

(37) Formation of the iron complex $[(\mathbf{1f})Fe^{II}(MeCN)](ClO_4)_2$ (**2f**) was confirmed by ESI-MS analysis of the dark purple solid that was obtained after evaporation of the solvent.

(38) Hawker, P. N.; Twigg, M. V. In *Comprehensive Coordination Chemistry*; Wilkinson, G., Ed.; Pergamon: Oxford, 1987; Vol. IV, p 1179.

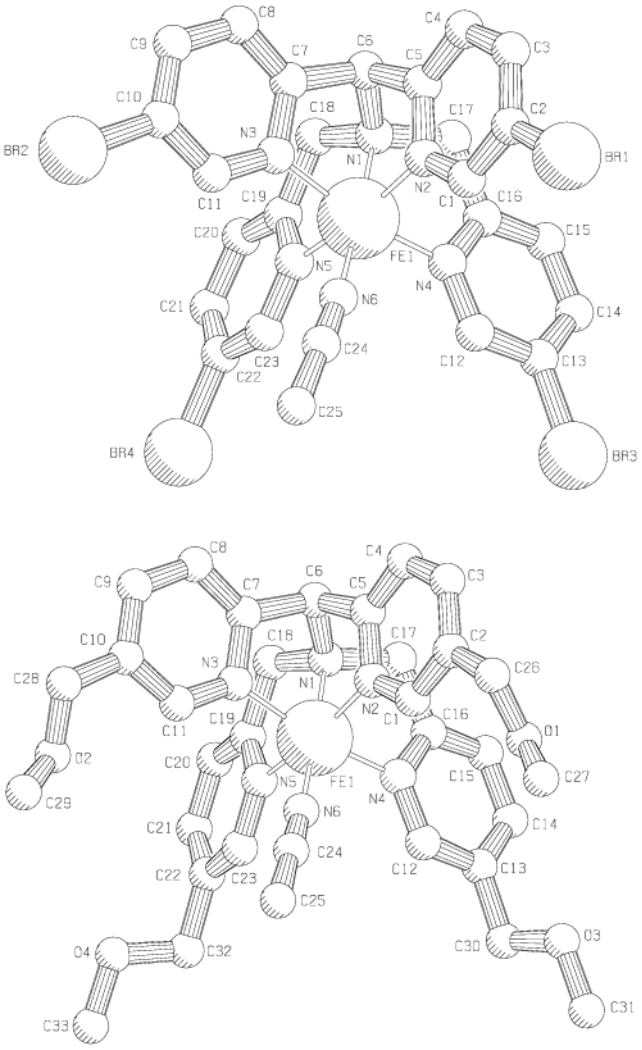


FIGURE 2. Pluto representation of **2b** (top) and **2c** (bottom). Hydrogen atoms and counterions are omitted for clarity.

The cyclic voltammetry measurements for **2b** in acetonitrile displayed a reversible oxidation wave with a redox potential for the Fe^{II}/Fe^{III} couple of 1198 mV vs SCE and an observed peak-to-peak separation of 105 mV. Forward and reverse differential pulse voltammetry at a scan rate of 10 mV s^{-1} confirmed this process to be reversible with a redox potential for the Fe^{II}/Fe^{III} couple of 1196 mV vs SCE. The redox potential of the tetrabromo N4PyFe complex **2b** was found to be substantially higher than that of N4PyFe **2a** ($E_{1/2}$ = 1010 mV vs SCE)[12] and a dibromo N4PyFe derivative[20,39] ($E_{1/2}$ = 1100 mV vs SCE). The general trend infers that the introduction of a greater number of electron-withdrawing groups into the N4Py ligand results in an increase in the redox potential of the corresponding iron(II) complex. This in turn gives rise to the observed relatively large red shift for the Fe^{III}-OOH species **3b** (vide infra).

Formation of Iron(III) Hydroperoxo Species. Upon the addition of excess hydrogen peroxide to **2a**−**e**, the iron complexes are converted to the corresponding purple iron(III) hydroperoxo species **3a**−**e** (Scheme 1). The UV-vis absorption data are compiled in Table 2. The nature of the $Fe^{III}OOH$ species was previously elucidated for N4PyFe **2a**.[13b] Based on the UV−vis data, we propose similar $Fe^{III}OOH$ structures for complexes **3b**−**e**. The mechanism of its formation and further reactions are also considered to be applicable to the new N4Py complexes described here. The complex **2d** was prepared as a model for peptide-bound N4PyFe complexes such as **2e** in order to study the stability of the complex and in particular its sulfide linkage toward the applied oxidative conditions. However, the $Fe^{III}OOH$ species **3d** was found to be considerably less stable than **3a**, and could not be detected by a stepwise titration of the iron(II) complex with hydrogen peroxide in acetone at room temperature. Conversely, the addition of a single aliquot of excess hydrogen peroxide to **2d** resulted in the formation and observation of the hydroperoxo species **3d** (∼2 min lifetime). Its lifetime was significantly extended to approximately 15 min by reducing the temperature of the solution to 0 °C.

Oxidation Activity of Iron(II) Complexes of N4Py and Derivatives. The peroxidase activity and the pH dependency of N4PyFe **2a** and complex **2e** were determined by monitoring spectrophotometrically the formation of the $ABTS^{\bullet+}$ radical cation[40] over time at pH 3.5 (ϵ_{660} 1.47 × 10^4 M^{-1} cm^{-1}) (Figure 3).

This probe had previously been used to establish the peroxidase-like activity of the dipeptide-bound N4PyFe catalyst.[16] Under these conditions complex **2a** is deactivated within 200 s, due to an oxidative degradation of the complex, which is attributed to hydroxyl radicals formed during the reaction.[14b,20] Indeed, the addition of a radical scavenger (2,4,6-tri-*tert*-butylphenol) increased the lifetime of the catalyst, but reduced the reaction rate of the ABTS oxidation. When compared to N4PyFe **2a**, the N4PyFe-peptide complex **2e** displayed a substantially lower activity for the peroxidation of ABTS under identical conditions (4.4 turnovers for **2e** vs 27 for **2a** at t = 50 s, Figure 3). This result may be attributed to steric interactions between the unfolded peptide chains on the catalyst and the ABTS substrate. Note that this is in contrast to our earlier findings[16] when two unfolded peptides, covalently attached to N4PyFe, had no effect on the catalyst's activity.

Conclusions

The synthesis of the tetrabromide N4Py derivative **1b** could be accomplished by taking advantage of a change in regioselectivity in the lithiation of 2,5-dibromopyridine by substituting a coordinating solvent by a noncoordinating solvent. This enabled the construction of the N4Py core while preserving the remaining bromide on the pyridine ring for further functionalization of the final ligand **1b**. The potential of this tetrabromo N4Py derivative to be functionalized further by cross-coupling methodologies was demonstrated by the successful preparation of tetraphenyl N4Py **1f**. This reaction entails four palladium-mediated Suzuki reactions on a single molecule with no detectable products from incomplete cross-coupling reactions on **1b**. In view of the positive results in the Suzuki reaction with **1b** it is very likely that other

(39) The iron(II) comlex of *N*,*N*-bis[(5-bromo-2-pyridinyl)methyl]-*N*-[di(2-pyridinyl)methyl]amine.

