

# **Structural Basis for Xenosiderophore Utilization by the Human Pathogen *Staphylococcus aureus***

## **Supplementary Information**

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## I. Supplementary Tables

**Supplementary Table 1.** Known iron acquisition virulence factors and associated genes in *S. aureus* pathogens.<sup>1</sup>

| Iron Source                                      | <i>S. aureus</i> Genes   |
|--|--|
| Heme Binding, extraction, and import             | <i>isdAB, isdH, isdCDEF, srtB, srtA</i>                                  |
| Degradation                                      | <i>isdG, isdI, iruO</i>  |
| Sensing and export                               | <i>hssRS-hrtAB</i>   |
| Endogenous siderophores                          | Staphyloferrin A, Staphyloferrin B                                       |
| Synthesis and export                             | Staphyloferrin A: <i>sfaABC, sfaD</i><br>Staphyloferrin B: <i>sbnA-I</i> |
| Import and iron release                          | Staphyloferrin A: <i>htsABC</i><br>Staphyloferrin B: <i>sirABC</i>       |
| Xenosiderophores                                 | Desferrioxamine B, ferrichrome, aerobactin, coprogen                     |
| Hydroxamate                                      | <i>fhuBGC<sub>2</sub>, fhuD1, fhuD2</i>                                  |
| Catecholate                                      | <i>sstABCD</i>   |
| Inorganic Iron (ferrous iron transport proteins) | <i>feoAB, fepABC</i>   |
| Major transcriptional regulator                  | <i>fur</i>   |

<sup>1</sup>Table adapted from reference 1.

**Supplementary Table 2.** Strains and plasmids used in this work.

| Strain                                  | Plasmid | Inducible Gene/Marker     | Origin/Reference     |
|---|---------|---------------------------|----------------------|
| <i>Staphylococcus aureus</i> SG511      | None    | Wild Type                 | Hans Knöll Institute |
| <i>Staphylococcus aureus</i> ATCC 11632 | None    | Wild Type                 | ATCC                 |
| <i>E. coli</i> TOP10                    | None    | Cloning strain            | Agilent              |
| <i>E. coli</i> BL21 (DE3)               | None    | Protein expression strain | Agilent              |
| <i>E. coli</i> Top10                    | pET28a  | FhuD2                     | This work            |
| <i>E. coli</i> BL21 (DE3)               | pET28a  | FhuD2                     | This work            |

**Supplementary Table 3.** Primary protein sequences of wild type FhuD2 from *S. aureus* and the truncated *N*-His<sub>6</sub>-FhuD2 used here in fluorescence quenching assays.

FhuD2-WT<sup>a</sup> from *S. aureus* (GenBank Accession # AAK92086.1):

MKKLLLPLIIMLLVLAACGNQGEKNNKAETKSYKMDDGKTVDIPKDPKRIAVVAPTYA  
GGLKKLGANIVAVNQVDQSKVLKDKFKGVTKIGDGDVEKVAKEKPDLIIVYSTDKDI  
KKYQKVAPTVVVDYNKHKYLEQQEMLGKIVGKEDKVKAWKKDWEETTAKDGKEIKK  
AIGQDATVSLFDEFDDKKLYTYGDNWGRGGEVLYQAFGLKMQPEQQKLTAKAGWAEV  
KQEEIEKYAGDYIVSTSEGKPTPGYESTNMWKNLKATKEGHIVKVDAGTYWYNDPYTL  
DFMRKDLKEKLIKAAK

FhuD2-*N*-His<sub>6</sub><sup>b</sup> used in this work:

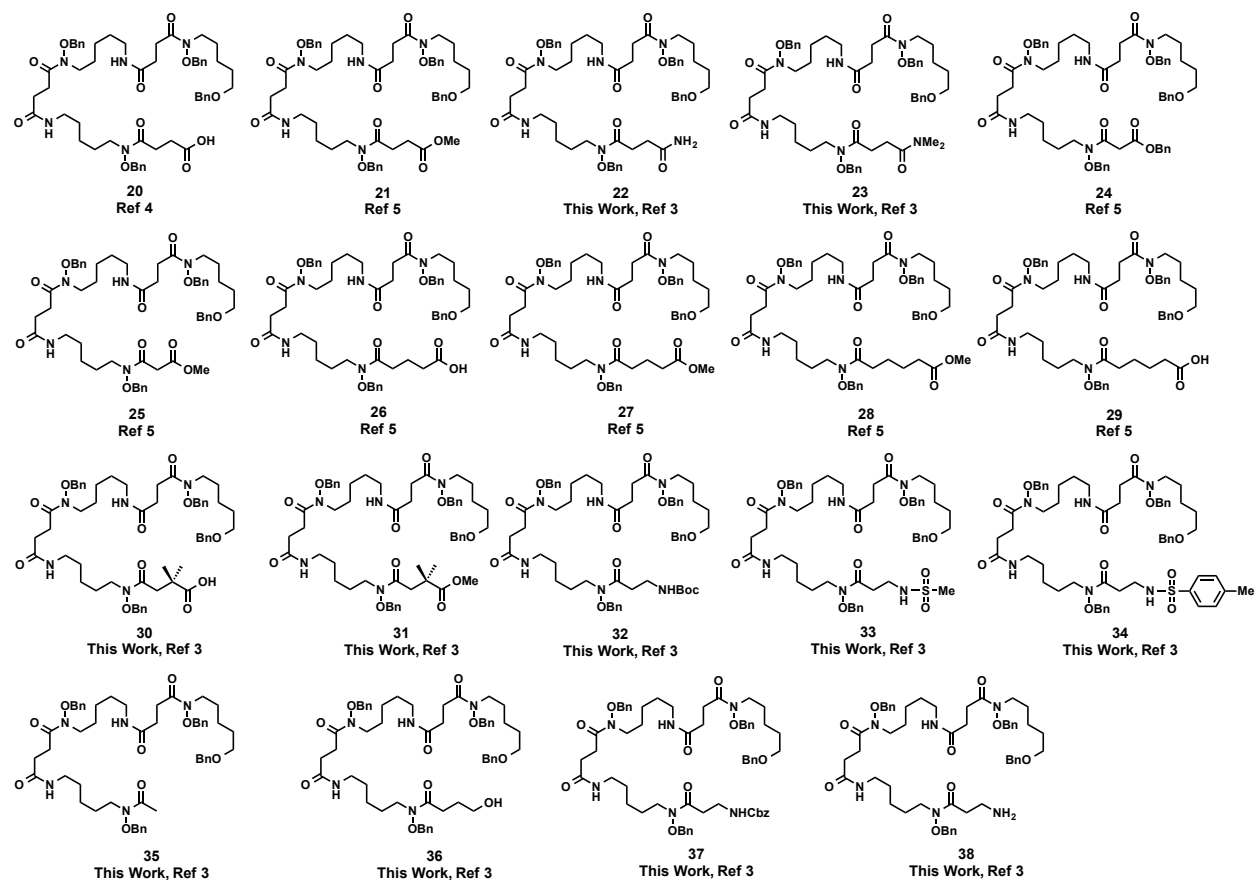
MGSSHHHHHHSSGLVPRGSHMNNKAETKSYKMDDGKTVDIPKDPKRIAVVAPTYAGG  
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YQKVAPTVVVDYNKHKYLEQQEMLGKIVGKEDKVKAWKKDWEETTAKDGKEIKKAI  
GQDATVSLFDEFDDKKLYTYGDNWGRGGEVLYQAFGLKMQPEQQKLTAKAGWAEVKQ  
EEIEKYAGDYIVSTSEGKPTPGYESTNMWKNLKATKEGHIVKVDAGTYWYNDPYTLDF  
MRKDLKEKLIKAAK

<sup>a</sup>Pre-lipoprotein signal sequence highlighted in magenta. Soluble siderophore-binding domain highlighted in teal. <sup>b</sup>Hexahistidine motif with thrombin cleavage site highlighted in yellow. Soluble siderophore-binding domain highlighted in teal.

**Supplementary Table 4.** Codon optimized nucleotide sequence of FhuD2-*N*-His<sub>6</sub> from *S. aureus* that was cloned into a pET28 vector for protein expression in *E. coli* BL21 (DE3).

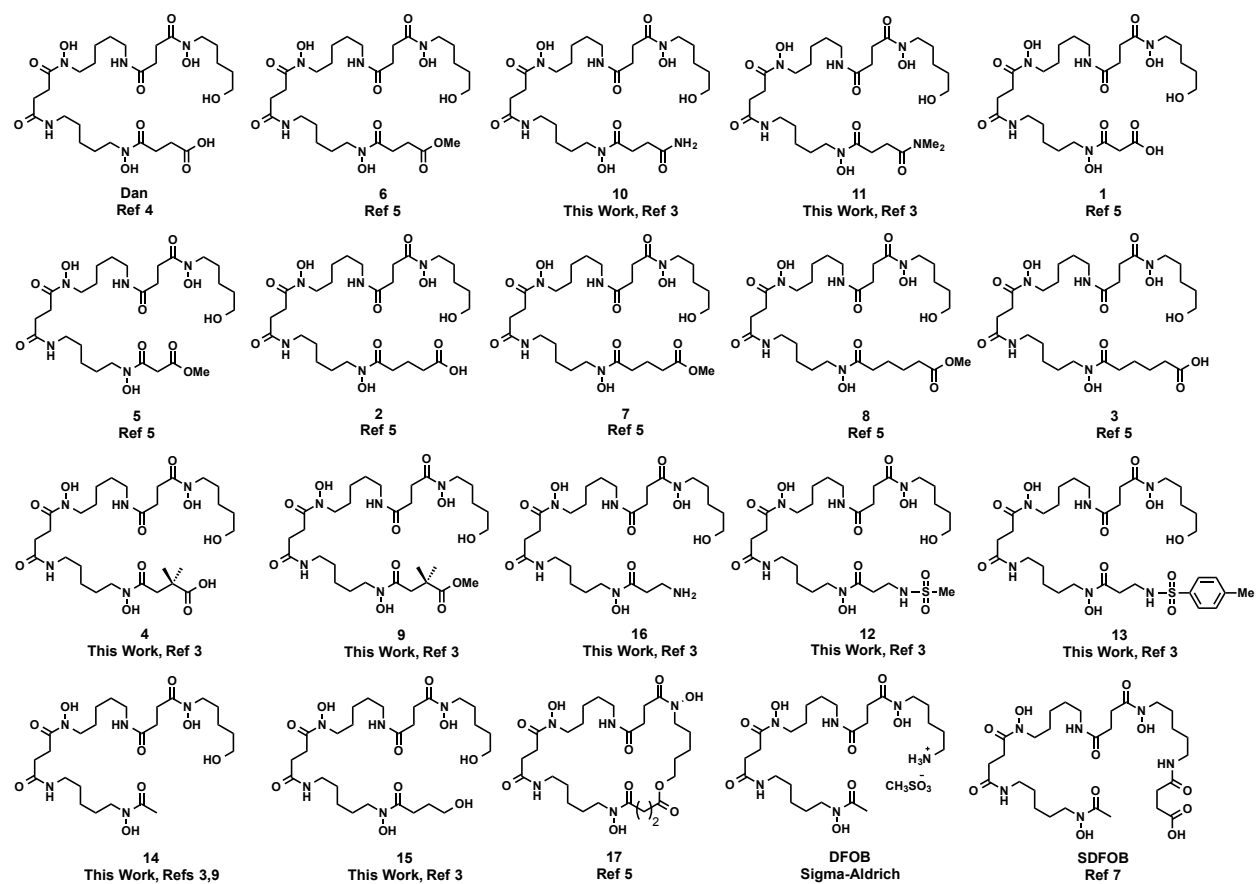
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CGTGGAAGAAAGACTGGGAGGAAACCACCGCGAAAGATGGCAAGGAGATCAAGAA  
AGCGATTGGCCAGGACGCGACCGTTAGCCTGTTTCGACGAATTTGATAAGAACTGT  
ACACCTATGGTGATAACTGGGGTTCGTGGTGGCGAGGTGCTGTACCAGGCGTTTCGGTC  
TGAAGATGCAACCGGAACAGCAAAAGCTGACCGCGAAAGCGGGTTGGGCGGAAGT  
GAAGCAAGAGGAAATCGAAAAATACGCGGGCGACTATATTGTGAGCACCAGCGAG  
GGTAAACCGACCCCGGGCTACGAAAGCACCAACATGTGGAAAAACCTGAAGGCGA  
CCAAAGAGGGTACATCGTTAAGGTGGATGCGGGCACCTACTGGTATAACGACCCG  
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## II. Supplementary Figures

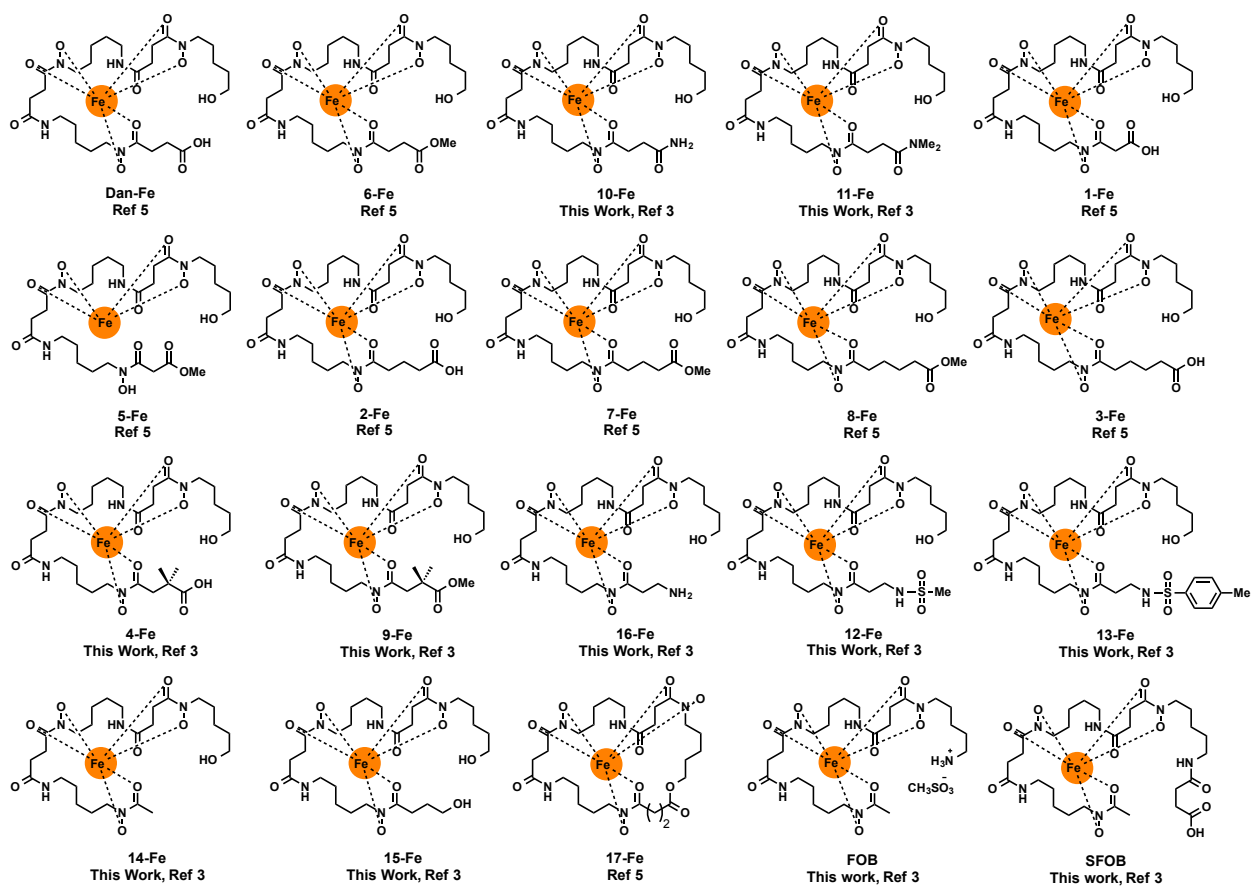


**Supplementary Figure 1.** Structures of benzyl protected siderophores **20–38**.

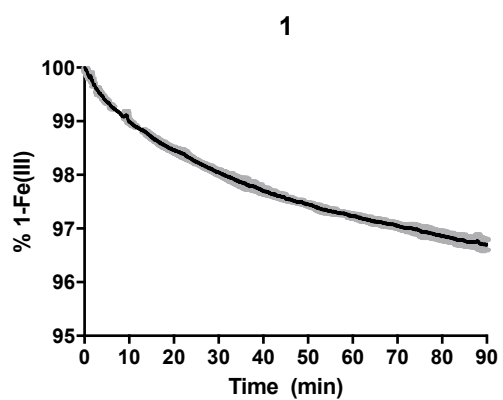
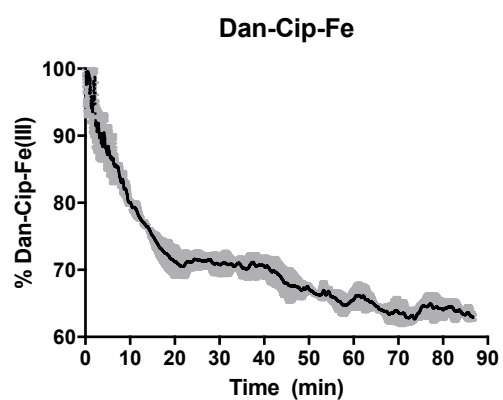
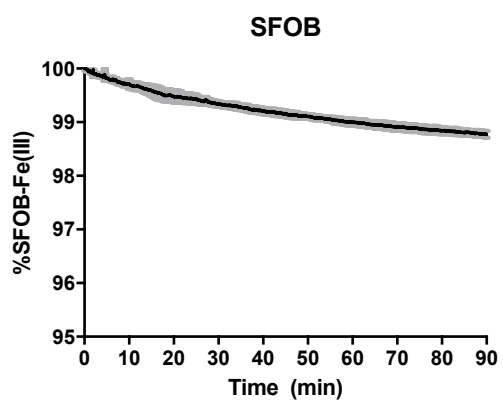
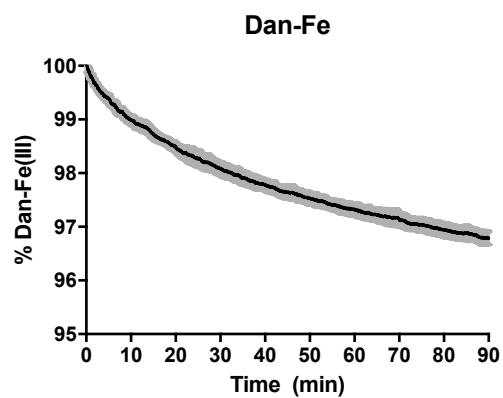
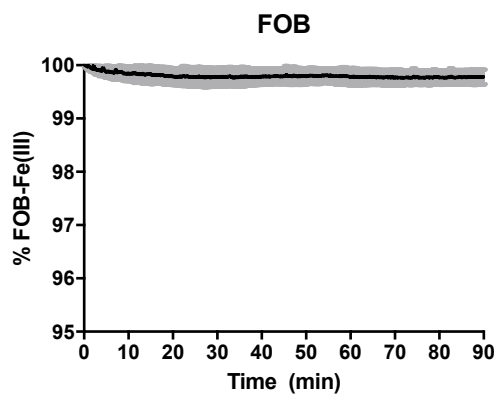


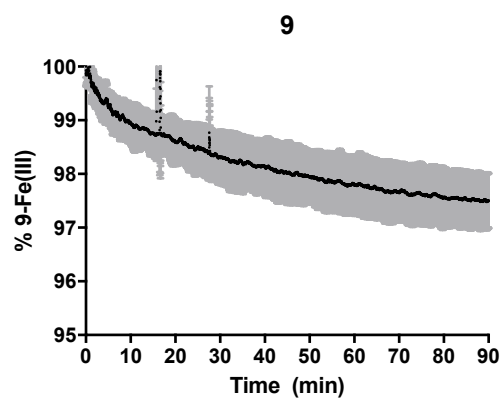
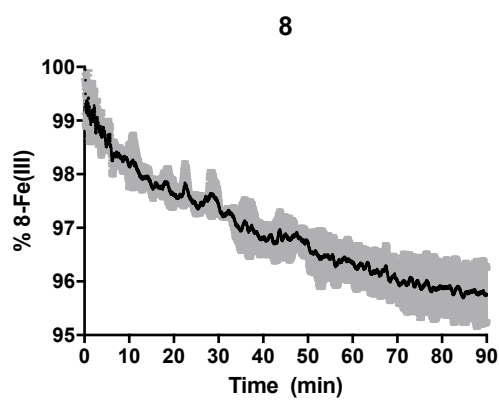
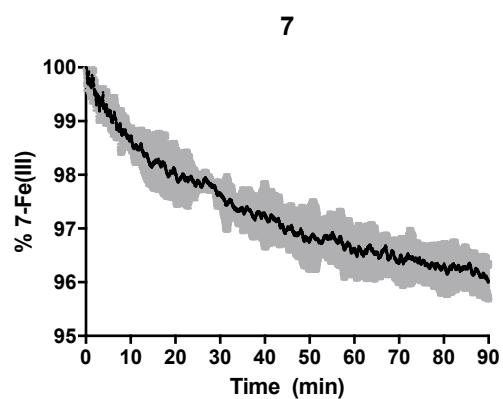
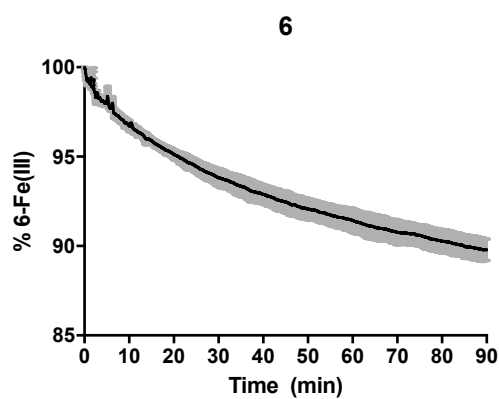
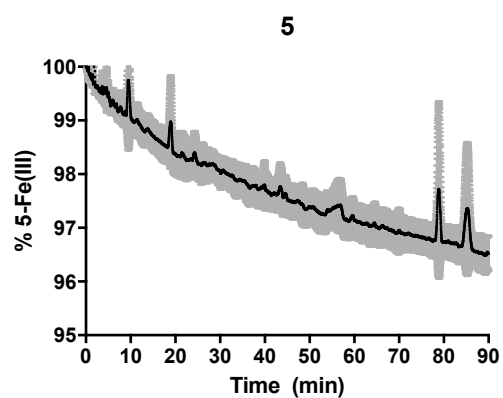
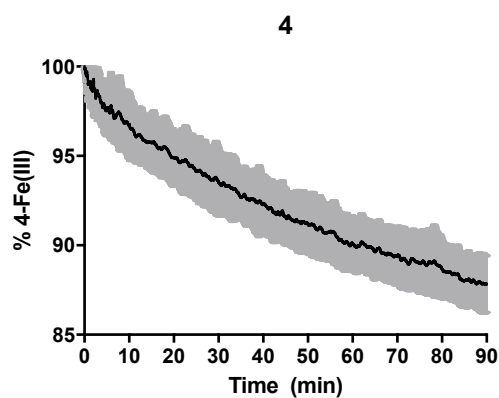
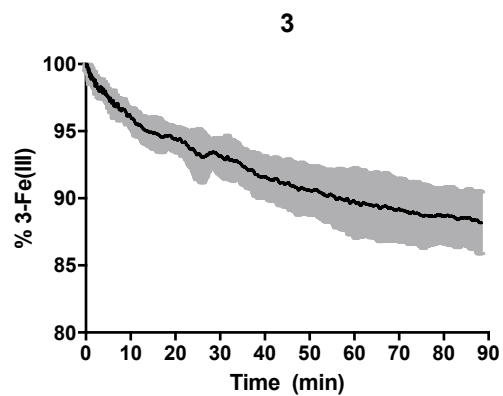
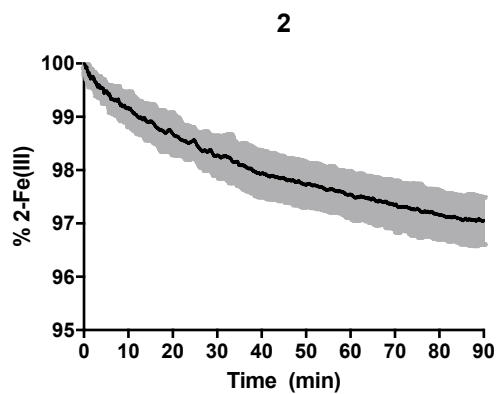


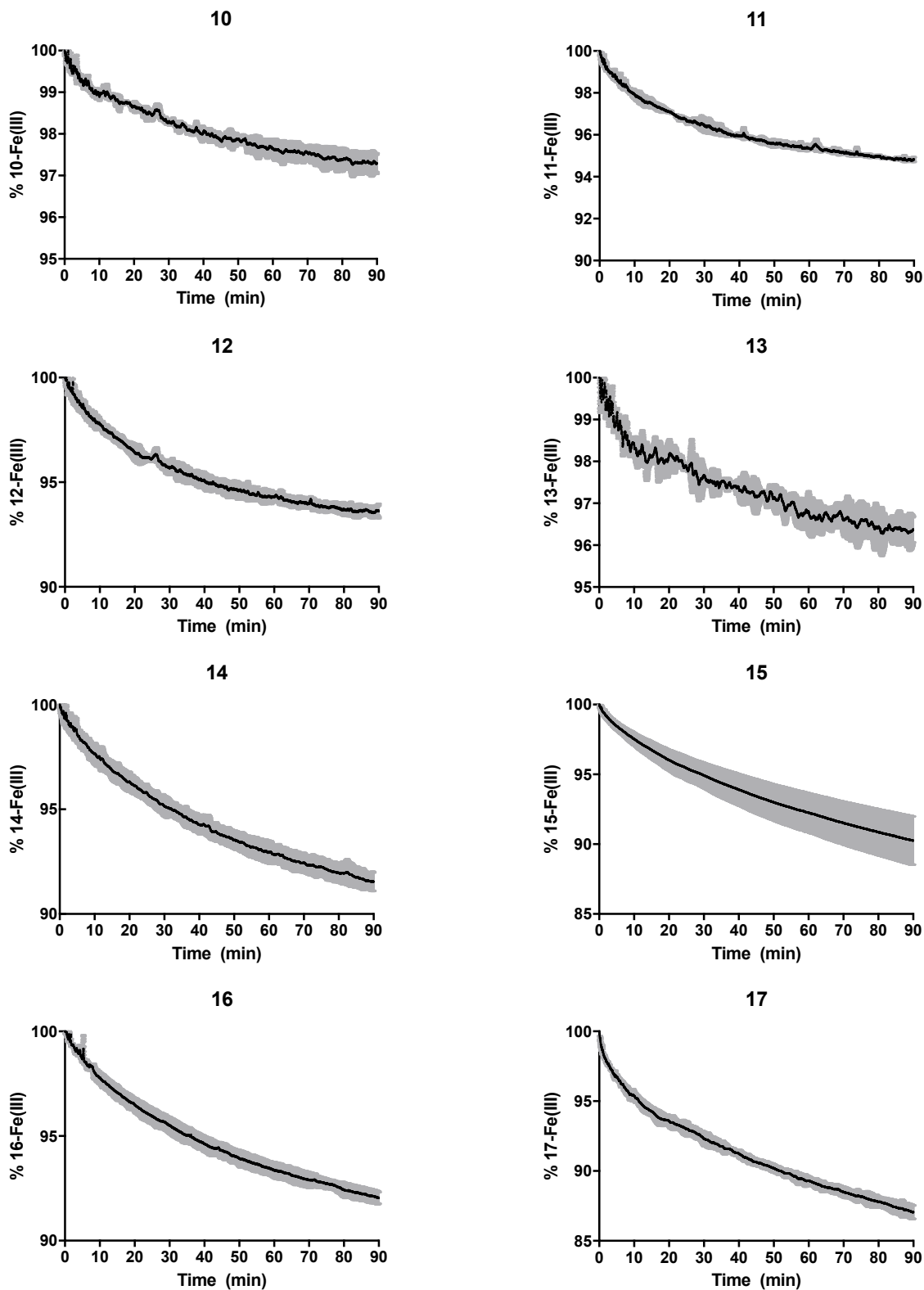
**Supplementary Figure 2.** Structures of siderophores **Dan**, **DFOB**, **SDFOB**, and **1–17**.



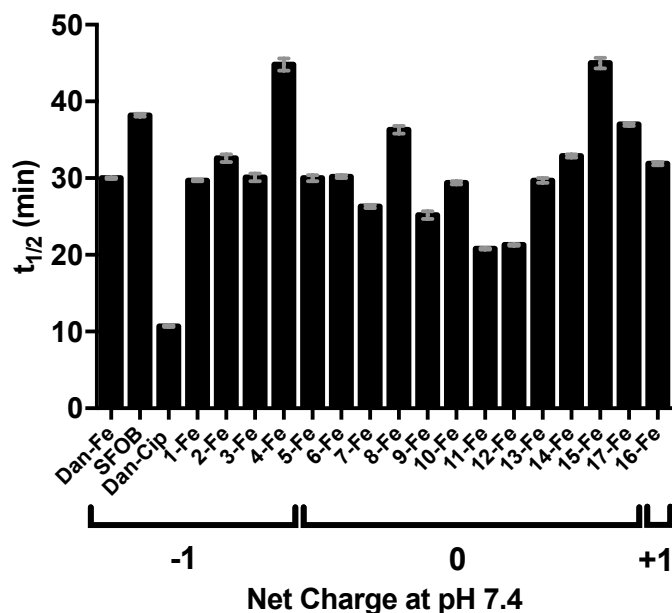
**Supplementary Figure 3.** Structures of siderophores **Dan-Fe**, **FOB**, **SFOB**, and **1-Fe–17-Fe**.



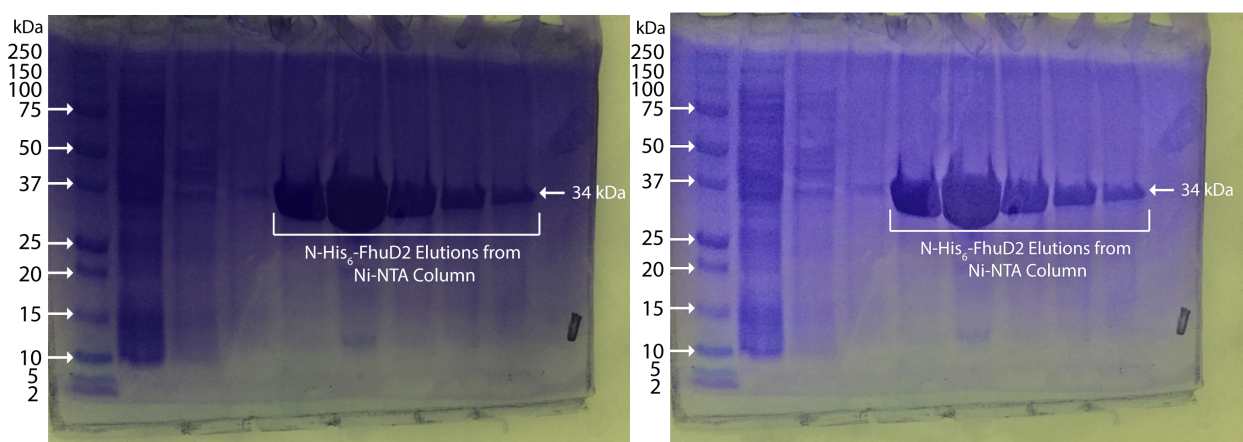




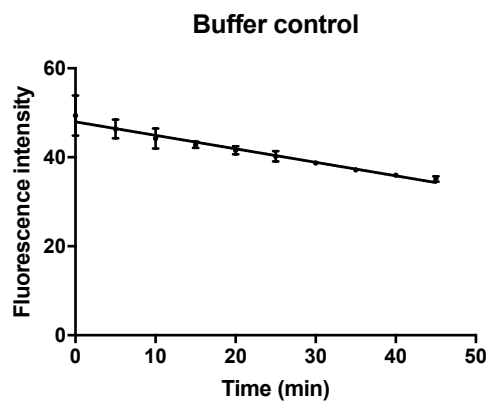
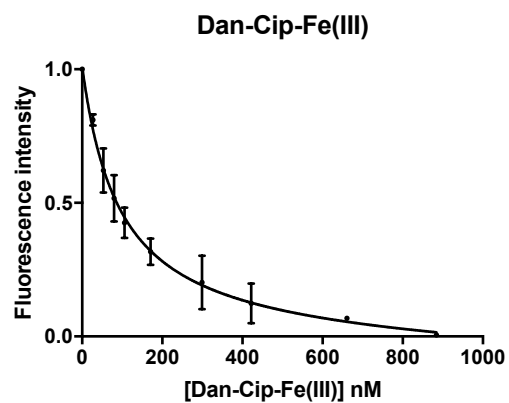
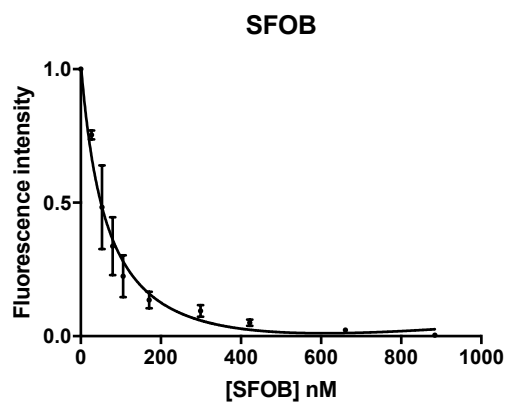
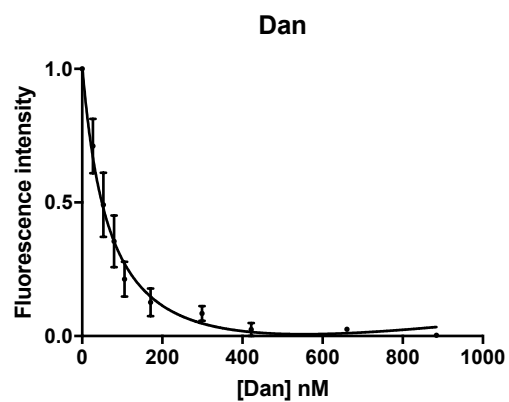
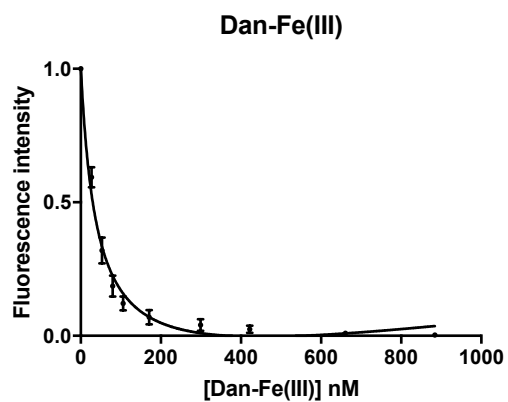
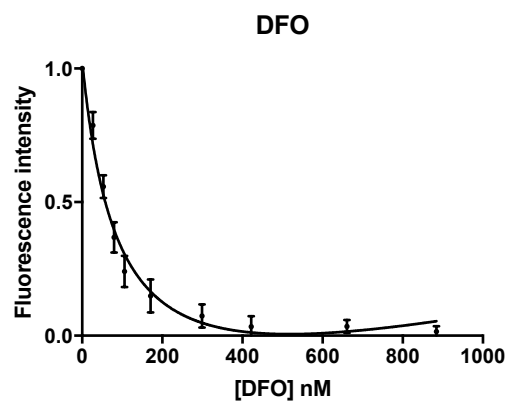
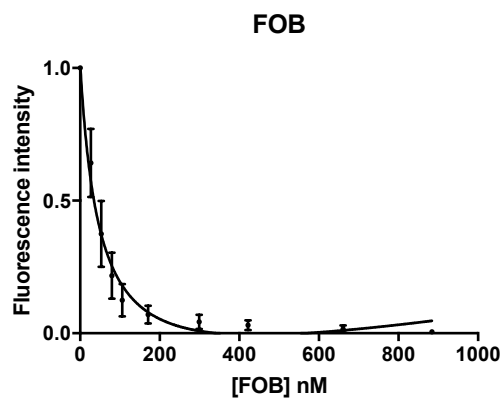
**Supplementary Figure 4.** Exponential decay plots for EDTA iron(III) binding competition assay with ferrioxamine siderophores. Error bars represent standard deviations for at least three independent trials.

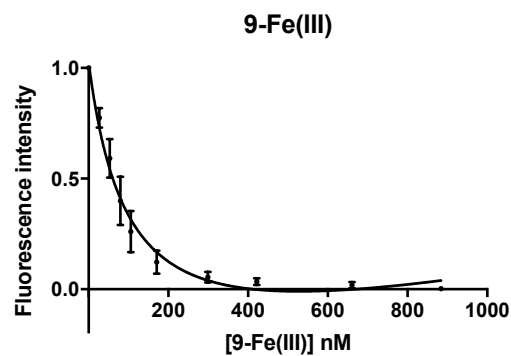
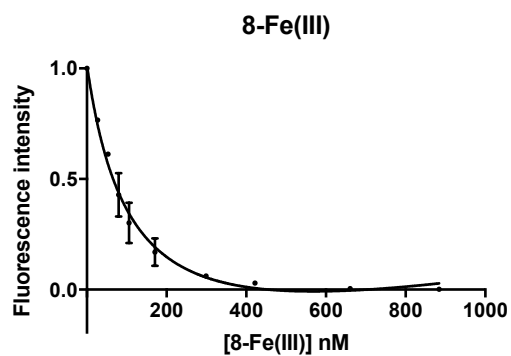
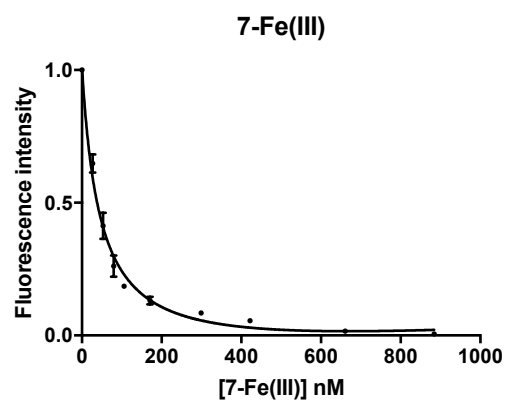
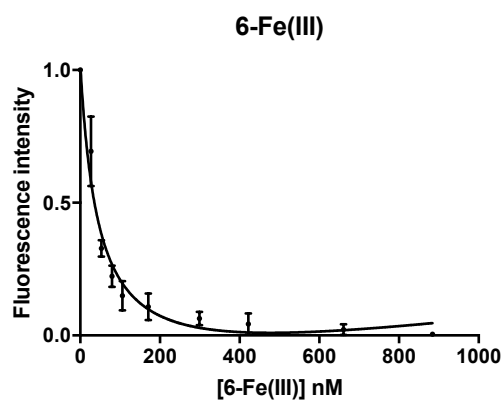
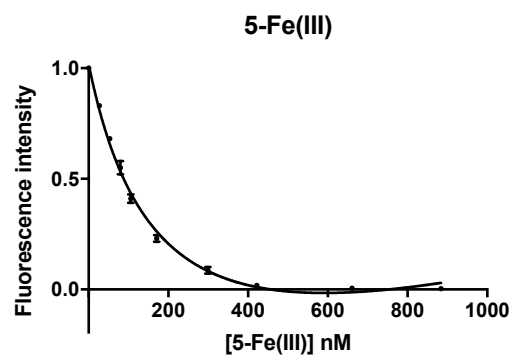
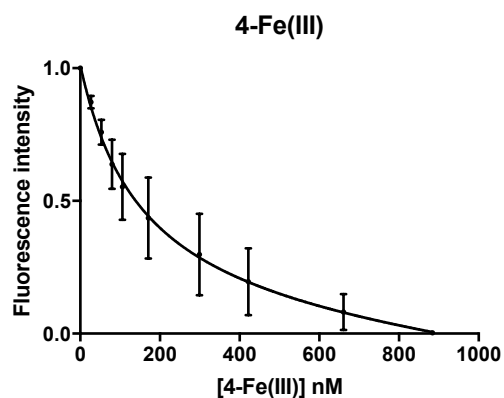
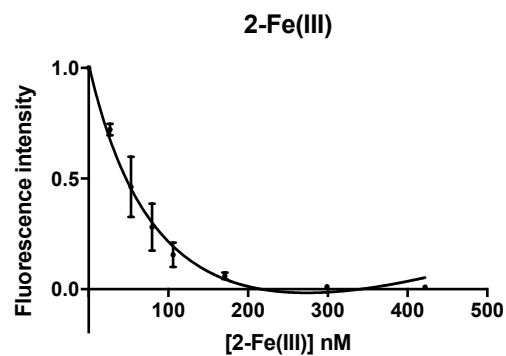
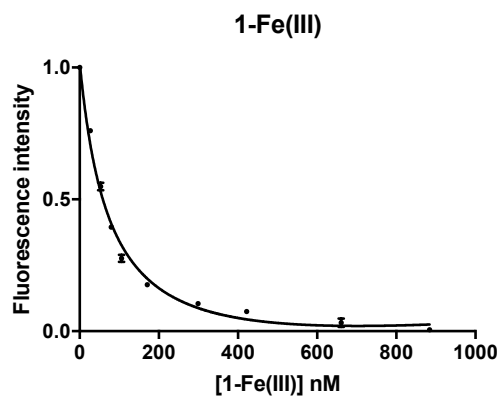


**Supplementary Figure 5.** Iron(III) exchange half-lives ( $t_{1/2}$ ) between ferrioxamine siderophores and EDTA in pH 7.4 buffer at room temperature determined from first-order rate plots. The  $t_{1/2}$  for FOB was too slow to measure an accurate first-order rate. Error bars represent standard deviations from the mean for at least three independent trials.