(40) Adams, P. A. *J. Chem. Soc., Perkin Trans. 2* **1990**, 1407−1414.

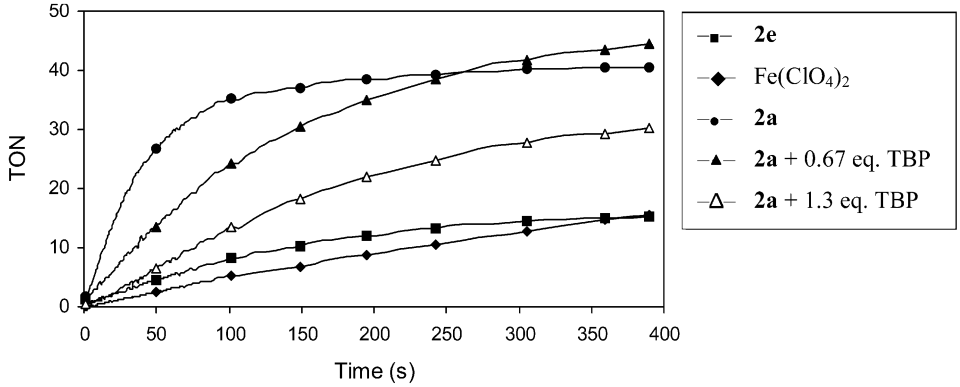


FIGURE 3. Formation of the $ABTS^{\bullet+}$ radical cation over time at 20 °C, expressed in turnover numbers (TON): background corrected oxidation of ABTS by **2a**, **2e**, and iron(II) perchlorate at pH 3.5 and by **2a** in the presence of the radical scavenger 2,4,6-tri-*tert*-butylphenol (TBP).

cross-coupling reactions can also be applied to provide a range of tetrasubstituted N4Py derivatives.

All described iron complexes of tetrafunctionalized N4Py derivatives show spectroscopic data that are characteristic for low-spin iron(II) complexes. X-ray crystallographic data of **2b** and **2c** have revealed that the four functionalities at the periphery of the N4Py ligand are directed away from the iron metal center. A similar orientation can therefore be expected for **2e** and other peptide-bound N4PyFe complexes. Complexes **2b** and **2c** also show a catalytic activity comparable to **2a** in the oxidation of organic compounds with hydrogen peroxide.[41]

After the successful preparation of several tetrasubstituted N4Py derivatives one, **27**, has been used as a functional template for the covalent attachment of four unprotected peptides. Although the C3-spacer on **27** is too long to be compatible with a folded four-helix bundle, the work described here provides a foundation for synthesizing alternative derivatives of the ligand.

Experimental Section

Representative Procedure for the Preparation of TBDMS Ethers. To a stirred solution of **6** (21.8 g, 116 mmol) and TEA (25.0 mL, 179 mmol, 1.5 equiv) in dry DMF (50 mL) was added *tert*-butyldimethylsilyl chloride (18.8 g, 125 mmol, 1.05 equiv). After being stirred for 15 min, protected from moisture, the mixture was diluted with diethyl ether and washed with water and brine. The organic phase was dried over Na_2SO_4 and concentrated by rotary evaporation to yield a yellow/brown oil (35.0 g, quant). Purification by flash column chromatography over silica using a gradient of hexane/diethyl ether (19:1 to 9:1) yielded 29.8 g of **7** as colorless oil.

2-Bromo-5-[[[*tert*-butyl(dimethyl)silyl]oxy]methyl]pyridine (7). Yield: 85%. R_f = 0.43 (SiO_2, hexane/diethyl ether = 9:1). ^{1}H NMR ($CDCl_3$, 300 MHz): δ 0.09 (s, 6H), 0.92 (s, 9H), 4.69 (s, 2H), 7.43 (d, J = 8.1 Hz, 1H), 7.52 (dd, J = 8.1, 2.2 Hz, 1H), 8.30 (d, J = 2.2 Hz, 1H). ^{13}C NMR ($CDCl_3$, 75.5 MHz): δ −5.4, 18.2, 25.7, 61.9, 127.5, 136.0, 136.5, 140.4, 148.0. HRMS: calcd for $C_{12}H_{20}BrNOSi$ 303.048, found 303.048.

Ethyl (*E*)-3-(6-Bromo-3-pyridinyl)-2-propenoate (8). A solution of triethyl phosphonatoacetate (12.0 g, 53.6 mmol) in dry diethyl ether (50 mL) was added over 10 min to a suspension of NaH (2.35 g, 58.8 mmol) in dry diethyl ether (130 mL) at 5 °C. The icebath was removed, and after 10 min a solution of **5** (9.97 g, 53.6 mmol) in dry THF/diethyl ether (250 mL, 1:1) was added over 10 min. After 15 min, water was added and the phases were separated. The aqueous layer was extracted with diethyl ether. The combined organic layer was washed with brine and dried over Na_2SO_4. Evaporation of the solvent produced **8** as a brown solid. Recrystallization from hexane yielded analytically pure product as white needles (9.96 g pure *E*-isomer, 75%). From the concentrated filtrate a further 1.24 g of **8** (85% combined yield) was obtained after flash column chromatography over silica using hexane/diethyl ether (9:1) as the eluent. R_f = 0.53 (SiO_2, hexane/ethyl acetate = 3:1). Mp 83.6−84.1 °C. ^{1}H NMR ($CDCl_3$, 300 MHz) δ 1.33 (t, J = 7.3 Hz, 3H), 4.26 (q, J = 7.3 Hz, 2H), 6.48 (d, J = 16.1 Hz, 1H), 7.50 (d, J = 8.4 Hz, 1H), 7.59 (d, J = 16.1 Hz, 1H), 7.67 (dd, J = 8.4, 2.2 Hz, 1H), 8.47 (d, J = 2.2 Hz, 1H). ^{13}C NMR ($CDCl_3$, 50.3 MHz) δ 14.2, 60.9, 121.2, 128.3, 129.5, 136.2, 139.3, 143.4, 149.8, 166.0. HRMS: calcd for $C_{10}H_{10}NO_2Br$ 254.989, found 254.990. Anal. Calcd for $C_{10}H_{10}BrNO_2$: C, 46.90; H, 3.90; N, 5.50; Br, 31.20. Found: C, 46.96; H, 3.97; N, 5.43; Br, 31.18.