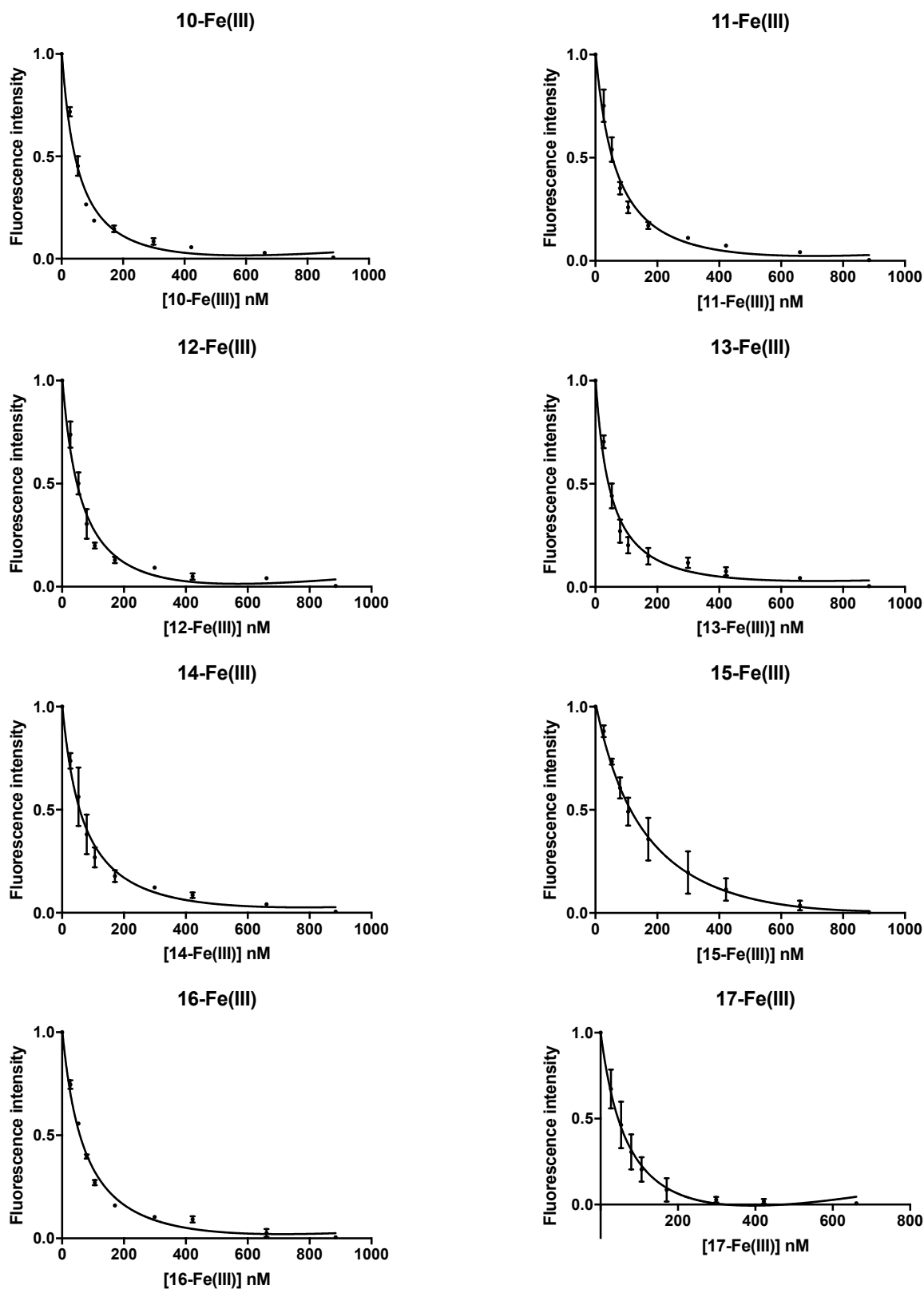


**Supplementary Figure 6.** SDS-PAGE analysis of purified *N*-His<sub>6</sub>-FhuD2. SDS-PAGE gel (Any kD, Bio-Rad) was loaded with protein ladder (ThermoFisher PageRuler Prestained Protein Ladder 10–180 kDa) in lane 1, Ni-NTA column flow through in lane 2, Ni-NTA column washes in lanes 3 and 4, and Ni-NTA elutions in lanes 5–9. Gels were stained with Coomassie blue and photographed. Image on left was not adjusted for contrast. Image on right was adjusted for contrast using Adobe Photoshop.

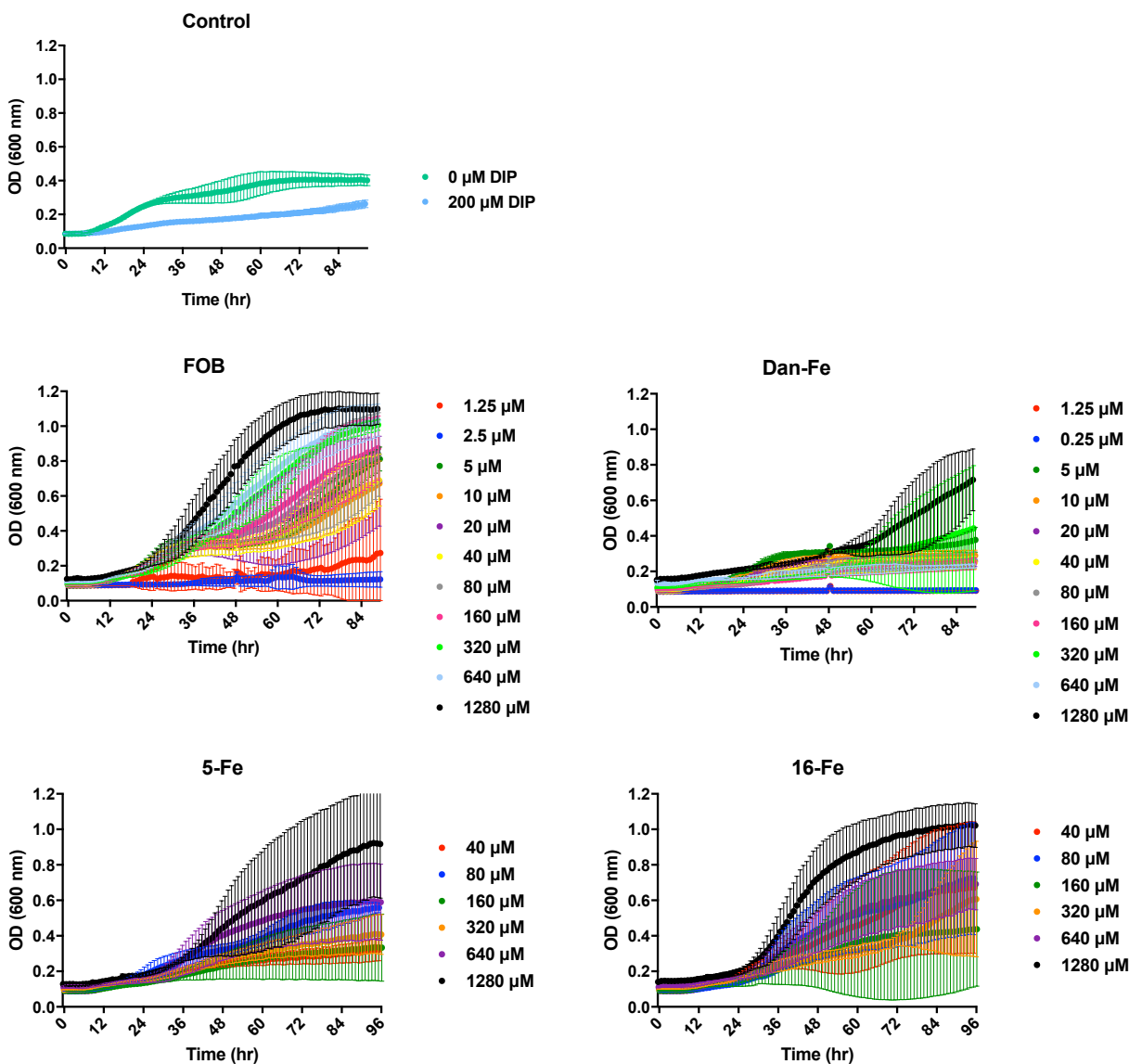








**Supplementary Figure 7.** Fluorescence quenching curves for titration of *N*-His<sub>6</sub>-FhuD2 with siderophores. Error bars represent standard deviations for at least three independent trials.



**Supplementary Figure 8.** *S. aureus* ATCC 11632 growth curves in iron restrictive TMS minimal media (pH 7.4) at 37 °C with siderophore supplementation. Error bars represent standard deviations for at least three independent trials.

### III. Supplementary Equations

Apparent siderophore Fe(III) affinity values ( $K_{Fe}$ ) were determined using a well established competition experiment with EDTA using optical absorbance spectroscopy<sup>2</sup>. Equations 1–11 and equilibria reactions were used to calculate  $K_{Fe}$ . Equations 1–11 show the equilibria for a general ligand (L), which in our case is the siderophore forming a 1:1 complex with Fe(III). We assumed there is no free iron in solution. We assumed that all absorbance at 430 nm was due to the presence of siderophore-Fe(III) complex and that a loss in absorbance at 430 nm is due to transfer of Fe(III) to EDTA. By starting with recrystallized highly pure siderophore-Fe(III) complex we initiated ligand exchange by addition of 1.2 equivalents of EDTA, as described in the materials and methods section of the main text. By monitoring the absorbance at 430 nm ( $Abs_{FeL}$ ) we calculated apparent  $K_{Fe}$  as shown below.

$$(1) \quad K_L = \frac{[FeL]}{[Fe^{3+}][L]} \quad \text{for the following equilibrium;} \quad [Fe^{3+}] + [L] \rightleftharpoons [FeL]$$

$$(2) \quad K_{FeEDTA} = \frac{[FeEDTA]}{[Fe^{3+}][EDTA]} \quad \text{for the following equilibrium; } [Fe^{3+}] + [EDTA] \rightleftharpoons [FeEDTA]$$

$$(3) \quad K_{Exchange} = \frac{K_L}{K_{FeEDTA}} \quad \text{for the following equilibrium; } [FeEDTA] + [L] \rightleftharpoons [FeL] + [EDTA]$$

$$(4) \quad K_{Exchange} = \frac{[FeL][EDTA]}{[FeEDTA][L]}$$

$$(5) \quad \Delta = \frac{Abs_{FeL} - Abs_{FeL+EDTA}}{\epsilon_L}$$

$$(6) \quad K_L = K_{FeEDTA} \times \frac{[FeL][EDTA]}{[FeEDTA][L]}$$

$$(7) \quad [FeL] = \frac{Abs_{FeL}}{\epsilon_L}$$

$$(8) \quad [EDTA] = [EDTA]_T - \Delta \quad \text{where} \quad [EDTA]_T = \text{total EDTA added}$$

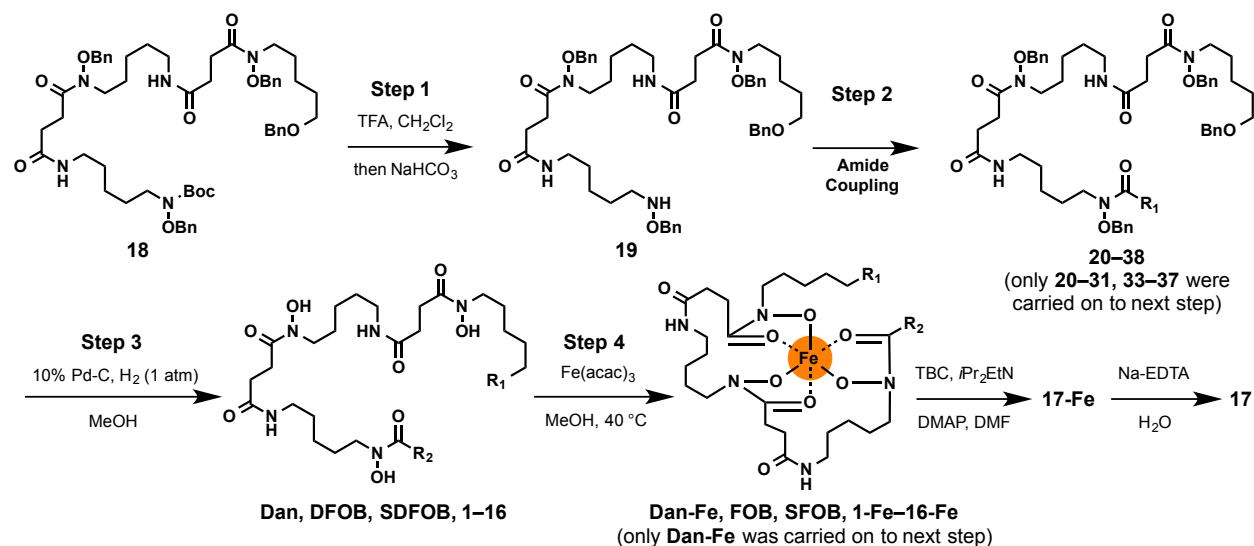
$$(9) \quad [FeEDTA] = \Delta$$

$$(10) \quad [L] = \Delta$$

$$(11) \quad K_{Fe} = \text{apparent } K_L$$

## IV. Synthesis of the Ferrioxamine Library

**General materials and methods.** The trihydroxamate siderophores **1–17** and the corresponding 1:1 iron(III) complexes **1-Fe–17-Fe** were synthesized as shown in **Supplementary Scheme 1**.<sup>3</sup> All reactions were carried out under a dry argon atmosphere unless otherwise stated. Dichloromethane ( $\text{CH}_2\text{Cl}_2$ ) was distilled from calcium hydride. Tetrahydrofuran was distilled from Na/benzophenone. Dimethylformamide (DMF), diisopropylamine ( $i\text{Pr}_2\text{EtN}$ ), and acetonitrile ( $\text{CH}_3\text{CN}$ ) were used from sealed anhydrous bottles purchased from Acros. Silica gel chromatography was performed using Sorbent Technologies silica gel 60 (32–63  $\mu\text{m}$ ).  $^1\text{H}$ -NMR and  $^{13}\text{C}$ -NMR spectra were recorded on a 600 MHz Varian DirectDrive spectrometer and FIDs were processed using ACD/ChemSketch version 10.04. Chemical shifts ( $\delta$ ) are given in parts per million (ppm) and are referenced to non-deuterated residual solvent. Coupling constants ( $J$ ) are reported in hertz (Hz). High-resolution mass spectrometry measurements were obtained on a Bruker micrOTOF II using electrospray ionization (ESI) in positive ion mode. Sample was introduced via flow injection at 4  $\mu\text{L}/\text{min}$  and mass spectra were recorded from 50–3000  $m/z$  for two min. HPLC was performed on a Waters 1525 binary pump instrument with a Waters 2487 dual  $\lambda$  absorbance detector set at 427 nm and 254 nm. The operating software for the HPLC was Breeze version 3.30. The HPLC column was a YMC Pro C18 reverse phase (3.0 x 50 mm) fit with a guard column (2.0 x 10 mm) of the same composition. Mobile phases were 10 mM ammonium acetate in  $\text{H}_2\text{O}$  (A) and 10 mM ammonium acetate in  $\text{CH}_3\text{CN}$  (B). A gradient was formed from 5%–80% (B) over 10 min, then 80%–95% (B) over 2 min, and then 95%–5% (B) over 3 min at a flow rate of 0.7 mL/min. Thin layer chromatography (TLC) was performed with aluminum-backed Merck RP-C18 F<sub>256</sub> silica gel plates using a 254 nm lamp or aqueous  $\text{FeCl}_3$  stain for visualization. IR spectra were recorded on a Bruker Tensor series FT-IR spectrometer using a diamond ATR accessory or as a thin film on NaCl disks. Melting points were determined in capillary tubes using a Thomas Hoover melting point apparatus and are uncorrected. The purity of compounds tested in biological assays was evaluated by analytical HPLC and verified to be  $\geq 95\%$  pure.

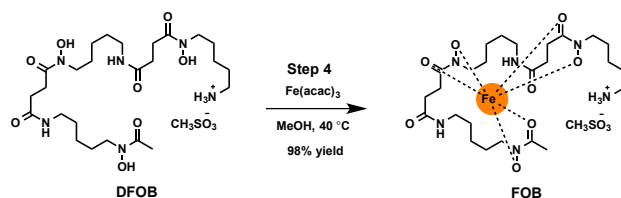


**Supplementary Scheme 1.** Synthesis of ferrioxamine siderophores used in this study.

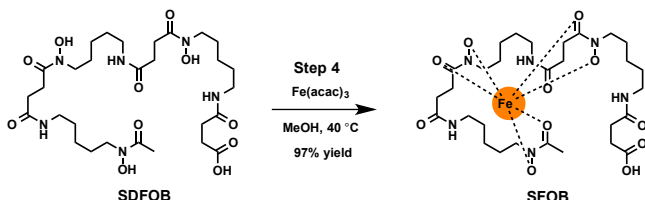
**Synthesis of siderophores.** The *N*-Boc-tetra-*O*-benzylated precursor **18** was prepared according to well established a literature protocol.<sup>4</sup> Treatment of **18** with TFA followed by an aqueous NaHCO<sub>3</sub> quench gave amine **19** according to a literature protocol (**Step 1**).<sup>4</sup> Amine **19** is the precursor for benzyl protected siderophores **20–38** (**Supplementary Fig. 7**) and deprotected siderophores **1–17**, **Dan**, and **Dan-Cip**. The synthesis and full characterization of siderophores **1**, **2**, **3**, **5**, **6**, **7**, **8**, and **17** and corresponding iron(III) complexes was reported previously.<sup>5</sup> The synthesis and full characterization of **Dan**, **Dan-Fe**, and **Dan-Cip** was reported previously.<sup>4,6</sup> **DFOB** was purchased from Sigma-Aldrich (St. Louis, MO). **SDFOB** was synthesized from **DFOB** as reported previously.<sup>7</sup> General procedures for **steps 3** and **4** are provided below along with detailed procedures for the synthesis of all new siderophores including benzyl protected precursors **22**, **23**, and **30–38**, deprotected siderophores **4** and **9–16**, and siderophore iron(III) complexes **FOB**, **SFOB**, and **4-Fe** and **9-Fe–16-Fe**. Siderophores were synthesized by Wencewicz at the University of Notre Dame and transferred to his laboratory at Washington University in St. Louis via a material transfer agreement.

**General procedure for step 3.** Benzyl protected siderophore (**22**, **23**, **30**, **31**, **33–37**) was dissolved in MeOH (~0.01 M) in an HCl-washed flask with a magnetic stir bar. The flask was flushed with argon, charged with 10% Pd-C (~10% w/w), and flushed with H<sub>2</sub> gas several times with intermediate vacuum evacuation. The mixture was left stirring under positive pressure from a H<sub>2</sub> balloon. Reaction progress was monitored by RP-C18 TLC (1.5:1 CH<sub>3</sub>CN:H<sub>2</sub>O; FeCl<sub>3</sub> stain). Once complete as judged by TLC the flask was flushed with argon and the mixture was vacuum filtered through a pad of celite and concentrated under reduced pressure. Crude product was dissolved in a minimal amount of MeOH and precipitated by adding Et<sub>2</sub>O. The siderophore (**4**, **9–16**) was isolated as a white solid after trituration with Et<sub>2</sub>O and drying under vacuum.

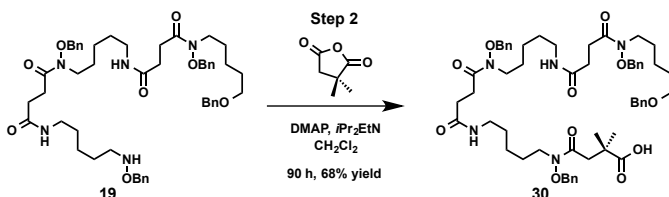
**General procedure for step 4.** The siderophore (**DFOB**, **SDFOB**, **4**, **9–16**) was dissolved in MeOH (0.002 M) at 40 °C (oil bath temp). Fe(acac)<sub>3</sub> (1.1 equiv) was added to give a clear, orange solution that was stirred for 2 hr. The MeOH was removed under reduced pressure using rotary evaporation to give the siderophore-iron(III) complex as an orange film. The crude product was dissolved in a minimal amount of MeOH and precipitated by addition of Et<sub>2</sub>O, trituated with Et<sub>2</sub>O, and dried under vacuum to give the pure siderophore-iron(III) complex (**FOB**, **SFOB**, **4-Fe**, **9-Fe–16-Fe**) as an orange powder.



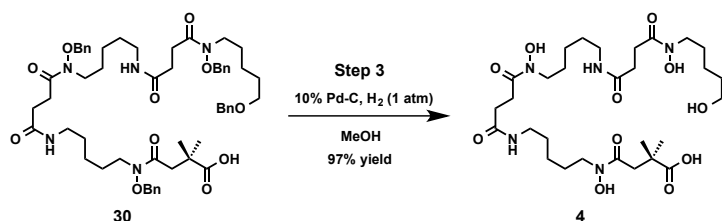
**FOB (mesylate salt).** Desferrioxamine B mesylate (**DFOB**; 47.5 mg, 0.072 mmol) was complexed to Fe(III) according to the general procedure for **step 4** to give siderophore-Fe(III) complex **FOB** in 98% yield as an orange powder (50.5 mg, 0.071 mmol). Mp 174–178 °C (dec.); HPLC retention time 2.06 min.



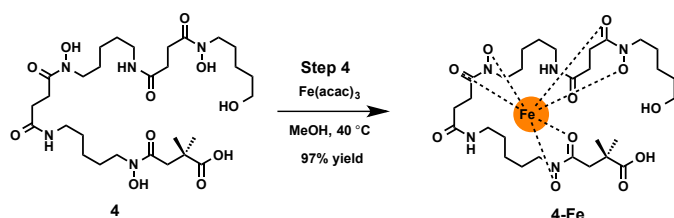
**SFOB.** Succinyl-desferrioxamine B (**SDFOB**; 51 mg, 0.077 mmol) was complexed to Fe(III) according to the general procedure for **step 4** to give siderophore-Fe(III) complex **SFOB** in 97% yield as an orange powder (53.2 mg, 0.075 mmol). Mp 143–148 °C; HPLC retention time 2.37 min.