3-(6-Bromo-3-pyridinyl)-1-propanol (9). To a stirred suspension of $LiAlH_4$ (1.90 g, 50.1 mmol) in dry diethyl ether (75 mL) at 0 °C was added a solution of **8** (6.40 g, 25.0 mmol) in dry diethyl ether (75 mL) over 20 min. After 10 min at 0 °C, the reaction mixture was allowed to warm to room temperature and stirred for 45 min. The reaction was quenched by carefully adding $Na_2SO_4 \cdot 10H_2O$ and stirring the mixture for 1 h. The solids were removed by filtration and washed with diethyl ether. Evaporation of the solvent yielded **9** as a light brown oil (3.27 g, 61%), which was used without any further purification. R_f = 0.29 (SiO_2, hexane/ethyl acetate = 1:1). ^{1}H NMR ($CDCl_3$, 300 MHz): δ 1.84 (m, 2H), 2.68 (t, J = 7.9 Hz, 2H), 3.66 (t, J = 6.0 Hz, 2H), 7.38 (s, 1H), 7.39 (s, 1H), 8.19 (s, 1H). ^{13}C NMR ($CDCl_3$, 75.5 MHz): δ 28.2, 33.3, 60.9, 127.6, 136.7, 138.8, 139.0, 149.8. HRMS: calcd for $C_8H_{10}NOBr$ 214.995, found 214.996.

Representative Procedures for the Preparation of 2-Pyridinecarbaldehydes. Method A. To a solution of **7** (10.4 g, 34.3 mmol) in dry THF (450 mL) at −80 °C under a nitrogen atmosphere was slowly added *n*-BuLi (24.0 mL, 38.4 mmol, 1.1 equiv). After 40 min at −80 °C, dry DMF (3.0 mL, 39 mmol, 1.1 equiv) was added to the red solution. The reaction mixture was allowed to warm to room temperature after 1 h at −80 °C. Brine and diethyl ether were added, and the layers were separated. The aqueous layer was extracted with diethyl ether. The combined organic layers were washed with brine and dried over Na_2SO_4. Concentration by rotary evaporation afforded a light brown oil (8.98 g). Purification by flash column chromatography over silica using a gradient of hexane/diethyl ether (9:1 to 3:1) yielded 6.88 g of **11** as a light brown oil.

Method B. To a solution of isopropylmagnesium bromide (11 mL, 22 mmol, 2 M in THF) in dry THF (10 mL) was added

(41) van den Heuvel, M. In *Non-Haem Ligands as Functional Templates for Peptide Attachment*. Ph.D. Thesis, Groningen, The Netherlands, 2002 (http://www.ub.rug.nl/eldoc/dis/science/m.van.den.heuvel/).

7 (6.02 g, 19.9 mmol). Heat was evolved during the reaction, and after 1 h dry DMF (1.7 mL, 22 mmol) was added. The reaction was quenched after an additional 1 h by the addition of a saturated aqueous NH_4Cl solution (20 mL). Diethyl ether was added, and the layers were separated. The aqueous layer was extracted with diethyl ether. The combined organic layers were washed with water and brine and dried over Na_2SO_4. Evaporation of the solvent furnished an orange/brown oil (5.12 g).

5-[[[*tert*-Butyl(dimethyl)silyl]oxy]methyl]-2-pyridinecarbaldehyde (11). Yield: 80%. $R_f = 0.54$ (SiO_2, hexane/ethyl acetate = 3:1). ^{1}H NMR ($CDCl_3$, 300 MHz): δ 0.12 (s, 6H), 0.94 (s, 9H), 4.84 (s, 2H), 7.82 (d, J = 8.1 Hz, 1H), 7.94 (d, J = 8.1 Hz, 1H), 8.72 (s, 1H), 10.06 (s, 1H). ^{13}C NMR ($CDCl_3$, 75.5 MHz): δ −5.4, 18.2, 25.8, 62.4, 121.9, 134.4, 141.7, 148.0, 151.8, 193.1. HRMS: calcd for $C_{13}H_{21}NO_2Si$ 251.134, found 251.133.

Representative Procedure for $NaBH_4$ Reductions. To a solution of **11** (5.87 g, 23.3 mmol) in methanol (90 mL) at 0 °C was slowly added $NaBH_4$ (0.44 g, 12 mmol, 0.5 equiv). The reaction mixture was allowed to warm to room temperature, and after 30 min dilute HCl (2 M) was added until pH 7, followed by a saturated aqueous Na_2CO_3 solution. The solvent was evaporated, and the residue was extracted with diethyl ether. The organic layer was washed with brine, dried over Na_2SO_4, and concentrated by rotary evaporation to yield **13** as a light yellow oil (5.73 g, 97%). The crude product was used without any further purification.

[5-[[[*tert*-Butyl(dimethyl)silyl]oxy]methyl]-2-pyridinyl]-methanol (13). ^{1}H NMR ($CDCl_3$, 300 MHz): δ 0.09 (s, 6H), 0.92 (s, 9H), 3.93 (br•s, 1H), 4.74 (s, 4H), 7.21 (d, J = 8.1 Hz, 1H), 7.64 (d, J = 8.1 Hz, 1 H), 8.48 (s, 1H). ^{13}C NMR ($CDCl_3$, 75.5 MHz): δ −5.3, 18.3, 25.8, 62.5, 64.1, 120.2, 134.9, 135.3, 146.5, 158.1. HRMS: calcd for $C_{13}H_{23}NO_2Si$ 253.150, found 253.149.

Representative Procedure for the Preparation of 2-Picolyl Chlorides. To a solution of **13** (5.98 g, 23.6 mmol) and TEA (6.6 mL, 47 mmol, 2 equiv) in dry CH_2Cl_2 (100 mL) at 0 °C under a nitrogen atmosphere was slowly added $SOCl_2$ (2.6 mL, 35 mmol, 1.5 equiv). The reaction mixture was allowed to warm to room temperature and stirred overnight. Saturated aqueous $NaHCO_3$ solution was added. After separation, the aqueous layer was extracted with CH_2Cl_2. The combined organic layers were washed with brine and dried over Na_2SO_4, and the solvent was evaporated by rotary evaporation. The product was obtained as a dark red/brown oil (6.08 g, 95%). The crude product was purified over alumina using hexane/ethyl acetate/TEA (200:9:1) as the eluent to provide 4.24 g of pure **15**.

5-[[[*tert*-Butyl(dimethyl)silyl]oxy]methyl]-2-(chloromethyl)pyridine (15). Yield: 66%. $R_f = 0.32$ (Al_2O_3, hexane/diethyl ether = 9:1). ^{1}H NMR ($CDCl_3$, 300 MHz): δ 0.10 (s, 6H), 0.93 (s, 9H), 4.66 (s, 2H), 4.75 (s, 2H), 7.43 (d, J = 8.1 Hz, 1H), 7.69 (dd, J = 8.1, 2.2 Hz, 1H), 8.51 (d, J = 2.2 Hz, 1H). ^{13}C NMR ($CDCl_3$, 75.5 MHz): δ −5.4, 18.2, 25.8, 47.5, 62.5, 120.7, 134.5, 134.6, 147.3, 160.5. HRMS calcd for $C_{13}H_{22}ClNOSi$ 271.116, found 271.118.

Representative Procedure for the Preparation of Dipyridyl Ketones. To a solution of **7** (6.36 g, 21.0 mmol) in dry THF (250 mL) at −80 °C was added *n*-BuLi (14.0 mL, 22.5 mmol, 1.07 equiv) under a nitrogen atmosphere. After 40 min at −80 °C, a solution of **11** (5.29 g, 21.0 mmol) in dry THF (50 mL) was added to the red reaction mixture. The blue/purple solution was stirred at −80 °C for 1 h and then allowed to warm to room temperature. Brine and diethyl ether were added, and the layers were separated. The aqueous layer was extracted with diethyl ether. The combined organic layers were washed with brine, dried over Na_2SO_4, and concentrated by rotary evaporation to yield an orange/brown oil. This residue was then dissolved in chloroform (130 mL). Activated MnO_2 (4.4 g, 43 mmol) was added, and the suspension was heated under reflux. At 1 h intervals, 13.2 g MnO_2 was added in portions of 4.4 g. The reaction mixture was refluxed overnight. After evaporation of the solvent the residue was filtered through Celite and washed with diethyl ether. The filtrate was dried over Na_2SO_4 and concentrated by rotary evaporation to yield a brown solid (8.97 g, 90%), which was purified over silica using hexane/ethyl acetate (5:1) as the eluent.

Bis[5-[[[*tert*-butyl(dimethyl)silyl]oxy]methyl]-2-pyridinyl]methanone (17). Yield: 78%. $R_f = 0.40$ (SiO_2, hexane/ethyl acetate = 3:1). ^{1}H NMR ($CDCl_3$, 300 MHz): δ 0.12 (s, 12H), 0.94 (s, 18H), 4.84 (s, 4H), 7.84 (d, J = 8.1 Hz, 2H), 8.08 (d, J = 8.1 Hz, 2H), 8.69 (s, 2H). ^{13}C NMR ($CDCl_3$, 75.5 MHz): δ −5.4, 18.2, 25.8, 62.4, 124.9, 134.0, 139.8, 147.0, 153.2, 192.5. HRMS: calcd for $C_{25}H_{40}N_2O_3Si_2$ 472.258, found 472.258.

Representative Procedure for the Preparation of Dipyridylketoximes. A mixture of hydroxylammonium chloride (2.60 g, 37.4 mmol, 2 equiv) and TEA (7.8 mL, 56 mmol, 3 equiv) in ethanol (50 mL) was stirred at room temperature for 0.5 h. The salts were removed by filtration, and the filtrate was added to **17** (8.84 g, 18.7 mmol). After heating under reflux for 1 h, the solvent was removed by rotary evaporation, and the residue was extracted with ethyl acetate. The organic layer was washed with brine, dried over Na_2SO_4, and concentrated to give a brown oil (7.51 g) that solidified on standing. Analytically pure product was obtained by recrystallization from ethanol/water to yield **19** as white plates.

Bis[5-[[[*tert*-butyl(dimethyl)silyl]oxy]methyl]-2-pyridinyl]methanone Oxime (19). Yield: 83%. Mp: 106.3−107.4 °C. ^{1}H NMR ($CDCl_3$, 300 MHz): δ 0.11 (s, 6H), 0.12 (s, 6H), 0.93 (s, 9H), 0.94 (s, 9H), 4.81 (s, 4H), 7.64 (d, J = 8.4 Hz, 1H), 7.75 (dd, J = 8.1, 2.0 Hz, 1H), 7.80 (dd, J = 8.4, 2.0 Hz, 2H), 8.55 (s, 1H), 8.59 (s, 1H). ^{13}C NMR ($CDCl_3$, 75.5 MHz): δ −5.4, 5.3, 18.3, 25.8, 25.9, 62.3, 62.6, 123.8, 124.7, 134.9, 135.2, 136.6, 138.4, 143.6, 146.5, 150.0, 150.1, 153.5. HRMS: calcd for $C_{25}H_{41}N_3O_3Si_2$ 487.269, found 487.268. Anal. Calcd for $C_{25}H_{41}N_3O_3Si_2$: C, 61.56; H, 8.47; N, 8.61. Found: C, 61.48; H, 8.58; N, 8.50.

Representative Procedure for the Preparation of Dipyridylmethylamines. A mixture of **19** (2.00 g, 4.10 mmol) and NH_4OAc (0.42 g, 5.5 mmol, 1.3 equiv) in ethanol (25 mL) and NH_4OH (15 mL, 25% v/v) was heated under reflux. Zinc powder (1.34 g, 20.5 mmol, 5 equiv) was added in portions through the condenser at 15 min intervals over 2 h. After refluxing for an additional 1 h, the reaction mixture was cooled to room temperature and concentrated NaOH was added until pH∼12 was reached. After filtration through Celite and washing with diethyl ether the layers were separated. The aqueous layer was extracted with diethyl ether. The organic phase was washed with brine, dried over Na_2SO_4, and concentrated in vacuo to yield 1.91 g of pure **21** as a green/brown viscous oil.

Bis[5-[[[*tert*-butyl(dimethyl)silyl]oxy]methyl]-2-pyridinyl]methylamine (21). Yield: 99%. ^{1}H NMR ($CDCl_3$, 300 MHz): δ 0.06 (s, 12H), 0.90 (s, 18H), 2.34 (br•s, 2H), 4.70 (s, 4H), 5.31 (s, 1H), 7.33 (d, J = 8.1 Hz, 2H), 7.58 (dd, J = 8.1, 2.2 Hz, 2H), 8.48 (d, J = 2.2 Hz, 2H). ^{13}C NMR ($CDCl_3$, 75.5 MHz): δ −5.4, 18.3, 25.8, 61.8, 62.5, 121.3, 134.7, 135.0, 147.2, 161.5. MS CI: $[M + H]^+$ calcd for $C_{25}H_{44}N_3O_2Si_2$ m/z 474.3, found 474.0.

Representative Procedure for the Preparation of N4Py Derivatives. A mixture of **21** (0.21 g, 0.44 mmol), **15** (0.36 g, 1.3 mmol, 3 equiv), and Cs_2CO_3 (0.44 g, 1.4 mmol, 3 equiv) in dry acetonitrile (30 mL) was heated under reflux under a nitrogen atmosphere for 60 h. The reaction mixture was filtered through Celite, washed with diethyl ether, and concentrated in vacuo. Purification by flash chromatography over alumina using a gradient of hexane/ethyl acetate/TEA (20:9:1 to 0:9:1) afforded 0.23 g of **23** as a brown viscous oil.