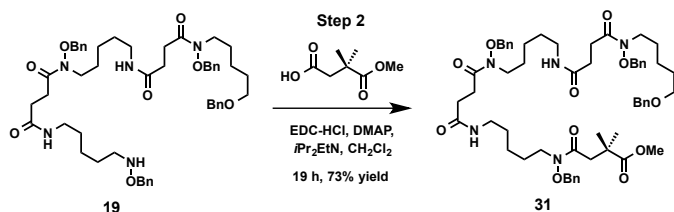
**Benzyl protected siderophore 30.** Hydroxylamine **19** (457.7 mg, 0.52 mmol), 2,2-dimethylsuccinic anhydride (150.0 mg, 0.71 mmol), *i*Pr<sub>2</sub>EtN (0.1 mL, 0.71 mmol), and catalytic DMAP (31 mg, 0.25 mmol) were dissolved in 8 mL of CH<sub>2</sub>Cl<sub>2</sub>, respectively. After 24 h, TLC (6% MeOH in CH<sub>2</sub>Cl<sub>2</sub>; FeCl<sub>3</sub> stain) showed only trace remaining starting material (**19**). After 90 h, the reaction was quenched with 1 N HCl (10 mL). The aq. layer was extracted with CH<sub>2</sub>Cl<sub>2</sub> (2 x 10 mL) and the combined CH<sub>2</sub>Cl<sub>2</sub> layers were washed with brine (10 mL), dried over anhydrous MgSO<sub>4</sub>, filtered, and concentrated under reduced pressure. This gave 579.6 mg of a cloudy, viscous oil that was purified by silica gel column chromatography (0.75 x 3 in silica gel; 3% MeOH in CH<sub>2</sub>Cl<sub>2</sub>). Pure product (**30**) was obtained in 68% yield as a clear, colorless oil (355.1 mg, 0.35 mmol). IR (thin film on NaCl plate) 3327, 2936, 2865, 1726, 1657, 1548, 1454, 1365, 1191 cm<sup>-1</sup>; <sup>1</sup>H-NMR (600 MHz, CDCl<sub>3</sub>) δ (ppm) 7.43–7.29 (m, 20 H), 6.81 (br s, 1 H), 6.58 (br s, 1 H), 4.85 (s, 4 H), 4.80 (s, 2 H), 4.48 (s, 2 H), 3.71–3.59 (m, 6 H), 3.44 (t, *J* = 6.5 Hz, 2 H), 3.23–3.16 (m, 4 H), 2.85 (t, *J* = 6.3 Hz, 2 H), 2.83–2.77 (m, 2 H), 2.69 (s, 2 H), 2.55 (t, *J* = 6.5 Hz, 2 H), 2.50 (t, *J* = 6.5 Hz, 2 H), 1.67–1.58 (m, 8 H), 1.52–1.42 (m, 4 H), 1.39–1.27 (m, 6 H), 1.25 (s, 6 H); <sup>13</sup>C-NMR (150 MHz, CDCl<sub>3</sub>) δ (ppm) 179.4, 174.5, 173.8, 173.2, 172.4, 172.3, 138.5, 134.3, 134.2, 129.2, 129.1, 129.0, 128.9, 128.8, 128.7, 128.6, 128.6, 128.3, 127.5, 127.4, 76.3, 76.3, 76.0, 72.8, 70.0, 45.4, 44.8, 43.9, 42.1, 39.9, 39.3, 39.2, 30.5, 30.4, 29.3, 28.6, 28.1, 28.0, 27.9, 26.6, 26.4, 26.3, 25.9, 23.6, 23.4, 23.3; HRMS–FAB (*m/z*): [*M*+H]<sup>+</sup> calcd. for C<sub>57</sub>H<sub>78</sub>N<sub>5</sub>O<sub>11</sub>: 1008.5698, found 1008.5700.



**Siderophore 4.** Benzyl protected siderophore **30** (51.8 mg, 0.051 mmol) was deprotected according to the general procedure for **step 3** to give the siderophore **4** in 97% yield as a white solid (32.0 mg, 0.049 mmol).  $^1\text{H-NMR}$  (600 MHz,  $\text{CD}_3\text{OD}$ )  $\delta$  (ppm) 3.62–3.57 (m, 4 H), 3.55 (t,  $J = 6.5$  Hz, 2 H), 3.21–3.13 (m, 6 H), 2.76 (t,  $J = 7.2$  Hz, 2 H), 2.51 (s, 2 H), 2.48–2.43 (m, 6 H), 1.70–1.60 (m, 4 H), 1.59–1.47 (m, 8 H), 1.44–1.29 (m, 6 H), 1.23 (s, 6 H); HRMS–ESI ( $m/z$ ):  $[\text{M}+\text{Na}]^+$  calcd. For  $\text{C}_{29}\text{H}_{53}\text{N}_5\text{NaO}_{11}$ : 670.3634, found 670.3621.

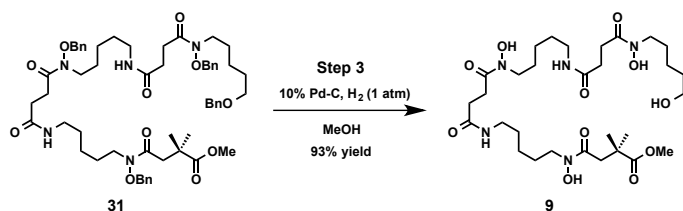


**Siderophore-Fe(III) complex 4-Fe.** Siderophore **4** (13.2 mg, 0.02 mmol) was complexed to Fe(III) according to the general procedure for **step 4** to give siderophore-Fe(III) complex **4-Fe** in 97% as an orange powder (13.9 mg, 0.02 mmol). Mp 130–140 °C (dec.); HRMS–ESI ( $m/z$ ):  $[\text{M}+\text{Na}]^+$  calcd. for  $\text{C}_{29}\text{H}_{50}\text{FeN}_5\text{NaO}_{11}$ : 723.2749, found 723.2742; HPLC retention time 2.80 min.

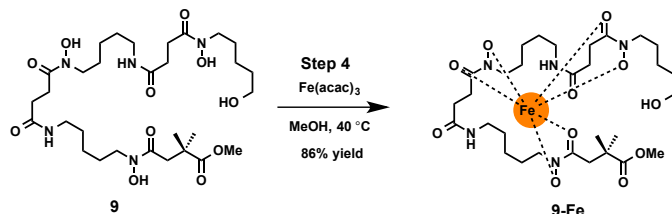


**Benzyl protected siderophore 31.** Hydroxylamine **19** (147.0 mg, 0.17 mmol), 3-(methoxycarbonyl)-3-methylbutanoic acid<sup>8</sup> (52.0 mg, 0.32 mmol),  $i\text{Pr}_2\text{EtN}$  (28.0 mg, 0.22 mmol), catalytic DMAP (5.1 mg, 0.04 mmol), and EDC-HCl (80.0 mg, 0.42 mmol) were dissolved in 10 mL of  $\text{CH}_2\text{Cl}_2$ . After 19 h, TLC (6% MeOH in  $\text{CH}_2\text{Cl}_2$ ;  $\text{FeCl}_3$  stain) showed no remaining starting material (**19**). The mixture was diluted with  $\text{CH}_2\text{Cl}_2$  (10 mL), washed with 10% aq. citric acid (2 x 10 mL), brine (10 mL), 10% aq.  $\text{NaHCO}_3$  (10 mL), and brine (10 mL), dried over anhydrous  $\text{MgSO}_4$ , filtered, and concentrated under reduced pressure to give 159.8 mg of a clear, colorless, viscous oil. The crude product was purified by silica gel column chromatography (1 x 3 in silica gel; 3%–6% MeOH in  $\text{CH}_2\text{Cl}_2$ ). Pure product (**31**) was isolated in 73% yield as a clear, colorless, viscous oil (125.2 mg, 0.12 mmol).  $^1\text{H-NMR}$  (600 MHz,  $\text{CDCl}_3$ )  $\delta$  (ppm) 7.43–7.29 (m, 20 H), 6.32 (br s, 2 H), 4.85 (s, 2 H), 4.85 (s, 2 H), 4.79 (s, 2 H), 4.48 (s, 2 H), 3.66 (s, 3 H), 3.65–3.56 (m, 6 H), 3.44 (t,  $J = 6.5$  Hz, 2 H), 3.22–3.17 (m, 4 H), 2.84–2.77 (m, 4 H), 2.67 (s, 2 H), 2.52–2.45 (m, 4 H), 1.67–1.59 (m, 8 H), 1.53–1.45 (m, 4 H), 1.40–1.33 (m, 2 H), 1.32–1.25 (m, 4 H), 1.22 (s, 6 H);  $^{13}\text{C-NMR}$  (150 MHz,  $\text{CDCl}_3$ )  $\delta$  (ppm) 177.9, 174.0, 173.8, 172.4, 172.1, 172.1, 138.5, 134.5, 134.5, 134.3, 129.1, 129.1, 128.9, 128.9, 128.8, 128.7, 128.7, 128.6, 128.3, 127.5, 127.4, 76.3, 76.3, 76.1, 72.8, 70.1, 51.9, 45.5, 44.9,

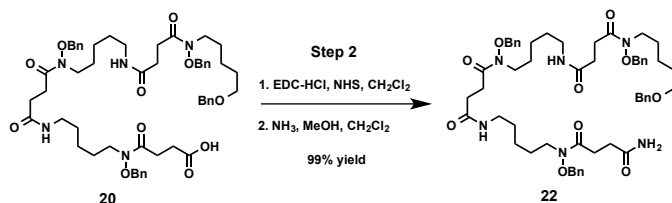
44.7, 42.4, 40.1, 39.3, 39.3, 30.7, 30.5, 29.3, 29.1, 28.4, 28.1, 28.0, 26.7, 26.4, 26.4, 25.6, 25.3, 23.9, 23.6, 23.4; HRMS–ESI ( $m/z$ ):  $[M+Na]^+$  calcd. for  $C_{58}H_{79}N_5NaO_{11}$ : 1044.5668, found 1044.5663.



**Siderophore 9.** Benzyl protected siderophore **31** (64.4 mg, 0.063 mmol) was deprotected according to the general procedure for **step 3** to give siderophore **9** in 93% yield as a white solid (38.7 mg, 0.058 mmol). Mp 138–140 °C;  $^1H$ -NMR (600 MHz,  $CD_3OD$ )  $\delta$  (ppm) 3.65 (s, 3 H), 3.59 (t,  $J$  = 6.5 Hz, 4 H), 3.55 (t,  $J$  = 6.9 Hz, 2 H), 3.55 (t,  $J$  = 6.5 Hz, 2 H), 3.16 (t,  $J$  = 6.9 Hz, 4 H), 2.79 (s, 2 H), 2.76 (t,  $J$  = 7.2 Hz, 4 H), 2.45 (t,  $J$  = 7.2 Hz, 4 H), 1.67–1.59 (m, 6 H), 1.59–1.49 (m, 6 H), 1.40–1.29 (m, 6 H), 1.24 (s, 6 H);  $^{13}C$ -NMR (150 MHz,  $CD_3OD$ )  $\delta$  (ppm) 180.1, 175.1, 175.0, 174.6, 174.5, 173.2, 62.9, 52.5, 49.0, 48.8, 43.5, 41.4, 40.4, 33.4, 31.6, 31.6, 30.1, 30.1, 29.1, 29.1, 27.7, 27.5, 26.2, 25.1, 25.0, 24.1; HRMS–ESI ( $m/z$ ):  $[M+H]^+$  calcd. for  $C_{30}H_{56}N_5O_{11}$ : 662.3971, found 662.3971.



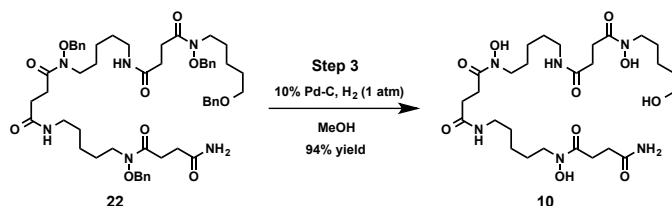
**Siderophore-Fe(III) complex 9-Fe.** Siderophore **9** (11.5 mg, 0.017 mmol) was complexed to Fe(III) according to the general procedure for **step 4** to give siderophore-Fe(III) complex **9-Fe** in 86% yield as an orange powder (10.6 mg, 0.015 mmol). Mp 170–173 °C; HRMS–ESI ( $m/z$ ):  $[M+H]^+$  calcd. for  $C_{30}H_{53}FeN_5O_{11}$ : 715.3086, found 715.3089; HPLC retention time 4.52 min.



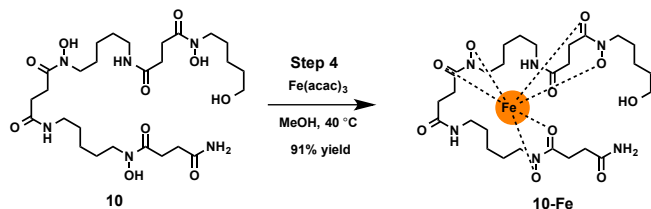
**Benzyl protected siderophore 22.** Benzyl protected siderophore **20** (298.2 mg, 0.30 mmol) was reacted with EDC-HCl (146.0mg, 0.76 mmol) and *N*-hydroxysuccinimide (NHS; 70.0 mg, 0.608 mmol) in 5 mL of  $CH_2Cl_2$  for 2 h. After an aqueous work-up, the NHS activated ester of **20** was isolated in 95% yield as a clear, viscous oil (312.4 mg, 0.29 mmol). The NHS activated ester of **20** (52.1 mg, 0.048 mmol) was reacted with  $NH_3$  in MeOH (7N, 2.0 mL, 49 mmol) in  $CH_2Cl_2$  (2 mL) for 2.5 h. All the volatiles were removed under reduced pressure giving 66.6 mg of a clear, tan oil. The crude product was purified by silica gel column chromatography (1 x 3 in silica gel; 3%–6% MeOH in  $CH_2Cl_2$ ) to give the desired product (**22**) in 99% yield as a clear, colorless, viscous oil (47.5 mg, 0.049 mmol).  $^1H$ -NMR (600 MHz,  $CDCl_3$ )  $\delta$  (ppm) 7.41–7.29 (m, 20 H), 6.91 (br s, 1 H), 6.50 (br s, 1 H), 6.36 (br s, 1 H), 6.06 (br s, 1 H), 4.85 (s, 2 H), 4.84 (s, 2 H),



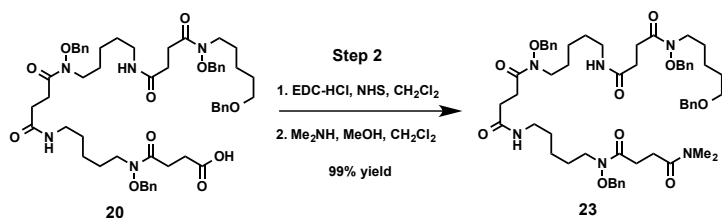
4.84 (s, 2 H), 4.48 (s, 2 H), 3.70–3.65 (m, 2 H), 3.65–3.59 (m, 4 H), 3.44 (t,  $J = 6.5$  Hz, 2 H), 3.22–3.15 (m, 4 H), 2.83–2.77 (m, 4 H), 2.77–2.73 (m, 2 H), 2.57–2.50 (m, 4 H), 2.48 (t,  $J = 6.5$  Hz, 2 H), 1.67–1.58 (m, 8 H), 1.51–1.44 (m, 4 H), 1.39–1.23 (m, 6 H);  $^{13}\text{C}$ -NMR (150 MHz,  $\text{CDCl}_3$ )  $\delta$  (ppm) 174.8, 174.2, 174.0, 173.8, 172.4, 172.3, 138.5, 134.4, 134.3, 129.2, 129.2, 129.1, 128.9, 128.8, 128.7, 128.3, 127.6, 127.5, 76.3, 76.1, 72.8, 70.1, 63.7, 45.5, 44.8, 44.5, 39.4, 39.2, 30.6, 30.5, 29.8, 29.6, 29.3, 28.6, 28.3, 28.1, 28.0, 27.3, 26.6, 26.4, 26.3, 23.6, 23.4; HRMS-ESI ( $m/z$ ):  $[\text{M}+\text{H}]^+$  calcd. for  $\text{C}_{55}\text{H}_{75}\text{N}_6\text{O}_{10}$ : 979.5539, found 979.5531.



**Siderophore 10.** Benzyl protected siderophore **22** (43.0 mg, 0.044 mmol) was deprotected according to the general procedure for **step 3** to give siderophore **10** in 94% yield as an off-white solid (25.5 mg, 0.041 mmol). Mp 152–155 °C;  $^1\text{H}$ -NMR (600 MHz,  $\text{DMSO}-d_6$ )  $\delta$  (ppm) 9.61 (br s, N-OH, 3 H), 7.78 (t,  $J = 5.4$  Hz, 2 H), 7.28 (br s, 1 H), 6.72 (br s, 1 H), 4.35 (t,  $J = 5.0$  Hz, 1 H), 3.45 (t,  $J = 7.0$  Hz, 6 H), 3.39–3.34 (m, 2 H), 2.99 (q,  $J = 6.7$  Hz, 4 H), 2.60–2.54 (m, 6 H), 2.26 (t,  $J = 7.2$  Hz, 6 H), 1.53–1.46 (m, 6 H), 1.44–1.34 (m, 6 H), 1.27–1.17 (m, 6 H);  $^{13}\text{C}$ -NMR (150 MHz,  $\text{DMSO}-d_6$ )  $\delta$  (ppm) 173.6, 172.0, 171.9, 171.9, 171.3, 60.6, 47.2, 47.1, 38.4, 32.2, 29.9, 29.5, 28.8, 27.6, 27.3, 26.2, 26.0, 23.5, 22.7; HRMS-ESI ( $m/z$ ):  $[\text{M}+\text{H}]^+$  calcd. for  $\text{C}_{27}\text{H}_{51}\text{N}_6\text{O}_{10}$ : 619.3661, found 619.3654.

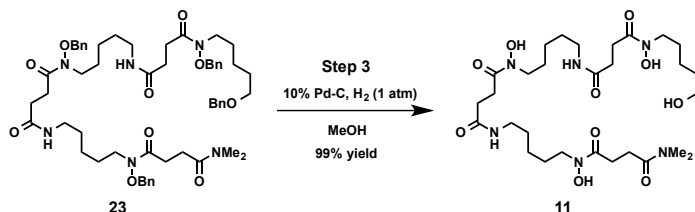


**Siderophore-Fe(III) complex 10-Fe.** Siderophore **10** (8.0 mg, 0.013 mmol) was complexed to Fe(III) according to the general procedure for **step 4** to give siderophore-Fe(III) complex **10-Fe** in 91% yield as an orange powder (7.9 mg, 0.012 mmol). Mp 138–144 °C (dec.); HRMS-ESI ( $m/z$ ):  $[\text{M}+\text{H}]^+$  calcd. for  $\text{C}_{27}\text{H}_{48}\text{FeN}_6\text{O}_{10}$ : 672.2776, found 672.2763; HPLC retention time 2.53 min.

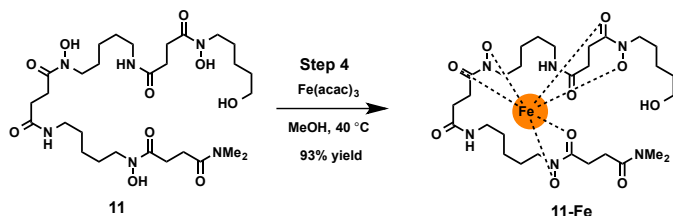


**Benzyl protected siderophore 23.** The NHS activated ester of **20** (52.1 mg, 0.048 mmol) was reacted with  $\text{Me}_2\text{NH}$  in MeOH (2.0 M, 2.0 mL, 4.0 mmol) in  $\text{CH}_2\text{Cl}_2$  (2 mL) for 2.5 h. All the volatiles were removed under reduced pressure to give 61.7 mg of a clear, viscous oil. The crude product was purified by silica gel column chromatography (1 x 3 in silica gel; 3%–6% MeOH in  $\text{CH}_2\text{Cl}_2$ ) to give pure product (**23**) in 99% yield as a clear, colorless, viscous oil (48.0 mg, 0.048

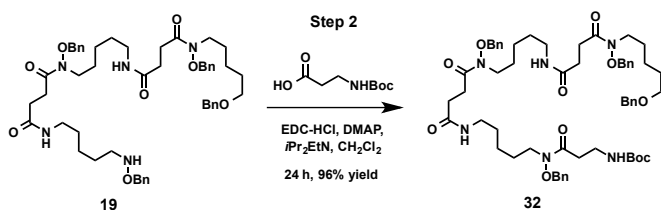
mmol).  $^1\text{H-NMR}$  (600 MHz,  $\text{CDCl}_3$ )  $\delta$  (ppm) 7.42–7.30 (m, 20 H), 6.56 (br s, 1 H), 6.36 (br s, 1 H), 4.90 (s, 2 H), 4.85 (s, 2 H), 4.84 (s, 2 H), 4.47 (s, 2 H), 3.68–3.59 (m, 6 H), 3.44 (t,  $J = 6.5$  Hz, 2 H), 3.22–3.16 (m, 4 H), 3.03 (s, 3 H), 2.94 (s, 3 H), 2.83–2.75 (m, 6 H), 2.65–2.60 (m, 2 H), 2.48 (t,  $J = 6.6$  Hz, 4 H), 1.67–1.58 (m, 8 H), 1.52–1.45 (m, 4 H), 1.39–1.25 (m, 6 H);  $^{13}\text{C-NMR}$  (150 MHz,  $\text{CDCl}_3$ )  $\delta$  (ppm) 174.0, 173.9, 173.8, 172.2, 172.1, 171.8, 138.5, 134.6, 134.3, 129.2, 129.1, 129.1, 128.8, 128.7, 128.6, 128.6, 128.3, 127.5, 127.4, 76.3, 76.3, 76.2, 72.8, 70.1, 63.8, 45.5, 45.0, 44.8, 39.3, 39.3, 37.1, 35.5, 30.7, 30.5, 29.6, 29.3, 28.9, 28.5, 28.1, 27.9, 27.5, 27.4, 26.7, 26.4, 26.3, 25.3, 23.9, 23.6, 23.4; HRMS–ESI ( $m/z$ ):  $[\text{M}+\text{Na}]^+$  calcd. for  $\text{C}_{57}\text{H}_{78}\text{N}_6\text{NaO}_{10}$ : 1029.5672, found 1029.5674.