***N*-[Bis[5-[[[*tert*-butyl(dimethyl)silyl]oxy]methyl]-2-pyridinyl]methyl]-*N*,*N*-bis[[5-[[[*tert*-butyl(dimethyl)silyl]oxy]-methyl]-2-pyridinyl]methyl]amine (23).** Yield: 56%. ^{1}H NMR ($CDCl_3$, 300 MHz): δ 0.08 (s, 24H), 0.91 (s, 36H), 3.92 (s, 4H), 4.69 (s, 4H), 4.70 (s, 4H), 5.30 (s, 1H), 7.61 (m, 8H),

8.43 (s, 2H), 8.49 (s, 2H). ^{13}C NMR ($CDCl_3$, 75.5 MHz): δ −5.4, 18.2, 25.8, 56.6, 62.6, 62.6, 71.0, 122.3, 123.4, 134.1, 134.4, 134.5, 134.7, 147.0, 147.2, 158.6, 158.7. ESI-MS: $[M + H]^+$ calcd for $C_{51}H_{86}N_5O_4Si_4$ *m/z* 944.8, found 944.6.

[6-[[[Bis[5-(hydroxymethyl)-2-pyridinyl]methyl][[5-(hydroxymethyl)-2-pyridinyl]methyl]amino]methyl]-3-pyridinyl]methanol (25). Dilute HCl (2 mL, 1 M) was added to a solution of **23** (0.22 g, 0.23 mmol) in ethanol (10 mL). After 0.5 h, the reaction mixture was extracted with diethyl ether. The aqueous layer was neutralized with solid Na_2CO_3 and lyophilized or azeotropically removed with ethanol. The residue was taken up in dry ethanol, and after filtration and evaporation of the solvent **25** was obtained as a viscous brown, hygroscopic oil (0.12 g, 100%). ^{1}H NMR (CD_3OD, 300 MHz): δ 3.86 (s, 4H), 4.58 (s, 4H), 4.91 (s, 4H), 5.32 (s, 1H), 7.39 (d, J = 8.1 Hz, 2H), 7.63 (d, J = 8.1 Hz, 4H), 7.73 (d, J = 8.1 Hz, 1H), 7.74 (d, J = 8.1 Hz, 1H), 8.38 (s, 2H), 8.49 (s, 2H). ^{13}C NMR (CD_3OD, 75.5 MHz): δ 58.3, 62.3, 62.4, 74.3, 124.6, 125.3, 137.0, 137.0, 137.1, 137.7, 148.7, 149.3, 158.8, 159.4. ESI-MS: $[M + H]^+$ calcd for $C_{27}H_{30}N_5O_4$ *m/z* 488.2, found 488.0.

***N*-[Bis[5-(methoxymethyl)-2-pyridinyl]methyl]-*N*,*N*-bis[[5-(methoxymethyl)-2-pyridinyl]methyl]amine (1c).** To a stirred suspension of NaH (55 mg, 1.4 mmol, 8 equiv) in dry DMF (1 mL) at 0 °C was added a solution of **25** (83 mg, 0.17 mmol) in dry DMF (1 mL). After 30 min, MeI (85 μL, 1.4 mmol, 8 equiv) was added at 0 °C. The reaction mixture was allowed to warm to room temperature and stirred for 2 h. Water was added and the mixture extracted with CH_2Cl_2. The organic layer was washed with water and brine. After drying over Na_2SO_4 and evaporation of the solvent, the product was obtained as a red/brown oil (83 mg, 89%). Purification by flash column chromatography over alumina using hexane/ethyl acetate/TEA (10:18:2) yielded **1c** as a light brown oil (37 mg, 40%). ^{1}H NMR ($CDCl_3$, 300 MHz): δ 3.33 (s, 12 H), 3.91 (s, 4H), 4.83 (s, 8H), 5.30 (s, 1H), 7.60 (s, 4H), 7.62 (s, 4H), 8.40 (s, 2H), 8.46 (s, 2H). ^{13}C NMR ($CDCl_3$, 75.5 MHz): δ 56.8, 58.1, 58.2, 71.4, 71.9, 122.5, 123.5, 131.4, 131.7, 135.7, 135.8, 148.4, 148.6, 159.3, 159.4. ESI-MS: $[M + H]^+$ calcd for $C_{31}H_{38}N_5O_4$ *m/z* 544.3, found 544.4.

[(1c)Fe(MeCN)](ClO$_4$)$_2$ (2c). To a solution of **1c** (32 mg, 0.059 mmol) in acetonitrile (1.5 mL) was added $Fe(ClO_4)_2 \cdot H_2O$ (17 mg, 0.066 mmol, 1.1 equiv). The deep red solution was placed in a sealed container, and ethyl acetate was allowed to diffuse slowly into the solution. A dark red oil was formed after 1 week. The solution was removed and the oil dissolved in acetonitrile. A fine red powder was formed which was isolated and redissolved in acetonitrile. Slow diffusion of ethyl acetate provided **2c** as red crystals after 2 weeks (9 mg, 18%). ^{1}H NMR (CD_3CN, 300 MHz): δ 3.29 (s, 6H), 3.34 (s, 6H), 4.32 (dd, J_{AB} = 27.9, 8.1 Hz, 4H), 4.38 (s, 4H), 4.47 (s, 4H), 6.31 (s, 1H), 7.04 (d, J = 8.1 Hz, 2H), 7.63 (d, J = 7.7 Hz, 2H), 7.85 (m, 4H), 8.82 (s, 2H), 8.92 (s, 2H). ESI-MS: $[M - (ClO_4)^- - (MeCN)]^+$ calcd for $C_{31}H_{38}ClFeN_5O_8$ *m/z* 698.2, found 698.0; $[M - 2(ClO_4)^-]^{2+}$ calcd for $C_{33}H_{40}FeN_6O_4$ *m/z* 320.1, found 320.3; $[M - 2(ClO_4)^- - (MeCN)]^{2+}$ calcd for $C_{31}H_{37}FeN_5O_4$ *m/z* 299.6, found 300.0.

***N*-[Bis[5-(3-bromopropyl)-2-pyridinyl]methyl]-*N*,*N*-bis[[5-(3-bromopropyl)-2-pyridinyl]methyl]amine (27).** PPh_3-Br_2 was freshly prepared by the addition of Br_2 (75 μL, 1.4 mmol, 4.7 equiv) to a stirred solution of Ph_3P (395 mg, 1.51 mmol, 5. equiv) in dry CH_2Cl_2 (1 mL) under a nitrogen atmosphere. To the resulting yellow suspension was added a solution of **24** (321 mg, 0.304 mmol) in dry CH_2Cl_2 (1 mL). The reaction mixture was stirred at room temperature until all the solids had disappeared (2 days) and extracted with 1 M HCl. The aqueous layer was neutralized with solid $NaHCO_3$ and extracted with CH_2Cl_2. The organic layer was washed with brine and dried over Na_2SO_4. Evaporation of the solvent afforded **27** as a red oil (190 mg, 74%). ^{1}H NMR ($CDCl_3$, 300 MHz): δ 2.07 (t, J = 6.6 Hz, 8H), 2.67 (t, J = 6.6 Hz, 8H), 3.32 (m, 8H), 3.84 (s, 4H), 5.22 (s, 1H), 7.34−7.58 (m, 8H), 8.28 (s, 2H), 8.34 (s, 2H). ^{13}C NMR ($CDCl_3$, 75.5 MHz): δ 30.5, 30.6, 32.5, 32.6, 33.3, 33.4, 56.6, 71.1, 122.3, 123.4, 133.6, 133.8, 136.1, 136.2, 148.8, 149.1, 157.5, 157.7. ESI-MS: $[M + H]^+$ calcd for $C_{35}H_{42}N_5Br_4$ *m/z* 848.0, found 848.0.[42]