**Siderophore 11.** Benzyl protected siderophore **23** (44.5 mg, 0.044 mmol) was deprotected according to the general procedure for **step 3** to give siderophore **11** in 99% yield as a white solid (28.0 mg, 0.043 mmol). Mp 143–145 °C;  $^1\text{H-NMR}$  (600 MHz,  $\text{CD}_3\text{OD}$ )  $\delta$  (ppm) 3.60 (t,  $J = 6.9$  Hz, 6 H), 3.55 (t,  $J = 6.6$  Hz, 2 H), 3.16 (t,  $J = 6.5$  Hz, 4 H), 3.09 (s, 3 H), 2.93 (s, 3 H), 2.80–2.73 (m, 6 H), 2.64 (t,  $J = 6.6$  Hz, 2 H), 2.45 (t,  $J = 7.2$  Hz, 4 H), 1.68–1.60 (m, 6 H), 1.59–1.48 (m, 6 H), 1.40–1.29 (m, 6 H);  $^{13}\text{C-NMR}$  (150 MHz,  $\text{CD}_3\text{OD}$ )  $\delta$  (ppm) 175.1, 175.1, 174.8, 174.6, 174.5, 62.9, 40.4, 37.8, 36.0, 33.4, 31.6, 30.1, 30.1, 29.1, 28.9, 28.6, 27.7, 27.5, 25.1, 25.0, 24.1; HRMS–ESI ( $m/z$ ):  $[\text{M}+\text{H}]^+$  calcd. for  $\text{C}_{29}\text{H}_{55}\text{N}_6\text{O}_{10}$ : 647.3974, found 647.3984.

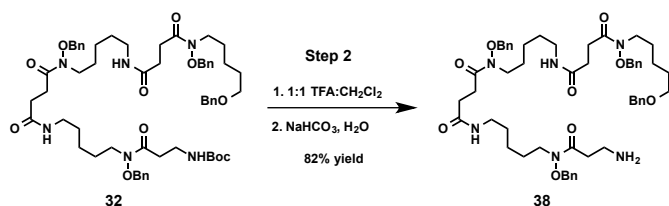


**Siderophore-Fe(III) complex 11-Fe.** Siderophore **11** (10.0 mg, 0.0155 mmol) was complexed to Fe(III) according to the general procedure for **step 4** to give siderophore-Fe(III) complex **11-Fe** in 93% yield as an orange powder (10.0 mg, 0.014 mmol). Mp 114–119 °C (dec.); HRMS–ESI ( $m/z$ ):  $[\text{M}+\text{Na}]^+$  calcd. for  $\text{C}_{29}\text{H}_{51}\text{FeN}_6\text{NaO}_{10}$ : 722.2908, found 722.2888; HPLC retention time 3.07 min.

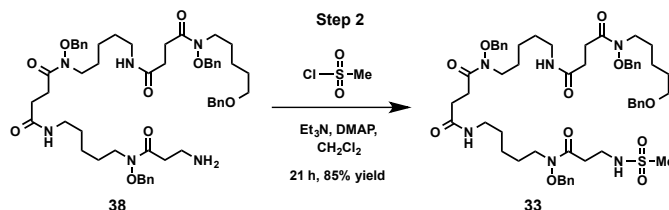


**Benzyl protected siderophore 32.** Hydroxylamine **19** (224.0 mg, 0.25 mmol), *N*-Boc- $\beta$ -alanine (52.0 mg, 0.275 mmol),  $i\text{Pr}_2\text{EtN}$  (0.05 mL, 0.29 mmol), catalytic DMAP (4.5 mg, 0.037 mmol), and EDC-HCl (125.0 mg, 0.65 mmol) were dissolved in 8 mL of  $\text{CH}_2\text{Cl}_2$ . After 24 h, TLC (5% MeOH in  $\text{CHCl}_3$ ;  $\text{FeCl}_3$  stain) showed no remaining starting material (**19**). The mixture was

diluted with CH<sub>2</sub>Cl<sub>2</sub> (25 mL) and washed with 10% aq. citric acid (2 x 10 mL), brine (10 mL), 10% aq. NaHCO<sub>3</sub> (2 x 10 mL), and brine (10 mL), dried over anhydrous MgSO<sub>4</sub>, filtered, and concentrated under reduced pressure. This gave 263.0 mg of a cloudy, waxy oil that was purified by silica gel column chromatography (0.5 x 4 in silica gel; 3% MeOH in CHCl<sub>3</sub>). Pure product (**32**) was obtained in 96% yield as a waxy solid (253.6 mg, 0.24 mmol). Mp 68–70 °C; IR (thin film on NaCl plate) 3327, 2936, 2863, 1707, 1653, 1545, 1454, 1412, 1366, 1252, 1172 cm<sup>-1</sup>; <sup>1</sup>H-NMR (600 MHz, CDCl<sub>3</sub>) δ (ppm) 7.40–7.22 (m, 20 H), 6.41 (br s, 1 H), 6.41 (br s, 1 H), 5.29 (br s, 1 H), 4.84 (s, 4 H), 4.76 (br s, 2 H), 4.46 (s, 2 H), 3.67–3.57 (m, 6 H), 3.43 (t, *J* = 6.5 Hz, 2 H), 3.40–3.35 (m, 2 H), 3.21–3.15 (m, 4 H), 2.83–2.75 (m, 4 H), 2.64–2.58 (m, 2 H), 2.51–2.44 (m, 4 H), 1.66–1.57 (m, 8 H), 1.51–1.45 (m, 4 H), 1.42 (s, 9 H), 1.38–1.32 (m, 2 H), 1.32–1.23 (m, 4 H); <sup>13</sup>C-NMR (150 MHz, CDCl<sub>3</sub>) δ (ppm) 173.9, 173.7, 173.4, 172.0, 172.0, 155.8, 138.4, 134.2, 134.0, 129.1, 129.1, 128.9, 128.8, 128.7, 128.6, 128.5, 128.5, 128.2, 127.4, 127.3, 78.8, 76.2, 76.2, 72.7, 69.9, 45.4, 45.0, 44.6, 39.2, 35.8, 32.5, 30.5, 30.4, 29.2, 28.9, 28.3, 28.0, 27.8, 26.5, 26.3, 26.2, 23.8, 23.5, 23.3; HRMS-ESI (*m/z*): [M+Na]<sup>+</sup> calcd. for C<sub>59</sub>H<sub>82</sub>N<sub>6</sub>NaO<sub>11</sub>: 1073.5934, found 1073.5959.

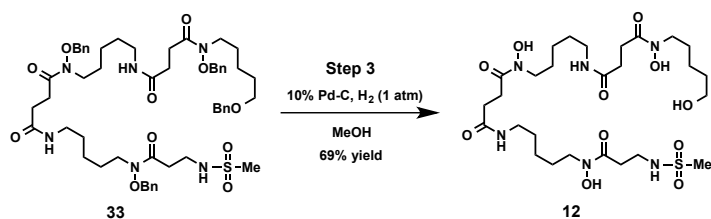


**Benzyl protected siderophore 38.** *N*-Boc-*O*-Benzyl protected siderophore **32** (121.0 mg, 0.115 mmol) was dissolved in a 1:1 mixture of TFA:CH<sub>2</sub>Cl<sub>2</sub> (4 mL). After 1.5 h, the TFA/CH<sub>2</sub>Cl<sub>2</sub> were evaporated and the resulting oil was dissolved in CHCl<sub>3</sub> (15 mL) and washed with satd. aq. NaHCO<sub>3</sub> (20 mL). The layers were separated and the CHCl<sub>3</sub> was dried over anhydrous MgSO<sub>4</sub>, filtered, and concentrated under reduced pressure to give the desired siderophore amine **38** in 82% yield as a clear, colorless, viscous oil (89.4 mg, 0.094 mmol). This material was used immediately in the next reactions without purification. <sup>1</sup>H-NMR (500 MHz, CDCl<sub>3</sub>) δ (ppm) 7.47–7.22 (m, 20 H), 6.41 (br s, 2 H), 4.85 (s, 4 H), 4.81 (s, 2 H), 4.47 (s, 2 H), 3.71–3.56 (m, 6 H), 3.44 (t, *J* = 6.4 Hz, 2 H), 3.24–3.15 (m, 4 H), 2.95 (t, *J* = 5.5 Hz, 2 H), 2.85–2.74 (m, 4 H), 2.59–2.52 (m, 2 H), 2.52–2.41 (m, 4 H), 1.75 (br s, 2 H), 1.69–1.56 (m, 8 H), 1.54–1.44 (m, 4 H), 1.40–1.21 (m, 6 H); <sup>13</sup>C-NMR (125 MHz, CDCl<sub>3</sub>) δ (ppm) 174.0, 173.8, 172.1, 172.1, 138.5, 134.3, 129.1, 129.1, 128.9, 128.9, 128.8, 128.7, 128.7, 128.6, 128.3, 127.5, 127.4, 76.3, 76.3, 76.2, 72.8, 70.1, 45.5, 44.9, 44.7, 39.3, 37.5, 35.7, 30.7, 30.5, 29.6, 29.3, 29.0, 28.4, 28.1, 28.0, 26.6, 26.5, 26.3, 23.9, 23.6, 23.4.

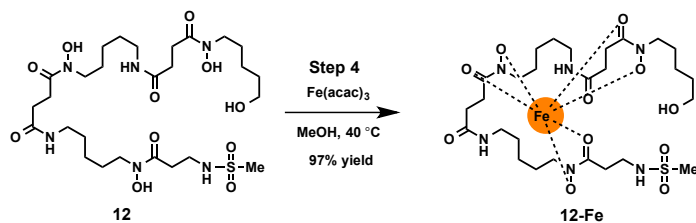


**Benzyl protected siderophore 33.** Amine **38** (29.8 mg, 0.313 mmol) was reacted with methanesulfonyl chloride (5.5 mg, 0.048 mmol) in the presence of Et<sub>3</sub>N (0.013 mL, 0.093 mmol), and catalytic DMAP (1.0 mg, 0.008 mmol) in 3 mL of CH<sub>2</sub>Cl<sub>2</sub>. After 21 h, TLC (5% MeOH in CH<sub>2</sub>Cl<sub>2</sub>; FeCl<sub>3</sub> stain) showed no remaining starting material (**38**). The reaction mixture

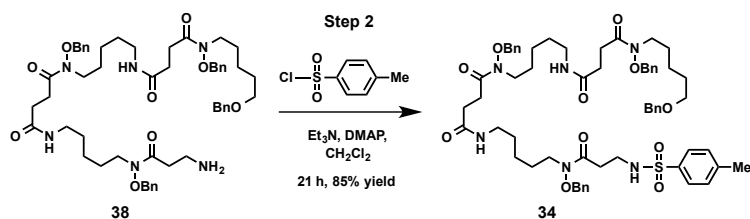
was diluted with CH<sub>2</sub>Cl<sub>2</sub> (10 mL), washed with 10% aq. citric acid (2 x 10 mL), brine (10 mL), 10% aq. NaHCO<sub>3</sub> (10 mL), and brine (10 mL), dried over anhydrous MgSO<sub>4</sub>, filtered, and concentrated to give 29 mg of a clear, colorless, viscous oil. The crude product was purified by silica gel column chromatography (0.5 x 4 in silica gel; 3%–5% MeOH in CH<sub>2</sub>Cl<sub>2</sub>) and pure product (**33**) was obtained in 85% yield as a clear, colorless, viscous oil (27.2 mg, 0.026 mmol). <sup>1</sup>H-NMR (600 MHz, CDCl<sub>3</sub>) δ (ppm) 7.43–7.24 (m, 20 H), 6.40 (br s, 1 H), 6.37 (br s, 1 H), 5.50 (t, *J* = 5.9 Hz, 1 H), 4.85 (s, 6 H), 4.80 (s, 2 H), 4.48 (s, 2 H), 3.70–3.57 (m, 6 H), 3.44 (t, *J* = 6.6 Hz, 2 H), 3.38–3.29 (m, 2 H), 3.23–3.15 (m, 4 H), 2.93 (s, 3 H), 2.85–2.74 (m, 4 H), 2.68 (t, *J* = 4.8 Hz, 2 H), 2.55–2.43 (m, 4 H), 1.69–1.55 (m, 8 H), 1.54–1.44 (m, 4 H), 1.41–1.33 (m, 2 H), 1.33–1.21 (m, 4 H); <sup>13</sup>C-NMR (150 MHz, CDCl<sub>3</sub>) δ (ppm) 174.1, 173.8, 172.8, 172.2, 172.2, 138.5, 134.3, 134.0, 129.3, 129.1, 129.1, 128.9, 128.9, 128.8, 128.7, 128.6, 128.3, 127.6, 127.5, 76.3, 76.3, 76.3, 72.8, 70.1, 45.5, 44.9, 44.7, 40.0, 39.3, 38.7, 32.8, 30.7, 30.6, 29.7, 29.3, 29.0, 28.4, 28.1, 28.0, 26.7, 26.4, 26.3, 23.8, 23.6, 23.4; HRMS–ESI (*m/z*): [*M*+*H*]<sup>+</sup> calcd. for C<sub>55</sub>H<sub>77</sub>N<sub>6</sub>O<sub>11</sub>S: 1029.5366, found 1029.5393.



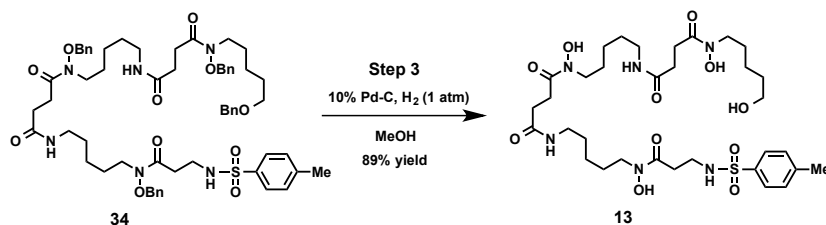
**Siderophore 12.** Benzyl protected siderophore **33** (21.0 mg, 0.020 mmol) was deprotected according to the general procedure for **step 3** to give siderophore **12** in 69% yield as an off-white solid (9.2 mg, 0.014 mmol). Mp 120–123 °C; <sup>1</sup>H-NMR (600 MHz, CD<sub>3</sub>OD) δ (ppm) 3.64–3.57 (m, 6 H), 3.55 (t, *J* = 6.6 Hz, 2 H), 3.33 (d, *J* = 7.0 Hz, 2 H), 3.17 (t, *J* = 6.7 Hz, 2 H), 3.16 (t, *J* = 6.7 Hz, 2 H), 2.95 (s, 3 H), 2.79–2.74 (m, 6 H), 2.45 (t, *J* = 7.2 Hz, 4 H), 1.68–1.60 (m, 6 H), 1.59–1.49 (m, 6 H), 1.40–1.30 (m, 6 H); <sup>13</sup>C-NMR (150 MHz, CD<sub>3</sub>OD) δ (ppm) 175.1, 62.9, 49.7, 40.4, 40.0, 33.4, 31.8, 31.7, 30.1, 30.1, 29.1, 27.7, 27.5, 25.1, 25.0, 24.1 (compound precipitated in NMR tube during FID acquisition); HRMS–ESI (*m/z*): [*M*+*H*]<sup>+</sup> calcd. for C<sub>27</sub>H<sub>53</sub>N<sub>6</sub>O<sub>11</sub>S: 669.3488, found 669.3493.



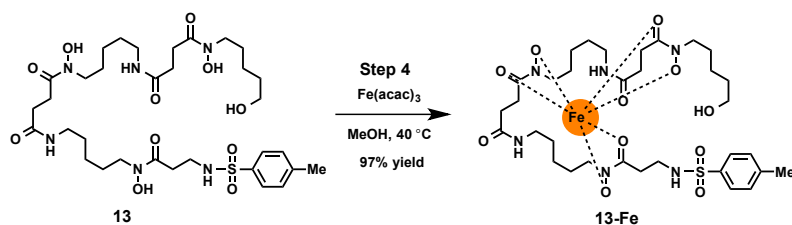
**Siderophore-Fe(III) complex 12-Fe.** Siderophore **12** (3.0 mg, 0.0045 mmol) was complexed to Fe(III) according to the general procedure for **step 4** to give siderophore-Fe(III) complex **12-Fe** in 97% yield as an orange powder (3.1 mg, 0.0043 mmol). Mp 147–152 °C (dec.); HRMS–ESI (*m/z*): [*M*+*H*]<sup>+</sup> calcd. for C<sub>27</sub>H<sub>50</sub>FeN<sub>6</sub>O<sub>11</sub>S: 722.2602, found 722.2617; HPLC retention time 3.12 min.



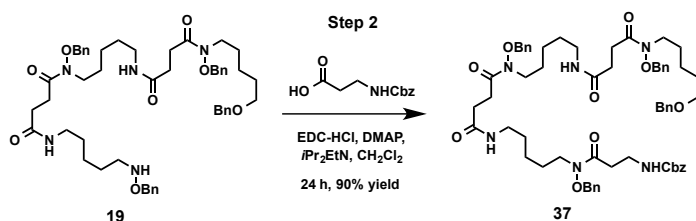
**Benzyl protected siderophore 34.** Amine **38** (29.8 mg, 0.31 mmol) was reacted with *p*-toluenesulfonyl chloride (9.0 mg, 0.047 mmol) in the presence of Et<sub>3</sub>N (0.013 mL, 0.093 mmol), and catalytic DMAP (1.0 mg, 0.008 mmol) in 3 mL of CH<sub>2</sub>Cl<sub>2</sub>. After 21 h, TLC (5% MeOH in CH<sub>2</sub>Cl<sub>2</sub>; FeCl<sub>3</sub> stain) showed no remaining starting material (**38**). The reaction mixture was diluted with CH<sub>2</sub>Cl<sub>2</sub> (10 mL), washed with 10% aq. citric acid (2 x 10 mL), brine (10 mL), 10% aq. NaHCO<sub>3</sub> (10 mL), and brine (10 mL), dried over anhydrous MgSO<sub>4</sub>, filtered, and concentrated to give 32.0 mg of a clear, colorless, viscous oil. The crude product was purified by silica gel column chromatography (0.5 x 4 in silica gel; 3%–5% MeOH in CH<sub>2</sub>Cl<sub>2</sub>) and pure product (**34**) was obtained in 85% yield as a clear, colorless, viscous oil (29.3 mg, 0.0265 mmol). <sup>1</sup>H-NMR (600 MHz, CDCl<sub>3</sub>) δ (ppm) 7.74 (d, *J* = 8.2 Hz, 2 H), 7.41–7.30 (m, 20 H), 7.28 (d, *J* = 7.9 Hz, 2 H), 6.37 (br s, 2 H), 5.71–5.66 (m, 1 H), 4.85 (s, 4 H), 4.74 (s, 2 H), 4.48 (s, 2 H), 3.68–3.58 (m, 6 H), 3.44 (t, *J* = 6.5 Hz, 2 H), 3.23–3.17 (m, 4 H), 3.16–3.11 (m, 2 H), 2.84–2.76 (m, 4 H), 2.62 (t, *J* = 5.3 Hz, 2 H), 2.52–2.44 (m, 4 H), 2.39 (s, 3 H), 1.68–1.56 (m, 8 H), 1.53–1.44 (m, 4 H), 1.40–1.32 (m, 2 H), 1.32–1.22 (m, 4 H); <sup>13</sup>C-NMR (150 MHz, CDCl<sub>3</sub>) δ (ppm) 174.0, 173.8, 172.8, 172.2, 143.1, 138.5, 137.1, 134.3, 134.0, 129.6, 129.2, 129.1, 129.1, 129.1, 128.9, 128.8, 128.7, 128.7, 128.6, 128.3, 127.5, 127.4, 126.9, 76.3, 76.3, 76.3, 72.8, 70.1, 45.5, 45.0, 44.7, 39.3, 38.7, 32.3, 30.7, 30.6, 29.7, 29.3, 29.0, 28.4, 28.1, 28.0, 26.7, 26.4, 26.3, 23.8, 23.6, 23.4, 21.4; HRMS–ESI (*m/z*): [M+H]<sup>+</sup> calcd. for C<sub>61</sub>H<sub>81</sub>N<sub>6</sub>O<sub>11</sub>S: 1105.5679, found 1105.5707.



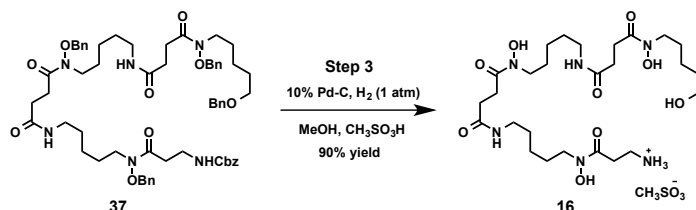
**Siderophore 13.** Benzyl protected siderophore **34** (22.0 mg, 0.02 mmol) was deprotected according to the general procedure for **step 3** to give siderophore **13** in 89% yield as an off-white solid (13.2 mg, 0.018 mmol). Mp 116–118 °C; <sup>1</sup>H-NMR (600 MHz, CD<sub>3</sub>OD) δ (ppm) 7.74 (d, *J* = 8.5 Hz, 2 H), 7.38 (d, *J* = 8.2 Hz, 2 H), 3.59 (t, *J* = 7.0 Hz, 2 H), 3.59 (t, *J* = 6.9 Hz, 2 H), 3.56 (t, *J* = 6.9 Hz, 2 H), 3.54 (t, *J* = 6.6 Hz, 2 H), 3.16 (t, *J* = 6.9 Hz, 2 H), 3.16 (t, *J* = 6.9 Hz, 2 H), 3.09 (t, *J* = 7.2 Hz, 2 H), 2.76 (t, *J* = 7.2 Hz, 4 H), 2.66 (t, *J* = 7.2 Hz, 2 H), 2.45 (t, *J* = 7.3 Hz, 2 H), 2.45 (t, *J* = 7.0 Hz, 2 H), 2.43 (s, 3 H), 1.68–1.58 (m, 6 H), 1.58–1.48 (m, 6 H), 1.40–1.28 (m, 6 H); <sup>13</sup>C-NMR (150 MHz, CD<sub>3</sub>OD) δ (ppm) 175.1, 174.3, 174.3, 172.7, 144.8, 138.9, 130.9, 128.2, 62.9, 49.2, 49.1, 49.0, 40.4, 40.4, 40.2, 33.8, 33.4, 31.8, 31.7, 30.1, 29.1, 29.1, 27.7, 27.5, 27.5, 25.1, 25.0, 24.1, 21.6; HRMS–ESI (*m/z*): [M+H]<sup>+</sup> calcd. for C<sub>33</sub>H<sub>57</sub>N<sub>6</sub>O<sub>11</sub>S: 745.3801, found 745.3787.



**Siderophore-Fe(III) complex 13-Fe.** Siderophore **13** (6.0 mg, 0.008 mmol) was complexed to Fe(III) according to the general procedure for **step 4** to give siderophore-Fe(III) complex **13-Fe** in 97% yield as an orange powder (6.2 mg, 0.008 mmol). Mp 149–154 °C (dec.); HRMS–ESI ( $m/z$ ):  $[M+Na]^+$  calcd. for  $C_{33}H_{53}FeN_6NaO_{11}S$ : 820.2735, found 820.2728; HPLC retention time 4.73 min.

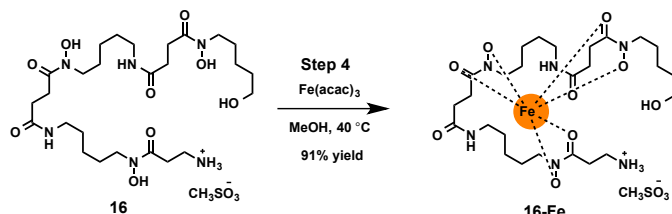


**Benzyl protected siderophore 37.** Hydroxylamine **19** (224.0 mg, 0.25 mmol), *N*-Cbz- $\beta$ -alanine (63.0 mg, 0.28 mmol),  $iPr_2EtN$  (0.05 mL, 0.29 mmol), catalytic DMAP (4.5 mg, 0.037 mmol), and EDC-HCl (125.0 mg, 0.65 mmol) were dissolved in 8 mL of  $CH_2Cl_2$ . After 24 h, TLC (5% MeOH in  $CHCl_3$ ;  $FeCl_3$  stain) showed no remaining starting material (**19**). The mixture was diluted with  $CH_2Cl_2$  (20 mL) and washed with 10% aq. citric acid (2 x 10 mL), brine (10 mL), 10% aq.  $NaHCO_3$  (2 x 10 mL), and brine (10 mL), dried over anhydrous  $MgSO_4$ , filtered, and concentrated under reduced pressure. This gave 301.8 mg of a cloudy oil that was purified by silica gel column chromatography (0.5 x 5 in silica gel; 3% MeOH in  $CHCl_3$ ). Pure product (**37**) was obtained in 90% yield as a clear, colorless oil (242.7 mg, 0.22 mmol).  $^1H$ NMR (600 MHz,  $CDCl_3$ )  $\delta$  (ppm) 7.41–7.21 (m, 25 H), 6.44 (br s, 1 H), 6.41 (br s, 1 H), 5.64 (br s, 1 H), 5.07 (s, 2 H), 4.84 (s, 4 H), 4.75 (br s, 2 H), 4.47 (s, 2 H), 3.70–3.56 (m, 6 H), 3.47–3.41 (m, 4 H), 3.21–3.14 (m, 4 H), 2.83–2.76 (m, 4 H), 2.66–2.60 (m, 2 H), 2.51–2.43 (m, 4 H), 1.66–1.57 (m, 8 H), 1.51–1.43 (m, 4 H), 1.39–1.32 (m, 2 H), 1.31–1.23 (m, 4 H);  $^{13}C$ NMR (150 MHz,  $CDCl_3$ )  $\delta$  (ppm) 173.9, 173.7, 173.2, 172.0, 172.0, 156.2, 138.4, 136.5, 134.2, 134.0, 129.1, 129.0, 128.9, 128.8, 128.7, 128.6, 128.5, 128.5, 128.3, 128.2, 127.8, 127.8, 127.4, 127.3, 76.2, 76.1, 72.7, 69.9, 66.3, 45.4, 44.9, 44.6, 39.1, 36.3, 32.4, 30.5, 30.4, 29.2, 28.9, 28.3, 28.0, 27.8, 26.5, 26.3, 26.2, 23.8, 23.5, 23.3; HRMS–ESI ( $m/z$ ):  $[M+Na]^+$  calcd. for  $C_{62}H_{80}N_6NaO_{11}$ : 1107.5777, found 1107.5760.

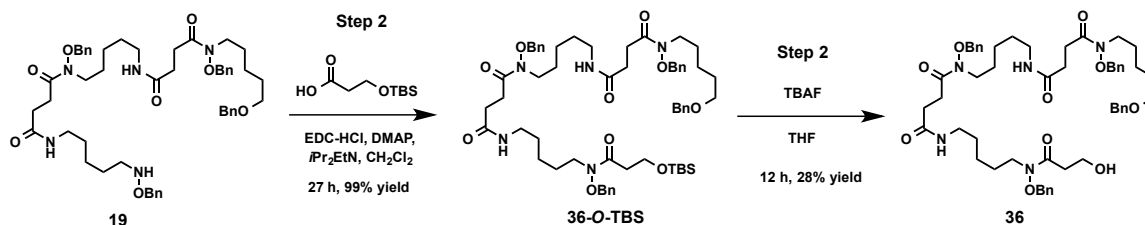


**Siderophore 16.** *N*-Cbz-*O*-Benzyl protected siderophore **37** (37.0 mg, 0.034 mmol) was deprotected according to the general procedure for **step 3** in the presence of methanesulfonic acid (4.0 mg, 0.042 mmol) to give the siderophore methanesulfonate salt of **16** in 90% yield as a light, pink solid (21.6 mg, 0.031 mmol). Mp 139–141 °C (dec.);  $^1H$ -NMR (600 MHz,  $CD_3OD$ )  $\delta$

(ppm) 3.62 (t,  $J = 7.0$  Hz, 2 H), 3.59 (t,  $J = 7.0$  Hz, 4 H), 3.54 (t,  $J = 6.5$  Hz, 2 H), 3.21–3.14 (m, 6 H), 2.88 (t,  $J = 6.3$  Hz, 2 H), 2.76 (t,  $J = 6.2$  Hz, 4 H), 2.70 (s, 3 H), 2.45 (t,  $J = 7.0$  Hz, 4 H), 1.69–1.60 (m, 6 H), 1.59–1.49 (m, 6 H), 1.40–1.30 (m, 6 H);  $^{13}\text{C}$ -NMR (150 MHz,  $\text{CD}_3\text{OD}$ )  $\delta$  (ppm) 175.1, 174.6, 174.5, 172.2, 62.9, 52.3, 49.7, 49.0, 40.4, 40.4, 39.6, 36.8, 33.4, 31.6, 31.6, 30.5, 30.2, 30.1, 29.2, 29.1, 29.0, 27.7, 27.5, 27.4, 25.0, 24.1; HRMS–ESI ( $m/z$ ):  $[\text{M}+\text{H}]^+$  calcd. for  $\text{C}_{26}\text{H}_{51}\text{N}_6\text{O}_9$ : 591.3712, found 591.3695.



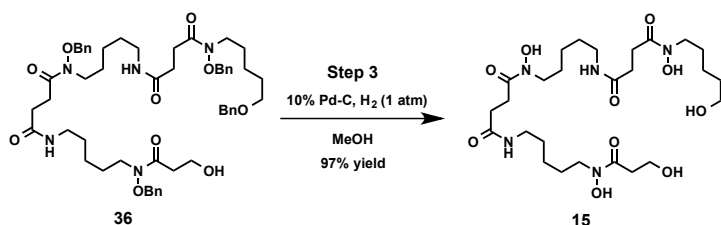
**Siderophore-Fe(III) complex 16-Fe.** Siderophore **16** (11.0 mg, 0.016 mmol) was complexed to Fe(III) according to the general procedure for **step 4** to give siderophore-Fe(III) complex **16-Fe** in 91% yield as an orange powder (10.5 mg, 0.014 mmol). Mp 153–158 °C (dec.); HRMS–ESI ( $m/z$ ):  $[\text{M}+\text{H}]^+$  calcd. for  $\text{C}_{26}\text{H}_{48}\text{FeN}_6\text{O}_9$ : 644.2827, found 642.2891; HPLC retention time 2.26 min.



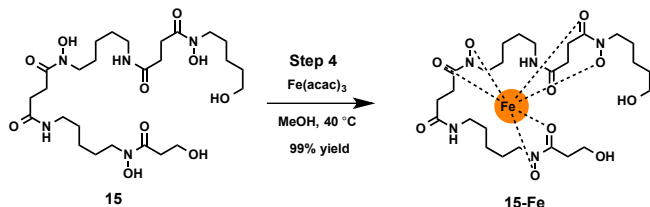
**Benzyl protected siderophore 36.** Hydroxylamine **19** (275.0 mg, 0.31 mmol), 4-((*tert*-butyldimethylsilyl)oxy)butanoic acid (84.0 mg, 0.41 mmol),  $i\text{Pr}_2\text{EtN}$  (0.13 mL, 0.75 mmol), catalytic DMAP (11.0 mg, 0.09 mmol), and EDC-HCl (150.0 mg, 0.78 mmol) were dissolved in 8 mL of  $\text{CH}_2\text{Cl}_2$ . After 27 h, TLC (9% MeOH in  $\text{CH}_2\text{Cl}_2$ ;  $\text{FeCl}_3$  stain) showed no remaining starting material (**19**). The mixture was diluted with  $\text{CH}_2\text{Cl}_2$  (30 mL) and washed with 10% aq. citric acid (30 mL) and brine (50 mL), dried over anhydrous  $\text{MgSO}_4$ , filtered, and concentrated under reduced pressure to give 417.8 mg of a waxy solid. The crude product was purified by silica gel column chromatography (1 x 5 in silica gel; 3%–5% MeOH in  $\text{CH}_2\text{Cl}_2$ ) to give pure **36-O-TBS** in 99% yield as a white, waxy solid (332.3 mg, 0.31 mmol). Mp 71–73 °C;  $^1\text{H}$ -NMR (600 MHz,  $\text{CDCl}_3$ )  $\delta$  (ppm) 7.41–7.30 (m, 20 H), 6.32 (br s, 1 H), 6.28 (br s, 1 H), 4.85 (s, 2 H), 4.85 (s, 2 H), 4.81 (s, 2 H), 4.48 (s, 2 H), 3.68–3.59 (m, 8 H), 3.44 (t,  $J = 6.5$  Hz, 2 H), 3.22–3.17 (m, 4 H), 2.84–2.77 (m, 4 H), 2.55–2.44 (m, 6 H), 1.86–1.80 (m, 2 H), 1.67–1.59 (m, 8 H), 1.50 (dt,  $J = 14.9$ , 7.4 Hz, 4 H), 1.36 (dt,  $J = 15.5$ , 7.7 Hz, 2 H), 1.33–1.25 (m, 4 H), 0.89 (s, 9 H), 0.04 (s, 6 H);  $^{13}\text{C}$ -NMR (150 MHz,  $\text{CDCl}_3$ )  $\delta$  (ppm) 174.7, 174.0, 173.8, 172.1, 172.0, 138.5, 134.5, 134.3, 129.1, 129.0, 128.9, 128.8, 128.8, 128.7, 128.6, 128.3, 127.5, 127.4, 76.3, 72.8, 70.1, 62.4, 45.5, 45.2, 44.7, 39.4, 39.3, 39.3, 30.7, 30.6, 29.3, 29.1, 28.8, 28.4, 28.1, 28.0, 27.6, 26.7, 26.5, 26.4, 25.9, 24.0, 23.6, 23.4, 18.3, -5.3; HRMS–ESI ( $m/z$ ):  $[\text{M}+\text{H}]^+$  calcd. for  $\text{C}_{61}\text{H}_{90}\text{N}_5\text{O}_{10}\text{Si}$ : 1080.6451, found 1080.6466. Purified **36-O-TBS** (224.0 mg, 0.21 mmol) was dissolved in 7 mL of anhydrous THF. TBAF (1 M in THF, 0.54 mL, 0.54 mmol) was added and the mixture was stirred at rt. After 12 h, the THF was evaporated under reduced pressure and the crude material was purified by silica gel column chromatography (1 x 6 in silica gel; 3%–7%



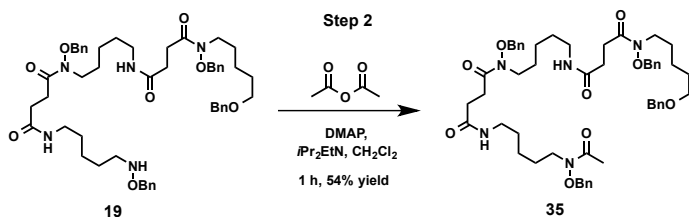
MeOH in CH<sub>2</sub>Cl<sub>2</sub>) to give the desired free alcohol product **36** in 28.4% yield (47.3% yield based on recovered **36-O-TBS**) as a clear oil (56.8 mg, 0.06 mmol). <sup>1</sup>H-NMR (600 MHz, CDCl<sub>3</sub>) δ (ppm) 7.43–7.29 (m, 20 H), 6.42 (br s, 1 H), 6.18 (br s, 1 H), 4.85–4.83 (m, 4 H), 4.83–4.80 (m, 2 H), 4.47 (s, 2 H), 3.71–3.56 (m, 8 H), 3.52–3.41 (m, 4 H), 3.24–3.15 (m, 4 H), 2.84–2.75 (m, 2 H), 2.70–2.66 (m, 2 H), 2.59–2.53 (m, 2 H), 2.50–2.43 (m, 2 H), 1.91–1.83 (m, 2 H), 1.69–1.46 (m, 12 H), 1.40–1.22 (m, 6 H); <sup>13</sup>C-NMR (150 MHz, CDCl<sub>3</sub>) δ (ppm) 177.3, 175.2, 173.8, 172.2, 172.1, 138.5, 134.3, 129.2, 129.1, 129.0, 128.9, 128.8, 128.7, 128.7, 128.6, 128.3, 127.6, 127.5, 76.3, 76.3, 76.2, 72.8, 70.1, 62.4, 45.5, 45.2, 44.7, 39.3, 38.5, 30.8, 30.7, 30.5, 29.5, 29.3, 28.7, 28.1, 28.0, 27.3, 27.2, 26.7, 26.4, 26.3, 23.9, 23.7, 23.6, 23.4; HRMS–ESI (m/z): [M+H]<sup>+</sup> calcd. for C<sub>55</sub>H<sub>76</sub>N<sub>5</sub>O<sub>10</sub>: 966.5587, found 966.5578.



**Siderophore 15.** Benzyl protected siderophore **36** (38.7 mg, 0.040 mmol) was deprotected according to the general procedure for **step 3** to give siderophore **15** in 97% yield as a tan, hygroscopic solid (23.4 mg, 0.039 mmol). <sup>1</sup>H-NMR (600 MHz, CD<sub>3</sub>OD) δ (ppm) 3.64–3.52 (m, 8 H), 3.47 (t, *J* = 7.2 Hz, 2 H), 3.17 (t, *J* = 6.7 Hz, 4 H), 2.79–2.73 (m, 2 H), 2.71–2.66 (m, 2 H), 2.55 (t, *J* = 7.5 Hz, 2 H), 2.45 (t, *J* = 7.0 Hz, 4 H), 1.81 (quintet, *J* = 7.0 Hz, 2 H), 1.67–1.60 (m, 6 H), 1.60–1.49 (m, 6 H), 1.40–1.26 (m, 6 H); <sup>13</sup>C-NMR (150 MHz, CD<sub>3</sub>OD) δ (ppm) 180.2, 175.7, 175.1, 174.5, 62.9, 62.6, 48.9, 40.4, 39.5, 33.4, 31.7, 30.1, 30.0, 29.2, 29.1, 28.9, 28.4, 27.6, 27.5, 27.3, 25.1, 25.0, 24.1; HRMS–ESI (m/z): [M+H]<sup>+</sup> calcd. for C<sub>27</sub>H<sub>52</sub>N<sub>5</sub>O<sub>10</sub>: 606.3709, found 606.3717.



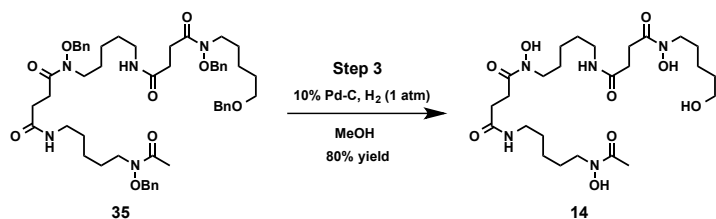
**Siderophore-Fe(III) complex 15-Fe.** Siderophore **15** (8.5 mg, 0.014 mmol) was complexed to Fe(III) according to the general procedure for **step 4** to give siderophore-Fe(III) complex **15-Fe** in 99% yield as an orange powder (9.1 mg, 0.014 mmol). Mp 192–195 °C; HRMS–ESI (m/z): [M+H]<sup>+</sup> calcd. for C<sub>27</sub>H<sub>49</sub>FeN<sub>5</sub>O<sub>10</sub>: 659.2823, found 659.2812; HPLC retention time 2.53 min.



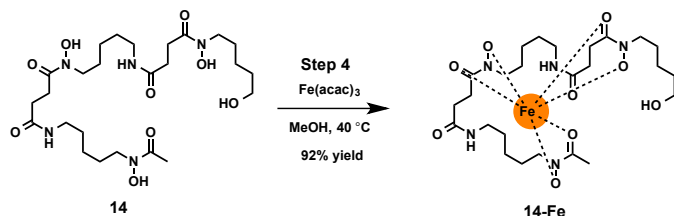
**Benzyl protected siderophore 35.** Hydroxylamine **19** (76.9 mg, 0.087 mmol), acetic anhydride (18 mg, 0.176 mmol), *i*Pr<sub>2</sub>EtN (22.0 mg, 0.17 mmol), and catalytic DMAP (1.1 mg, 0.009 mmol) were dissolved in 6.0 mL of CH<sub>2</sub>Cl<sub>2</sub>. After 1 h, TLC (6% MeOH in CH<sub>2</sub>Cl<sub>2</sub>; FeCl<sub>3</sub> stain) showed



no remaining starting material (**19**). The mixture was diluted with CH<sub>2</sub>Cl<sub>2</sub> (15 mL) and washed with 10% aq. citric acid (10 mL), brine (10 mL), 10% aq. NaHCO<sub>3</sub> (10 mL), and brine (10 mL), dried over anhydrous MgSO<sub>4</sub>, filtered, and concentrated under reduced pressure. This gave 68.5 mg of a clear, colorless oil that was purified via silica gel column chromatography (0.75 x 4 in silica gel; 3%–6% MeOH in CH<sub>2</sub>Cl<sub>2</sub>). Pure product (**35**) was obtained in 54% yield as a clear, colorless oil (43 mg, 0.05 mmol). All characterization data matched that previously reported in the literature.<sup>9</sup> <sup>1</sup>H-NMR (600 MHz, CDCl<sub>3</sub>)  $\delta$  (ppm) 7.41–7.26 (m, 20 H), 6.34 (br s, 1 H), 6.32 (br s, 1 H), 4.85 (s, 2 H), 4.84 (s, 2 H), 4.80 (s, 2 H), 4.48 (s, 2 H), 3.67–3.58 (m, 6 H), 3.44 (t,  $J$  = 6.46 Hz, 2 H), 3.22–3.18 (m, 4 H), 2.83–2.77 (m, 4 H), 2.50–2.47 (m, 4 H), 2.09 (s, 3 H), 1.67–1.59 (m, 8 H), 1.52–1.48 (m, 4 H), 1.39–1.35 (m, 2 H), 1.33–1.26 (m, 4 H); <sup>13</sup>C-NMR (150 MHz, CDCl<sub>3</sub>)  $\delta$  (ppm) 174.0, 173.8, 172.1, 172.1, 138.5, 134.3, 129.1, 129.1, 128.9, 128.9, 128.8, 128.7, 128.7, 128.6, 128.3, 127.5, 127.4, 76.3, 76.3, 76.2, 72.8, 70.1, 45.5, 44.9, 44.7, 39.3, 39.3, 30.7, 30.6, 29.3, 29.0, 28.4, 28.1, 28.0, 26.7, 26.5, 26.4, 23.9, 23.6, 23.4, 20.5; HRMS–ESI ( $m/z$ ):  $[M+H]^+$  calcd. for C<sub>53</sub>H<sub>72</sub>N<sub>5</sub>O<sub>9</sub>: 922.5325, found 922.5362.