Tetracysteine N4Py Derivative 1d. Methyl *N*-acetylcysteine (114 mg, 0.64 mmol) was dissolved in nitrogen-purged DMF (1 mL) and titrated with a Cs_2CO_3 solution (~100 mg/mL) to pH 9 under a nitrogen atmosphere. A solution of **27** (53 mg, 0.062 mmol) in DMF (1 mL) was added, and the mixture was stirred at room temperature overnight. The solution was diluted with water and lyophilized. The brown residue was purified by preparative reverse-phase HPLC with a gradient of 0.05 M ammonium acetate/MeCN (50:50) to 100% MeCN over 10 min. The product eluted at t_R = 7.2 min, was collected and lyophilized to yield a brown oil (9.5 mg, 12%). ^{1}H NMR ($CDCl_3$, 500 MHz): δ 1.88 (m, 8H), 2.04 (s, 12H), 2.55 (m, 8H), 2.68 (m, 8H), 3.00 (m, 8H), 3.78 (m, 12H), 3.90 (s, 4H), 4.83 (m, 4H), 5.28 (s, 1H), 6.39 (br•d, 4H), 7.47 (m, 8H), 8.35 (s, 2H), 8.41 (s, 2H). ESI-MS: $[M + H]^+$ calcd for $C_{59}H_{82}N_9O_{12}S_4$ *m/z* 1236.5, found 1236.1.

Tetrapeptide N4Py derivative 2e was first prepared as described for **1d**, using the peptide sequence Ac-Cys-Gly-Leu-His-Glu-Leu-Leu-Lys-Gly-NH_2.[43] After stirring overnight, the solution was acidified with 0.1% TFA in water and $Fe(ClO_4)_2$ (1.1 equiv) was added as a solution in MeCN. The orange/red solution was purified by preparative reverse-phase HPLC using a linear gradient of 20−40% solvent B over 10 min. The product eluted at t_R = 8.6 min and was collected and lyophilized to yield a yellow/orange solid (2.1 mg, 18%). MALDI: calcd for $[M - 2(ClO_4)^- - (MeCN)]^{2+}$ *m/z* 4620.38 (monoisotopic), 4623.43 (average isotope composition), found 4619.2.

Bis(5-bromo-2-pyridinyl)methanone (30). To a suspension of 2,5-dibromopyridine (10.7 g, 44.3 mmol) in dry toluene (1 L) at −80 °C was slowly added *n*-BuLi (29 mL, 46 mmol, 1.05 equiv) under a nitrogen atmosphere. After 2 h at −80 °C the reaction was quenched with freshly distilled diethyl carbonate (2.7 mL, 22 mmol, 0.5 equiv). After further workup and extractions using ethyl acetate, removal of the solvent furnished a dark brown oil, which solidified on standing. By triturating the crude product with diethyl ether pure **30** was obtained as a light brown solid (2.77 g, 37%). Recrystallization from diethyl ether afforded the analytically pure product. Mp: 191.8−192.7 °C. ^{1}H NMR ($CDCl_3$, 300 MHz): δ 7.98 (d, J = 8.2 Hz, 2H), 8.03 (dd, J = 8.2, 2.0 Hz, 2H), 8.77 (d, J = 2.0 Hz, 2H). ^{13}C NMR ($CDCl_3$, 50.3 MHz): δ 124.7, 126.3, 139.4, 150.3, 152.1, 191.0. HRMS: calcd for $C_{11}H_6N_2OBr_2$ 339.885, found 339.885. Anal. Calcd for $C_{11}H_6N_2OBr_2$: C, 38.60; H, 1.80; N, 8.20; Br, 46.70. Found: C, 38.33; H, 1.80; N, 8.01; Br, 47.04.

Bis(5-bromo-2-pyridinyl)methanone Oxime (31). A mixture of **30** (2.77 g, 8.10 mmol), hydroxylammonium chloride (3.0 g, 42 mmol, 5 equiv), and pyridine (3.4 mL, 42 mmol, 5 equiv) in ethanol (60 mL) was refluxed for 1.5 h and subsequently cooled to room temperature. The mixture was poured into ice−water and stirred in an ice bath for 1 h. The solids were collected by filtration and washed with cold dilute ethanol (50%). After drying in air the product was obtained as a light brown solid (2.63 g, 91%). Recrystallization from ethanol/water afforded analytically pure **31** as fine, lilac/grey needles. Mp: 191.1−191.7 °C. ^{1}H NMR (CD_3OD, 300 MHz): δ 7.63 (d, J = 8.4 Hz, 1H), 7.76 (d, J = 8.4 Hz, 1H), 7.92 (dd, J = 8.4, 1.8 Hz, 1H), 7.99 (dd, J = 8.4, 1.8 Hz, 1H), 8.65 (d, J = 1.8 Hz, 1H), 8.70 (d, J = 1.8 Hz, 1H). ^{1}H NMR (CD_3OD, 300 MHz): δ 7.56 (d, J = 8.4 Hz, 1H), 7.83 (d, J = 8.4 Hz, 1H), 8.01 (dd, J = 8.4, 2.2 Hz, 1H), 8.10 (dd, J = 8.4, 2.2 Hz, 1H), 8.50 (d, J = 2.2 Hz, 1H), 8.68 (d, J = 2.2 Hz, 1H). ^{13}C NMR ($CDCl_3$, 50.3 MHz): δ 119.8, 120.3, 122.8, 127.0, 138.7, 139.4, 149.5, 150.0, 150.6, 153.0, 153.9. HRMS: calcd for $C_{11}H_7N_3OBr_2$ 354.895, found

(42) Observed isotope pattern corresponds to the calculated isotope pattern.

(43) The peptide was synthesized by standard automated methods on a peptide synthesizer using Fmoc-protected amino acids and purified by preparative reversed-phase HPLC.

354.896. Anal. Calcd for $C_{11}H_7N_3OBr_2$: C, 37.00; H, 2.00; N, 11.80; Br, 44.80. Found: C, 36.58; H, 2.06; N, 11.41; Br, 44.58.