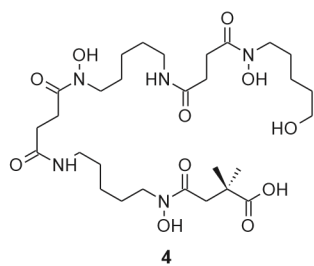


**Siderophore 14.** Benzyl protected siderophore **35** (34.8 mg, 0.038 mmol) was deprotected according to the general procedure for **step 3** to give siderophore **14** in 80% yield as a white solid (16.9 mg, 0.03 mmol). All characterization data matched that previously reported in the literature.<sup>9</sup> Mp 123–125 °C; <sup>1</sup>H-NMR (600 MHz, CD<sub>3</sub>OD)  $\delta$  (ppm) 3.60 (t,  $J$  = 6.9 Hz, 6 H), 3.19–3.13 (m, 6 H), 2.77 (t,  $J$  = 7.2 Hz, 4 H), 2.48–2.43 (m, 4 H), 2.09 (s, 3 H), 1.67–1.60 (m, 4 H), 1.58–1.48 (m, 8 H), 1.41–1.30 (m, 6 H); <sup>13</sup>C-NMR (150 MHz, CD<sub>3</sub>OD)  $\delta$  (ppm) 175.1, 174.7, 174.3, 174.2, 173.2, 67.1, 62.9, 49.7, 49.2, 49.1, 48.9, 48.7, 40.5, 40.5, 40.4, 33.4, 32.5, 32.5, 32.5, 31.7, 31.7, 30.3, 30.2, 30.1, 30.1, 29.1, 27.7, 27.5, 27.5, 25.4, 25.3, 25.1, 25.0, 24.4, 24.1, 22.7, 20.3, 15.6; HRMS–ESI ( $m/z$ ):  $[M+Na]^+$  calcd. for C<sub>25</sub>H<sub>47</sub>N<sub>5</sub>NaO<sub>9</sub>: 584.3266, found 584.3291.

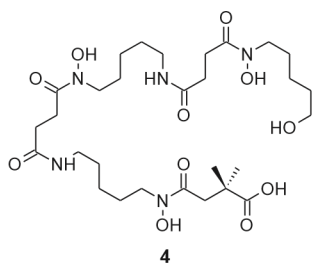
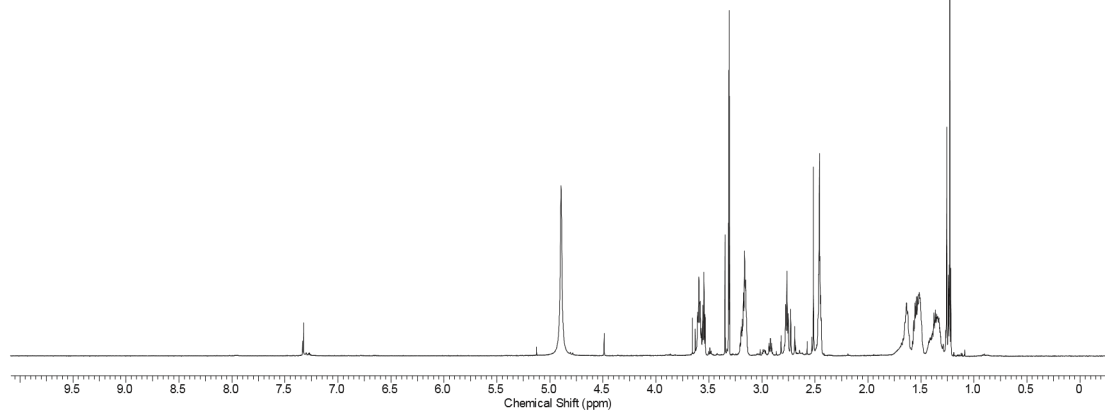


**Siderophore-Fe(III) complex 14-Fe.** Siderophore **14** (10.0 mg, 0.018 mmol) was complexed to Fe(III) according to the general procedure for **step 4** to give siderophore-Fe(III) complex **14-Fe** in 92% yield as an orange powder (10.0 mg, 0.016 mmol). Mp 164–167 °C; HRMS–ESI ( $m/z$ ):  $[M+Na]^+$  calcd. for C<sub>25</sub>H<sub>44</sub>FeN<sub>5</sub>NaO<sub>9</sub>: 637.2381, found 637.2403; HPLC retention time 2.75 min.

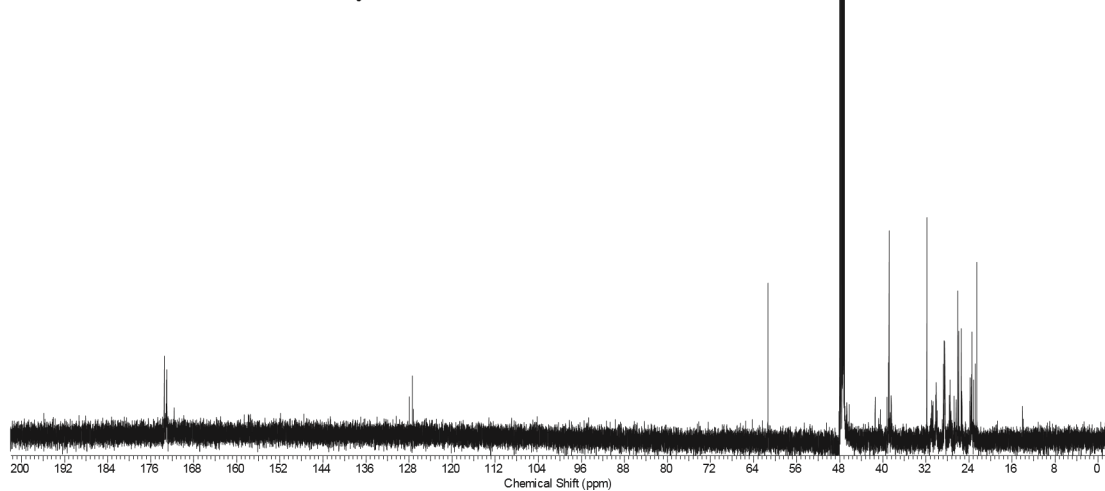
## V. $^1\text{H}$ -NMR and $^{13}\text{C}$ -NMR Spectra of New Compounds

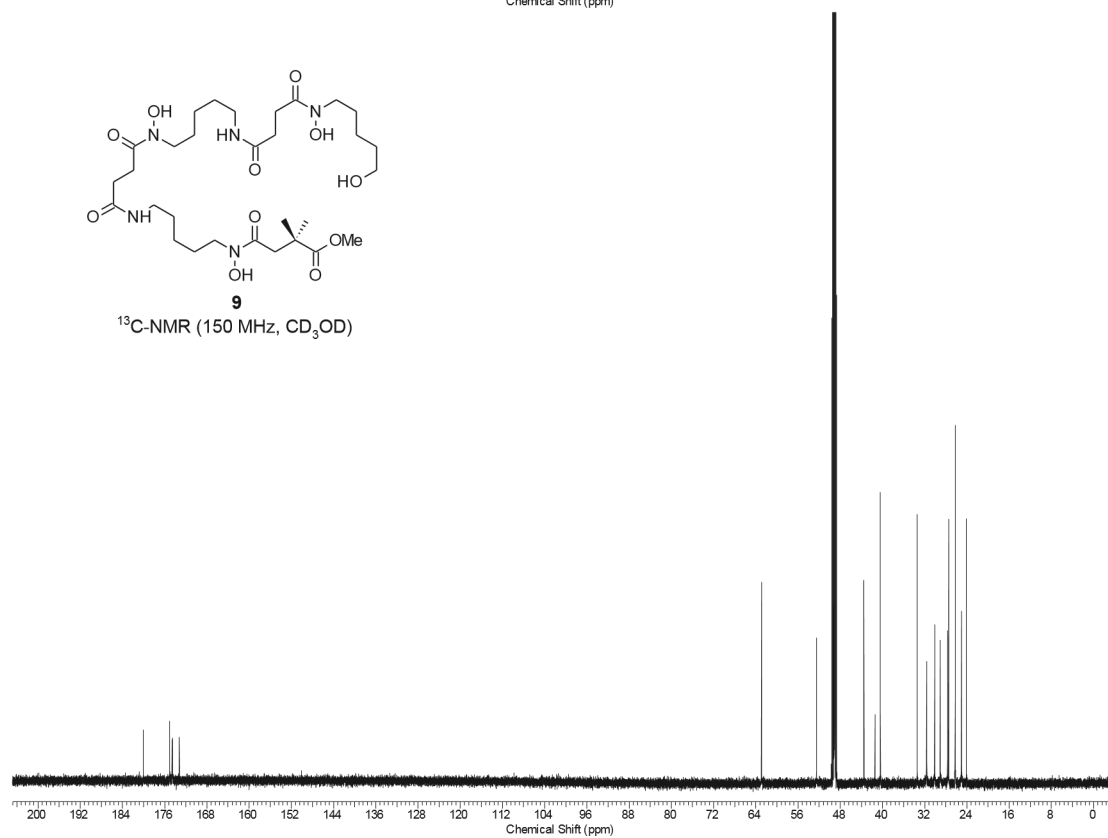
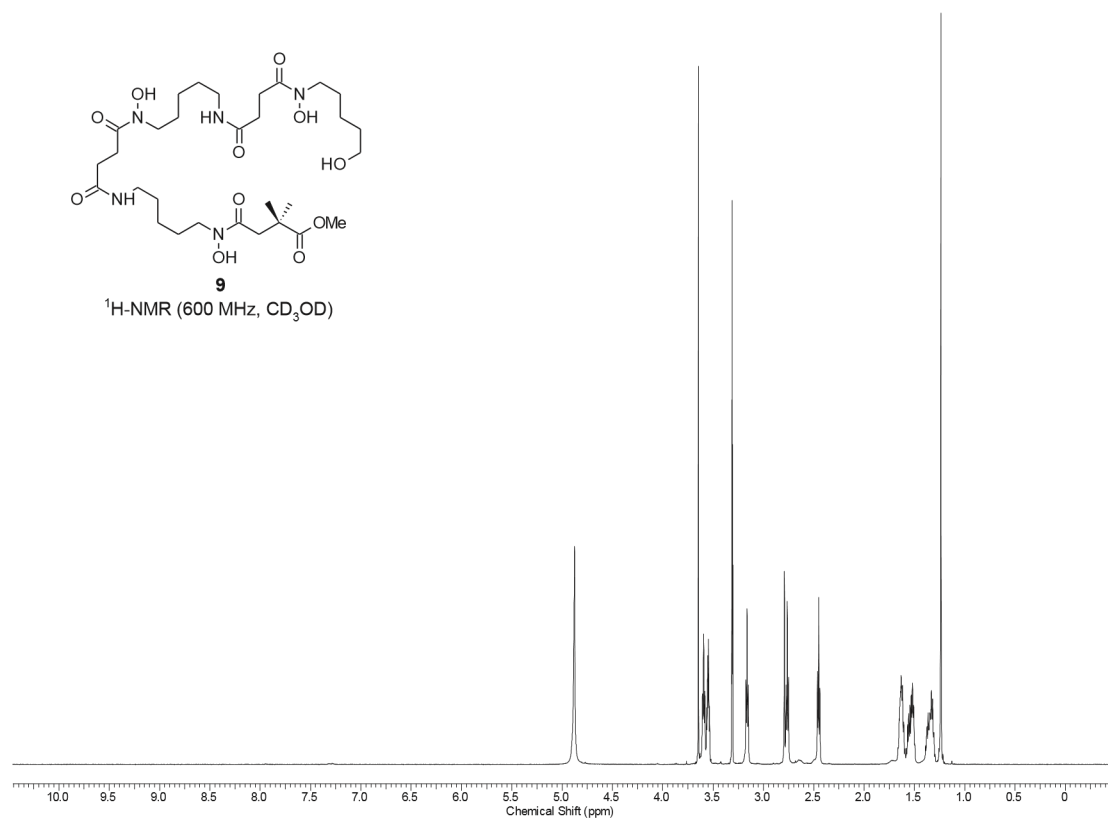


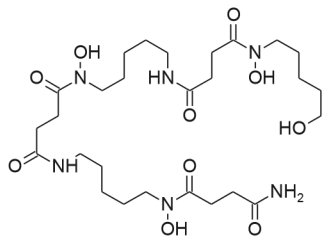
$^1\text{H}$ -NMR (600 MHz,  $\text{CD}_3\text{OD}$ )



$^{13}\text{C}$ -NMR (150 MHz,  $\text{CD}_3\text{OD}$ )

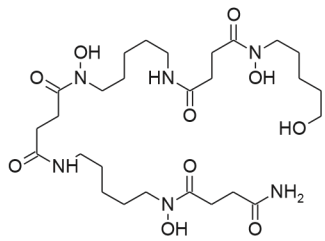
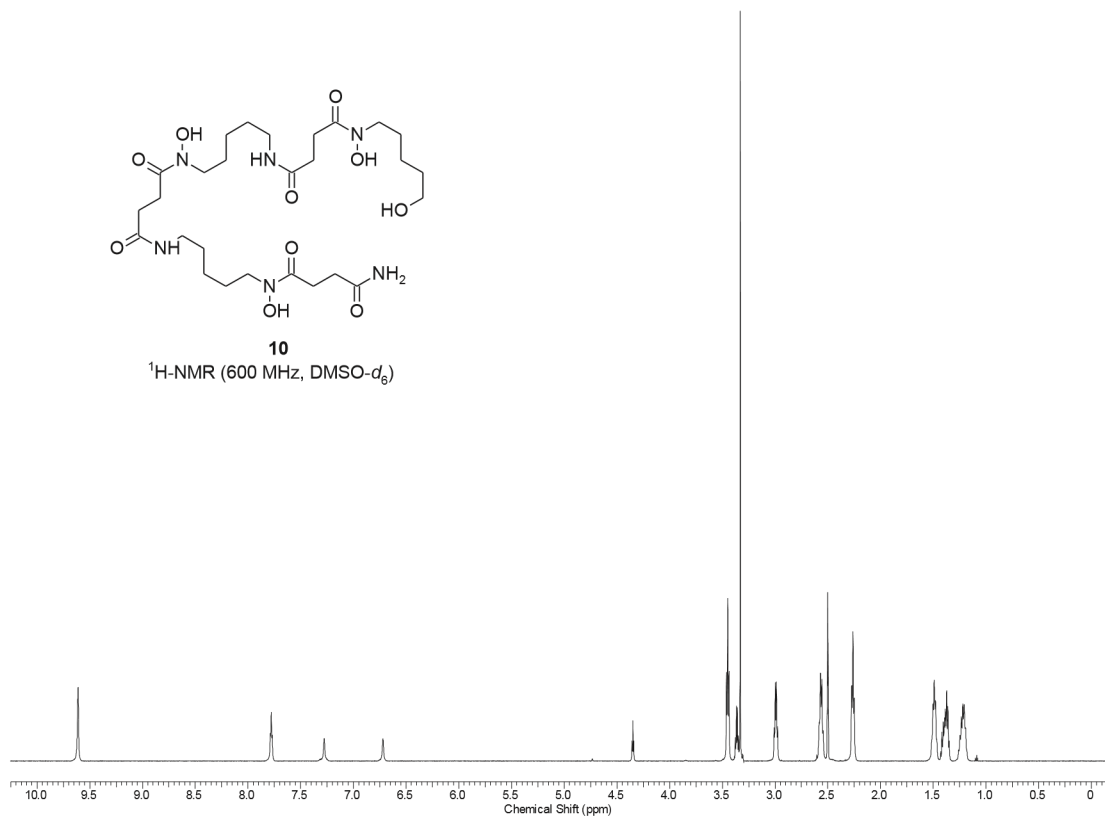






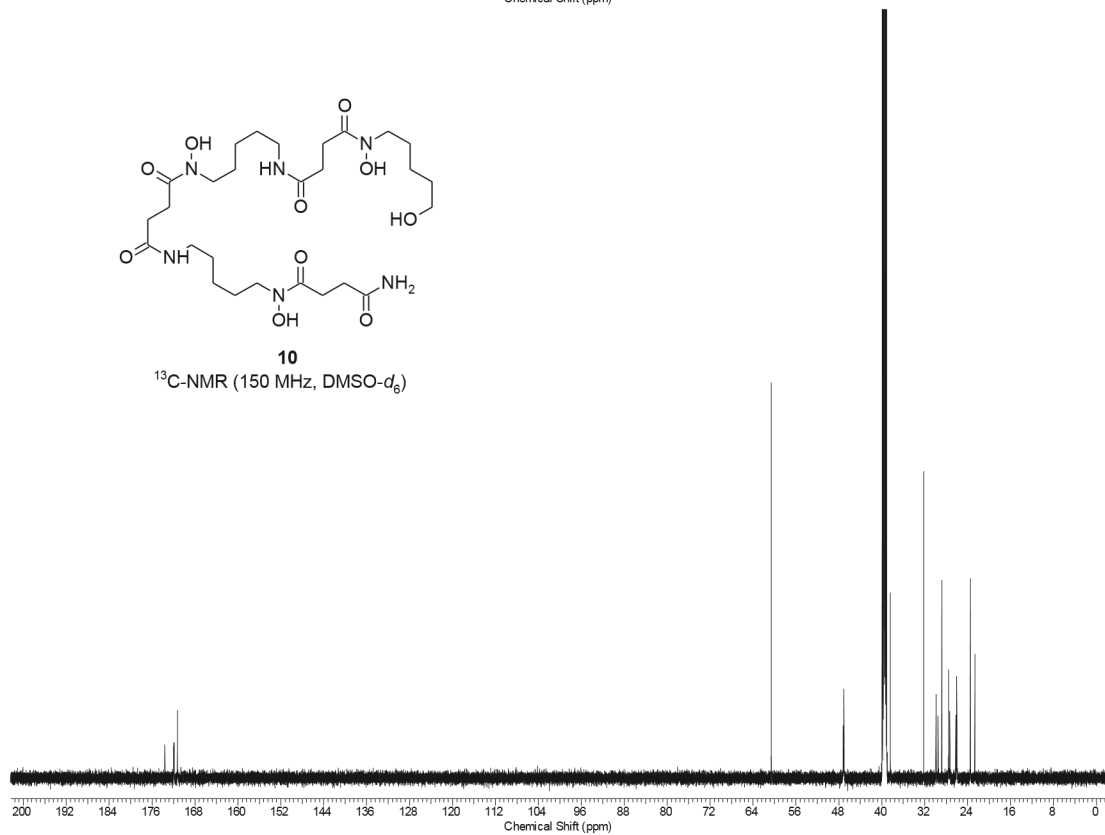
**10**

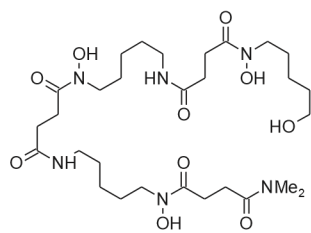
<sup>1</sup>H-NMR (600 MHz, DMSO-*d*<sub>6</sub>)



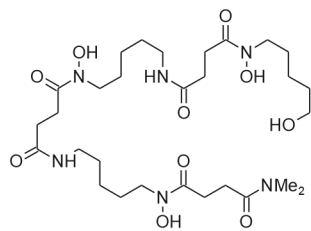
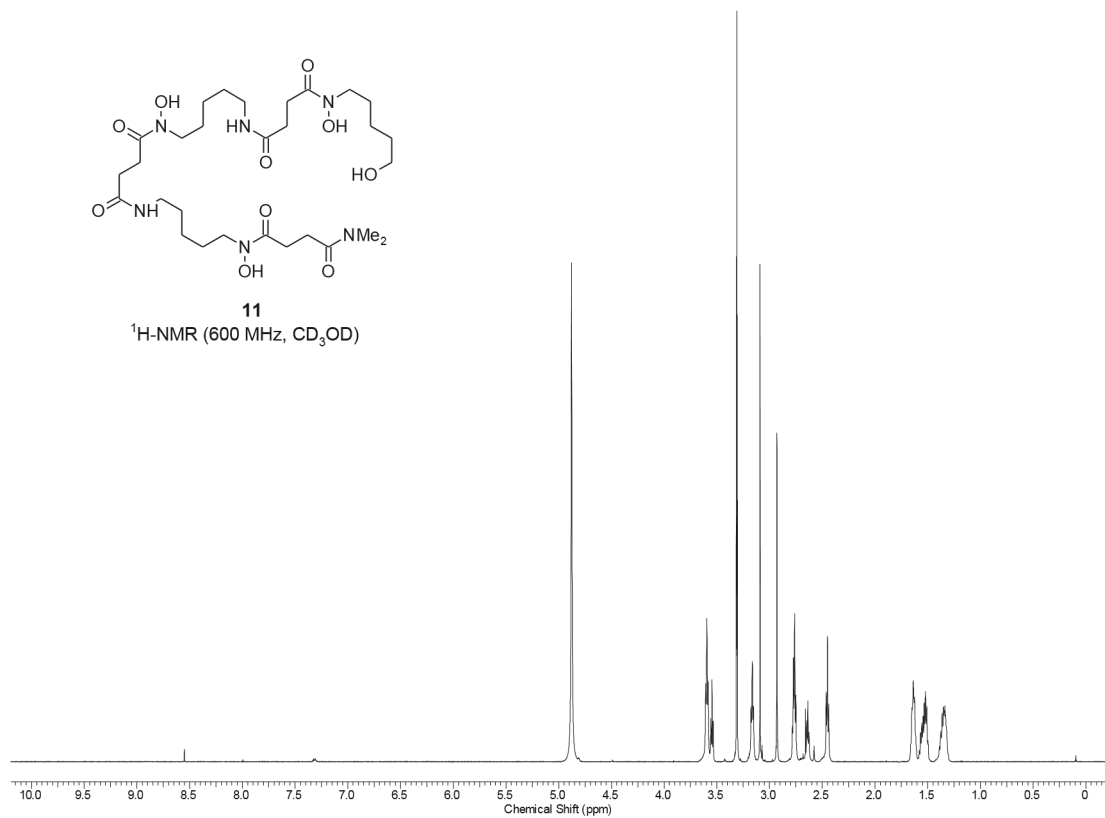
**10**

<sup>13</sup>C-NMR (150 MHz, DMSO-*d*<sub>6</sub>)