Bis(5-bromo-2-pyridinyl)methylamine (32). Zinc dust (1.1 g, 17 mmol, 3.4 equiv) was carefully added in small portions over 5 min to a suspension of **31** (1.79 g, 5.01 mmol) in acetic acid (30 mL) and water (15 mL). The mixture was stirred at room temperature for 30 min, filtered over Celite, and washed with water. After evaporation of the solvent the residue was neutralized with concentrated ammonia (25% v/v) and extracted with CH_2Cl_2. The organic layer was washed with brine, dried over Na_2SO_4, and concentrated in vacuo to furnish pure **32** as a brown viscous oil that solidified on standing (1.62 g, 94%). ^{1}H NMR ($CDCl_3$, 300 MHz): δ 2.38 (br. s, 2H), 5.24 (s, 1H), 7.29 (d, J = 8.4 Hz, 2H), 7.72 (dd, J = 8.4, 1.8 Hz, 2H), 8.55 (d, J = 1.8 Hz, 2H). ^{13}C NMR ($CDCl_3$, 75.5 MHz): δ 61.1, 119.1, 123.0, 139.2, 150.1, 160.6. HRMS: calcd for $C_{11}H_9N_3Br_2$ 340.916, found 340.916.

5-Bromo-2-picoline *N*-Oxide (34). A mixture of 5-bromo-2-picoline[18] (11.7 g, 68.0 mmol) and *m*-CPBA (20.9 g, 97.0 mmol, 1.4 equiv) in CH_2Cl_2 (300 mL) was stirred at room temperature overnight. Saturated Na_2CO_3 (300 mL) was added, and after stirring for 30 min the layers were separated. The water layer was extracted with CH_2Cl_2, and the combined organic layers were washed with brine and dried over Na_2SO_4. Evaporation of the solvent yielded **34** as a light yellow solid that was recrystallized from diethyl ether (12.5 g, 98%). Mp: 119.3−120.4 °C (lit.[44] mp 117−118 °C). ^{1}H NMR ($CDCl_3$, 300 MHz): δ 2.43 (s, 3H), 7.11 (d, J = 8.4 Hz, 1H), 7.29 (dd, J = 8.4, 1.1 Hz, 1H), 8.38 (d, J = 1.1 Hz, 1H). ^{13}C NMR ($CDCl_3$, 75.5 MHz): δ 17.2, 117.1, 126.3, 128.2, 140.5, 147.9. HRMS: calcd for C_6H_6BrNO 186.963, found 186.963.

(5-Bromo-2-pyridinyl)methanol (35). Protected from moisture, trifluoroacetic acid anhydride (10 mL, 71 mmol, 5 equiv) was carefully and slowly added to **34** (2.63 g, 14.0 mmol). After the vigorous thermal reaction had ceased, the orange mixture was stirred at room temperature for 30 min and then refluxed for 30 min. Saturated $NaHCO_3$ was carefully added after cooling to room temperature until pH 8 was reached. The resulting red solution was stirred at room temperature under nitrogen overnight. The mixture was extracted with CH_2Cl_2, and the combined organic layers were washed with brine and dried over Na_2SO_4. Evaporation of the solvent yielded **35** as a dark brown oil (2.15 g, 81%), which was used without any further purification. ^{1}H NMR ($CDCl_3$, 300 MHz): δ 3.41 (br. s, 1H), 4.72 (s, 2H), 7.20 (d, J = 8.3 Hz, 1H), 7.81 (dd, J = 8.3, 2.2 Hz, 1H), 8.60 (d, J = 2.2 Hz, 1H). ^{13}C NMR ($CDCl_3$, 75.5 MHz): δ 63.9, 118.8, 121.9, 139.3, 149.3, 158.5. HRMS: calcd for C_6H_6NOBr 186.963, found 186.965.

5-Bromo-2-(chloromethyl)pyridine (36). Thionyl chloride (0.89 mL, 12 mmol, 1.5 equiv) was slowly added to a solution of crude **35** (1.51 g, 8.03 mmol) in CH_2Cl_2 (30 mL) at 0 °C, protected from moisture. After being allowed to warm to room temperature the reaction mixture was stirred overnight. Saturated $NaHCO_3$ was added, and the layers were separated. The water layer was extracted with CH_2Cl_2. The combined organic layers were washed with brine and dried over Na_2SO_4. Concentration by rotary evaporation furnished **36** as a dark brown oil (1.63 g, 97%) that was used without further purification.[45] ^{1}H NMR ($CDCl_3$, 300 MHz): δ 4.61 (s, 2H), 7.37 (d, J = 8.4 Hz, 1H), 7.84 (dd, J = 8.4, 2.2 Hz, 1H), 8.62 (d, J = 2.2 Hz, 1H). ^{13}C NMR ($CDCl_3$, 75.5 MHz): δ 45.8, 120.0, 124.0, 139.5, 150.3, 155.0. HRMS: calcd for C_6H_6NClBr 204.929, found 204.929.

***N*-[Bis(5-bromo-2-pyridinyl)methyl]-*N*,*N*-bis[(5-bromo-2-pyridinyl)methyl]amine (1b).** A mixture of **32** (0.72 g, 2.1 mmol), **36** (1.8 g, 8.7 mmol, 4 equiv), DIEA (7.2 mL, 41 mmol, 20 equiv), and dry acetonitrile (1.5 mL) was heated at 80 °C under a nitrogen atmosphere for 2 days. After evaporation of the solvent, the residue was taken up in CH_2Cl_2, washed with water and brine, and dried over Na_2SO_4. Concentration by rotary evaporation afforded a dark brown, viscous oil that solidified on standing. After purification by flash chromatography over aluminum oxide using a gradient of hexane/ethyl acetate/TEA (40:9:1−30:9:1) and evaporation of the solvents, the residue was precipitated from diethyl ether to furnish **1b** as a white solid (0.83 g, 58%). Recrystallization from CH_2Cl_2/diethyl ether afforded pure **1b** as white needles. Mp: 151.6−152.2 °C. ^{1}H NMR ($CDCl_3$, 300 MHz): δ 3.84 (s, 4H), 5.21 (s, 1H), 7.40 (d, J = 8.4 Hz, 2H), 7.55 (d, J = 8.4 Hz, 2H), 7.72 (dd, J = 8.4, 2.2 Hz, 2H), 7.78 (dd, J = 8.4, 2.2 Hz, 2H), 8.56 (d, J = 2.2 Hz, 2H), 8.59 (d, J = 2.2 Hz, 2H). ^{13}C NMR ($CDCl_3$, 75.5 MHz): δ 56.3, 70.7, 119.0, 119.5, 124.4, 125.2, 138.9, 139.0, 150.1, 150.3, 157.7. MS CI: $[M + H]^+$ calcd for $C_{23}H_{18}Br_4N_5$ m/z 679.8, found 679.9. Anal. Calcd for $C_{23}H_{17}Br_4N_5 \cdot H_2O$: C, 39.41; H, 2.73; N, 9.99; Br, 45.59. Found: C, 39.54; H, 2.39; N, 9.96; Br, 45.60.

[(1b)Fe(MeCN)](ClO$_4$)$_2$ (2b). To a suspension of **1b** (93 mg, 0.14 mmol) in methanol (3 mL) was added $Fe(ClO_4)_2 \cdot 6H_2O$ (56 mg, 0.15 mmol, 1.1 equiv) in acetonitrile (1.5 mL). The resulting clear dark red/brown solution was stirred at room temperature for 15 min. Slow diffusion of ethyl acetate into this solution furnished **2b** as red crystals (115 mg, 87%). ^{1}H NMR (CD_3CN, 300 MHz): δ 4.31 (dd, J_{AB} = 51.8, 18.5 Hz, 4H), 6.32 (s, 1H), 7.00 (d, J = 8.4 Hz, 2H), 7.75 (d, J = 8.4 Hz, 2H), 7.88 (dd, J = 8.4, 1.8 Hz, 2H), 8.10 (dd, J = 8.4, 1.8 Hz, 2H), 9.01 (d, J = 1.8 Hz, 2H), 9.10 (d, J = 1.8 Hz, 2H). ESI-MS: $[M - (ClO_4)^- - (MeCN)]^+$ calcd for $C_{23}H_{17}Br_4ClFeN_5O_4$ m/z 833.7, found 833.6; $[M - 2(ClO_4)^- - (MeCN)]^{2+}$ calcd for $C_{23}H_{17}Br_4FeN_5$ m/z 367.4, found 367.8. Anal. Calcd for $C_{25}H_{20}Br_4Cl_2FeN_6O_8$: C, 30.68; H, 2.07; N, 8.59. Found: C, 30.48; H, 2.27; N, 8.36.

***N*-[Bis(5-phenyl-2-pyridinyl)methyl]-*N*,*N*-bis[(5-phenyl-2-pyridinyl)methyl]amine (1f).** A mixture of **1b** (0.20 g, 0.30 mmol), phenylboronic acid (0.36 g, 3.0 mmol, 10 equiv), $Pd(PPh_3)_4$ (40 mg, 34 μmol, 12 mol %), aqueous Na_2CO_3 (2 M, 2 mL), and 4 droplets of ethylene glycol was heated at reflux in toluene (4 mL) under a nitrogen atmosphere overnight. The layers were separated, and the aqueous phase was extracted with CH_2Cl_2. The combined organic layers were washed with aqueous Na_2CO_3 (2 M) and brine and dried over Na_2SO_4. Evaporation of the solvent afforded the pure product as a dark yellow oil that solidified on standing (0.16 g, 80%). ^{1}H NMR (300 MHz, $CDCl_3$): δ 4.10 (s, 4H), 5.50 (s, 1H), 7.30−7.90 (m, 28H), 8.72 (d, J = 2.2 Hz, 2H), 8.82 (s, 2H). ^{13}C NMR (50.3 MHz, $CDCl_3$): δ 57.3, 72.4, 115.6, 119.0, 123.0, 123.8, 126.8, 126.9, 127.8, 128.2, 128.4, 128.8, 131.8, 132.0,134.6, 134.7, 134.9, 137.5, 147.2, 147.6, 158.3, 158.6. MS CI: $[M + H]^+$ calcd for $C_{47}H_{38}N_5$ m/z 672.3, found 672.0 (fragmentations m/z 352, 323, 170).

[(1f)Fe(MeCN)](ClO$_4$)$_2$ (2f) was prepared as described for **2c**. After failed crystallization attempts the solution was concentrated and analyzed by mass analysis. ESI-MS: [M-(MeCN)] calcd for $C_{47}H_{37}Cl_2FeN_5O_8$ m/z 925.1, found 925.1; $[M - (ClO_4)^- - (MeCN)]^+$ calcd for $C_{47}H_{37}ClFeN_5O_4$ m/z 826.3, found 826.3; $[M - 2(ClO_4)^-]^{2+}$ calcd for $C_{49}H_{40}FeN_6$ m/z 384.1, found 384.3; $[M - 2(ClO_4)^- - (MeCN)]^{2+}$ calcd for $C_{47}H_{37}FeN_5$ m/z 363.6, found 363.7.

6-[[[Bis(5-bromo-2-pyridinyl)methyl][(5-formyl-2-pyridinyl)methyl]amino]methyl]nicotinaldehyde (38). To **1b** was added *i*-PrMgBr (4.5 equiv, 2 M in THF), and the mixture was stirred at room temperature for 1 h. ESI-MS analysis of a sample quenched with water revealed the molecular ion of **37a** ($[M + H]^+$ 526). When quenched with D_2O, the molecular ion of **37b** ($[M + H]^+$ 529) was observed. More *i*-PrMgBr (4 equiv) was added, and the mixture was heated to reflux for 1 h. Dry DMF (15 equiv) was added. The mixture was washed with water and brine and dried over Na_2SO_4. Evaporation of the solvent afforded a brown oil. ^{1}H NMR (300 MHz, $CDCl_3$): δ 4.03 (s, 4H), 5.27 (s, 1H), 7.52 (d, J = 8.2 Hz, 2H), 7.71 (d, J = 8.2 Hz, 2H), 7.76 (dd, J = 8.2, 1.8 Hz, 2H), 8.07 (dd, J =

(44) Blanz, E. J., Jr.; French, F. A.; DoAmaral, J. R.; French, D. A. *J. Med. Chem.* **1970**, *13*, 1124−1130.

(45) The product can be stored at −20 °C under nitrogen for many months without any detectable signs of degradation.

8.2, 1.8 Hz, 2H), 8.58 (d, J = 1.8 Hz, 2H), 8.93 (d, J = 1.8 Hz, 2H), 10.04 (s, 2H). ^{13}C NMR (50.3 MHz, $CDCl_3$): δ 58.0 (t), 71.8 (d), 120.5 (s), 124.1 (d), 126.0 (d), 130.9 (s), 136.9 (d), 139.8 (d), 150.2 (d), 152.4 (d), 158.2 (s), 166.2 (s), 191.1 (d). MS CI: $[M + H]^+$ calcd for $C_{25}H_{19}Br_2N_5O_2$ m/z 582.3, found 582.3 (fragmentations m/z 329, 254, 122).

ABTS Oxidation. The ABTS oxidations were performed in 0.1 M acetate buffers at pH 3.5 using a magnetically stirred, thermostatically controlled quartz cuvette at 20 °C. The increase in absorption due to the $ABTS^{+\bullet}$ radical cation formation was monitored by a UV−vis spectrophotometer at 660 nm (ϵ 1.47 × 10^4 $M^{-1}cm^{-1}$)[40] at intervals of 1 s for 400 s with an incremental cycle time of 10% after an initial time of 60 s. The experimental conditions were identical to those described previously.[16]

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Supporting Information Available: Experimental procedures and analytical data of compounds **5**, **6**, **10**, **12**, **14**, **16**, **18**, **20**, **22**, and **24**, and a summary of the crystallographic data and the cyclic voltammetry measurements. This material is available free of charge via the Internet at http://pubs.acs.org. Further details of the crystal structure may be obtained from the Cambridge Crystallographic Data Centre by quoting the references CCDC 210556 for **2b** and CCDC 210557 for **2c**.

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