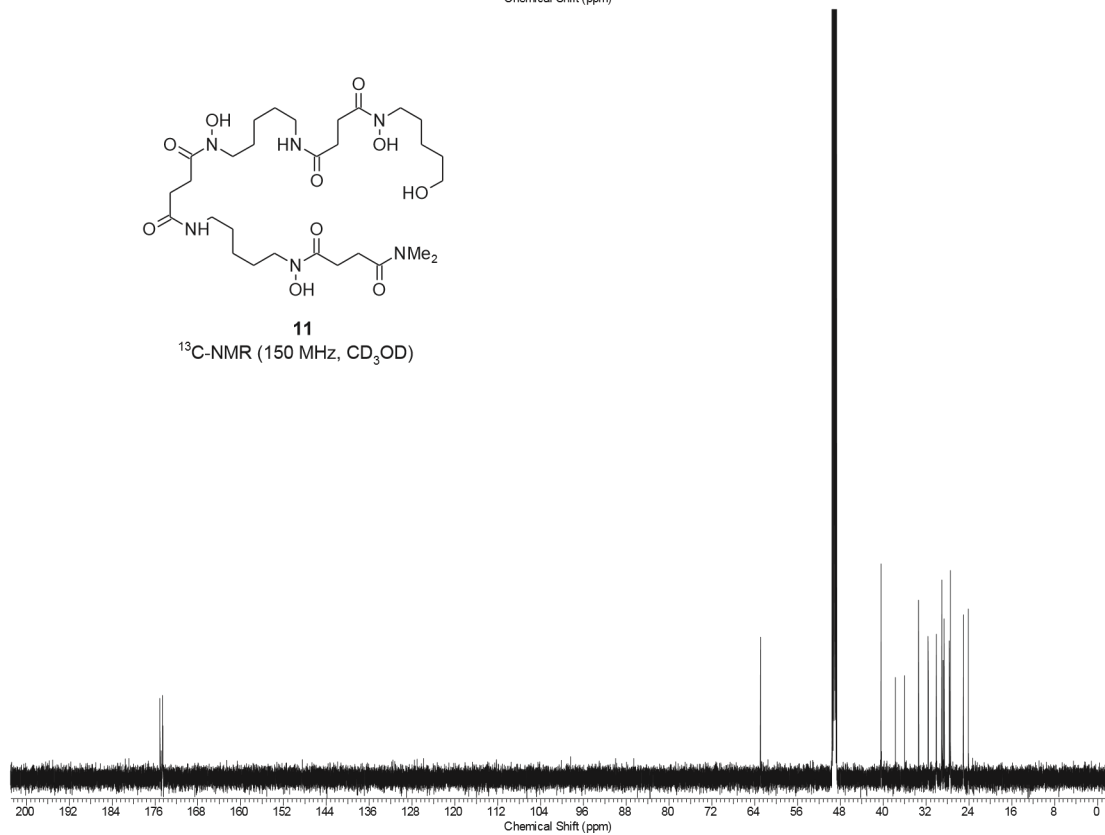


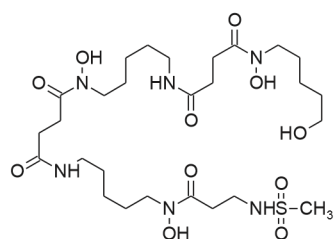
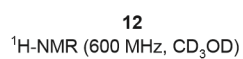


**11**  
<sup>1</sup>H-NMR (600 MHz, CD<sub>3</sub>OD)



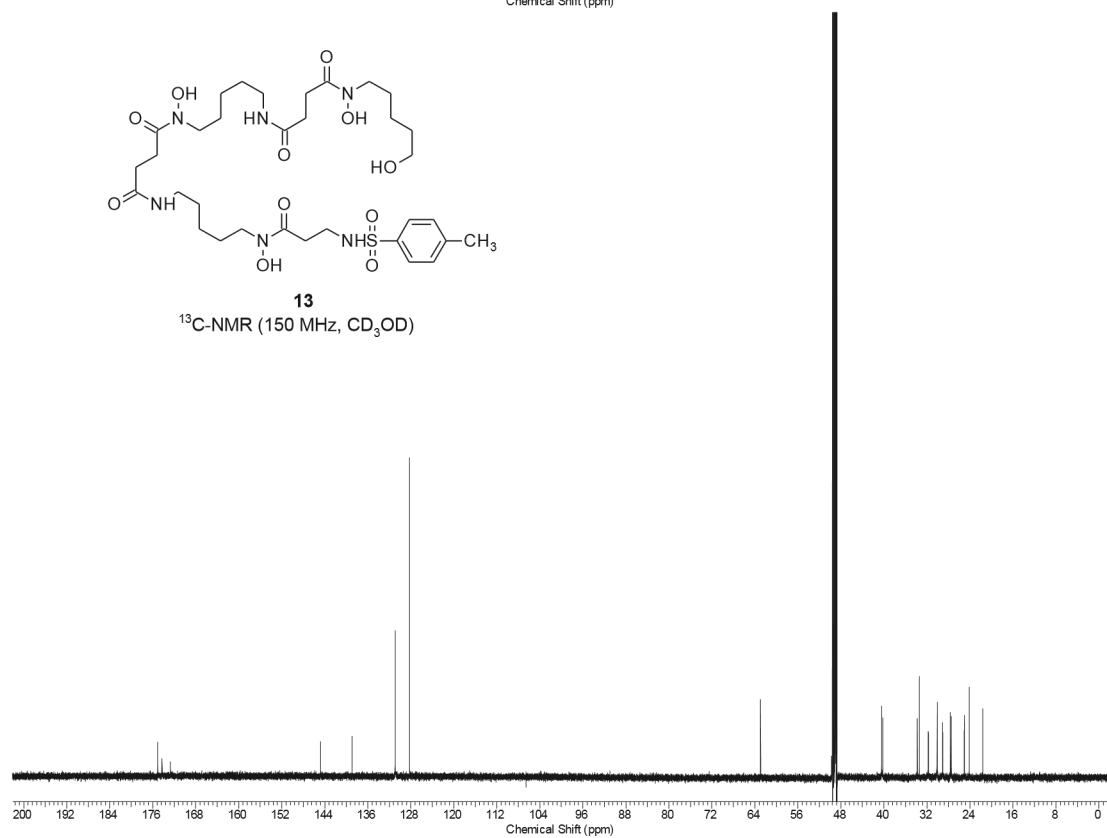
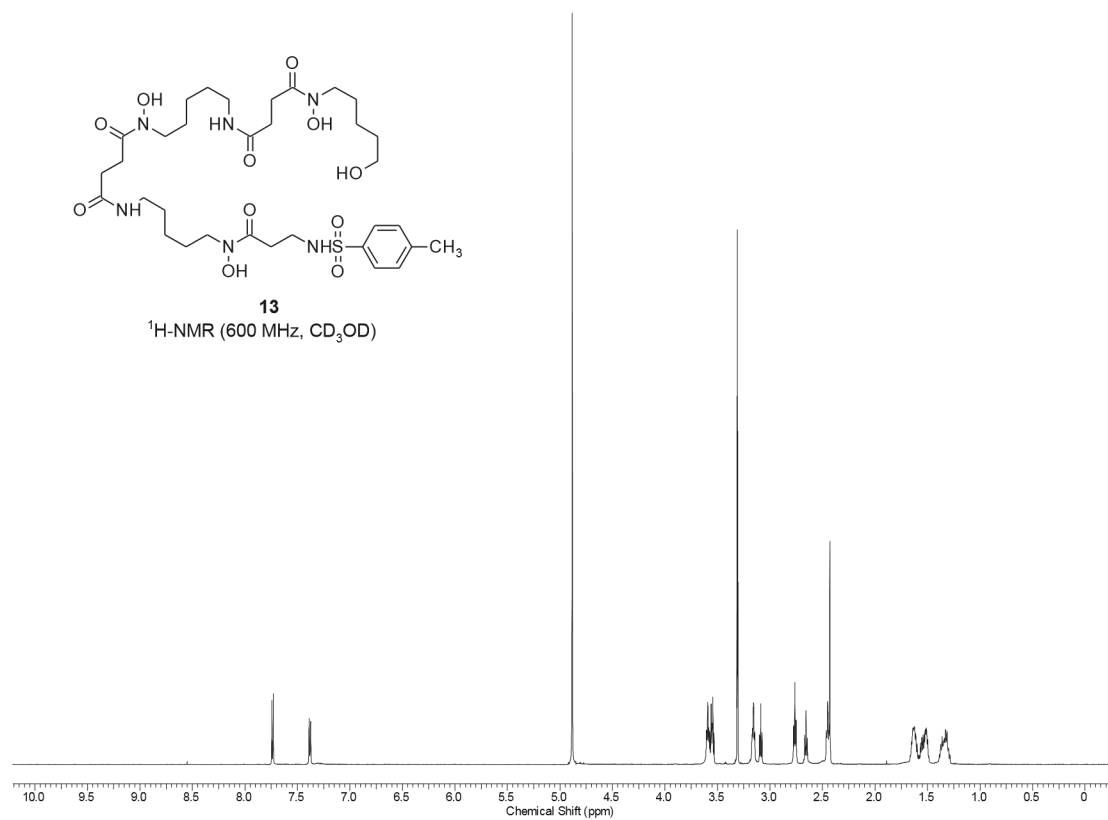
**11**  
<sup>13</sup>C-NMR (150 MHz, CD<sub>3</sub>OD)

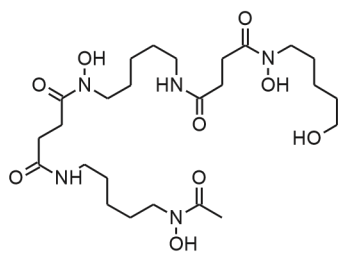




**12**  
<sup>1</sup>H-NMR (600 MHz, CD<sub>3</sub>OD)  
 Compound precipitated in NMR  
 tube during FID-acquisition.



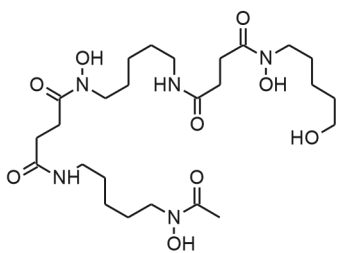
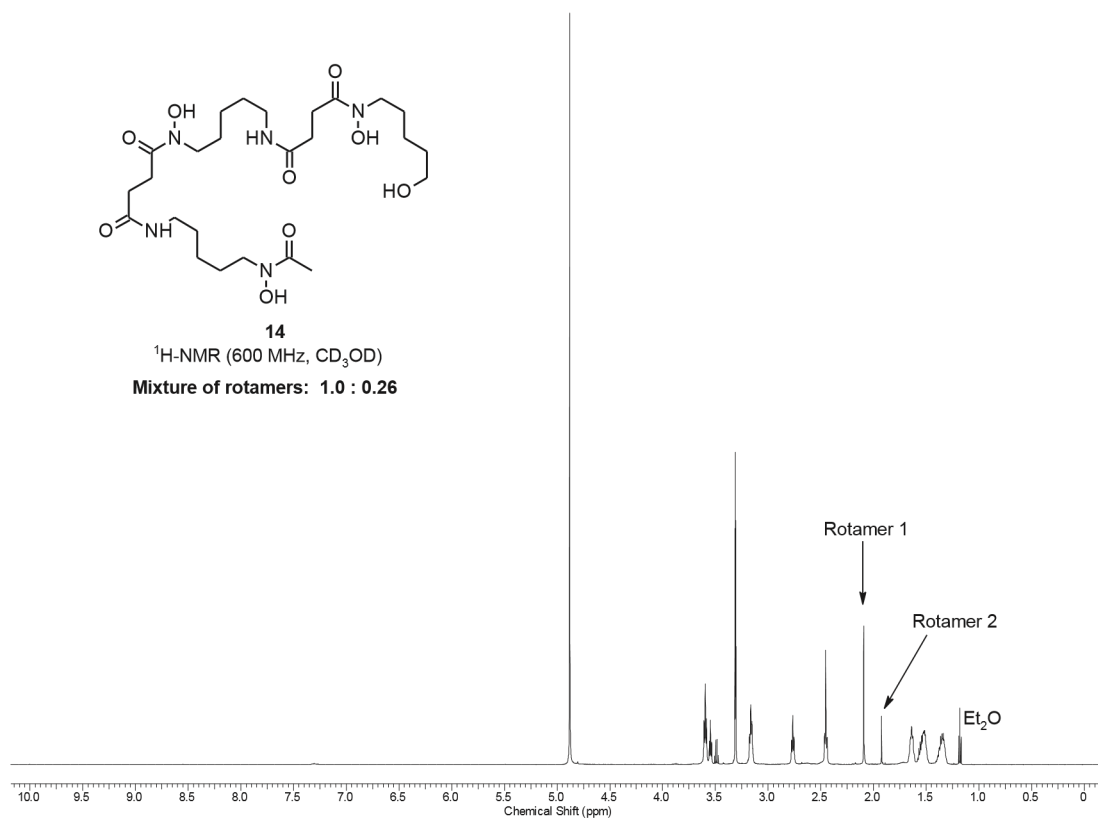




**14**

<sup>1</sup>H-NMR (600 MHz, CD<sub>3</sub>OD)

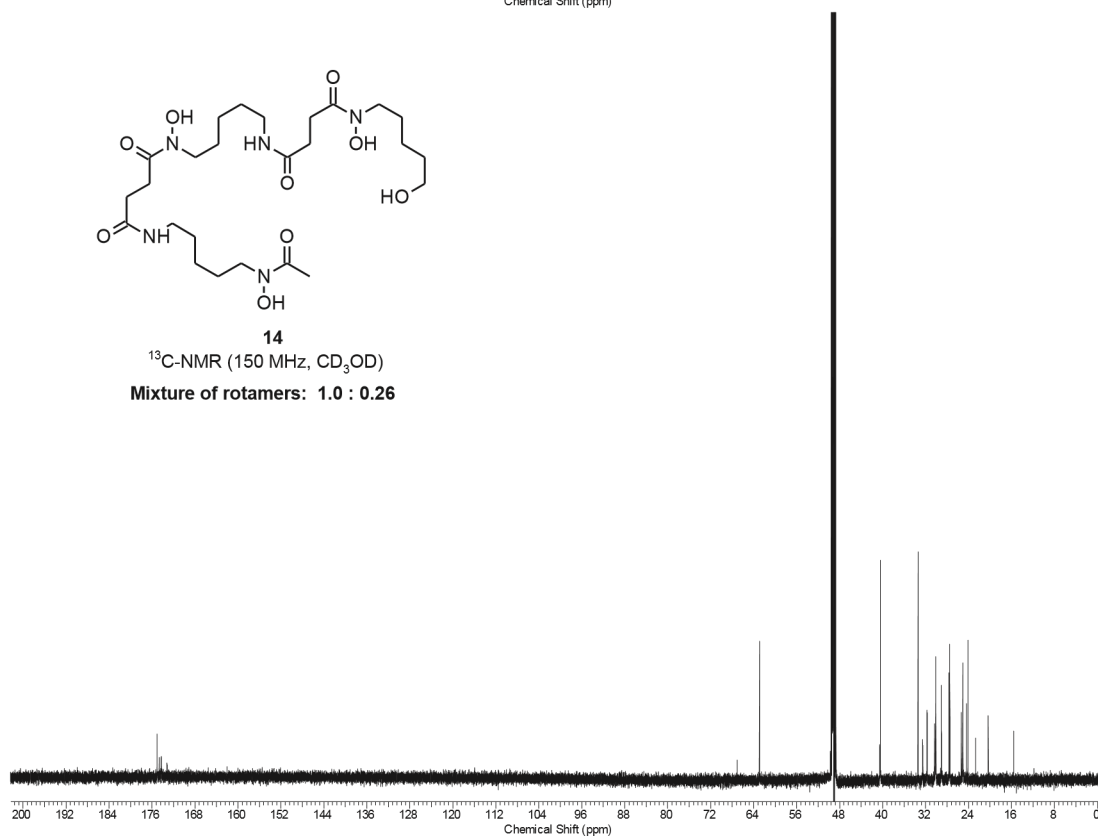
Mixture of rotamers: 1.0 : 0.26



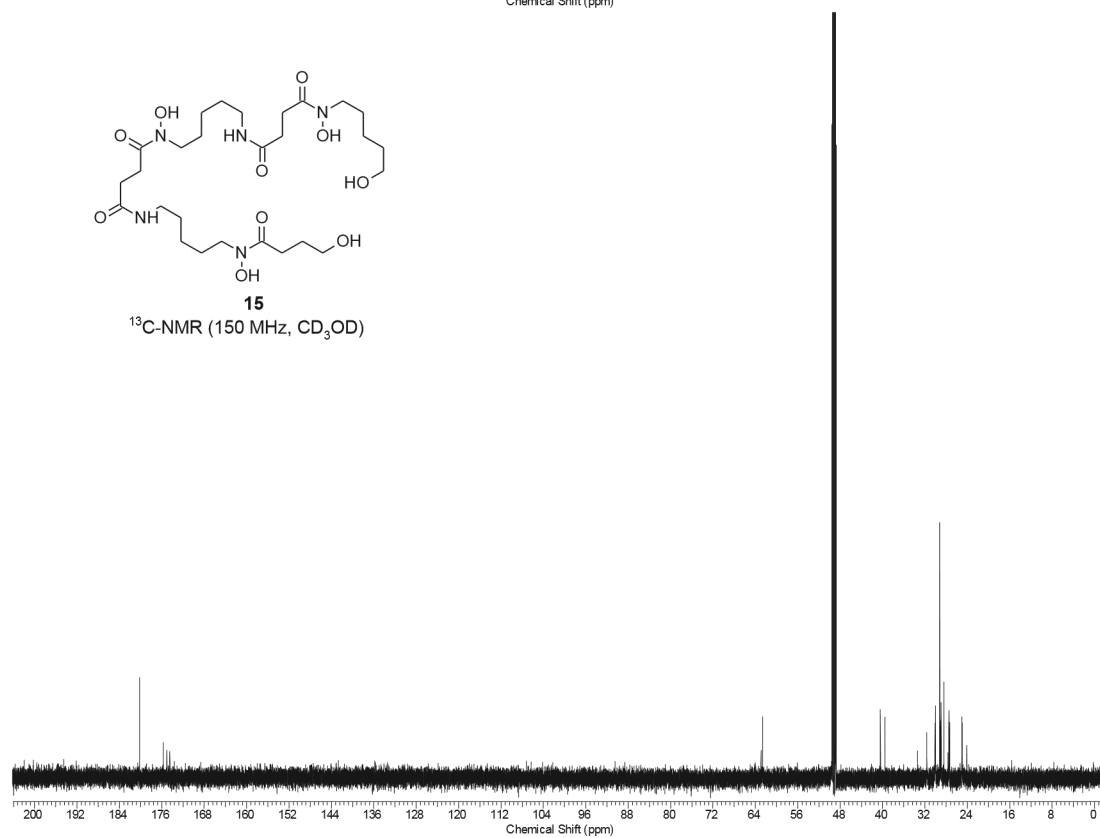
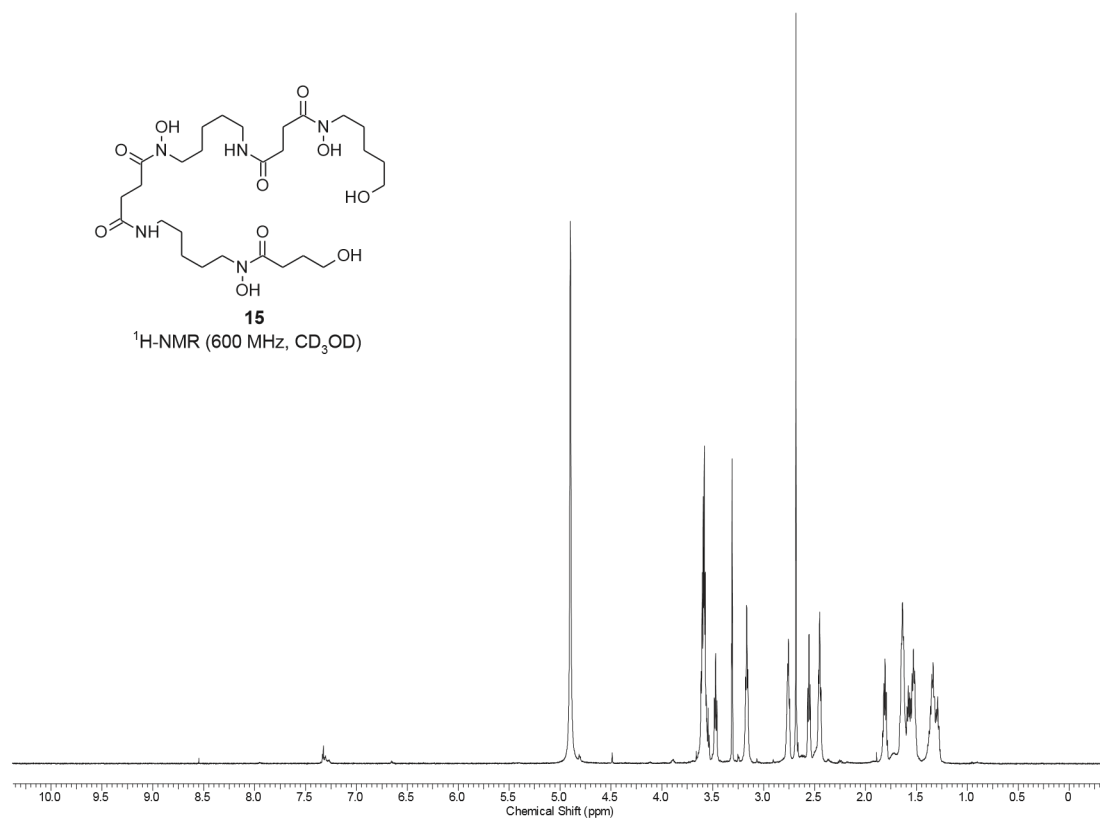
**14**

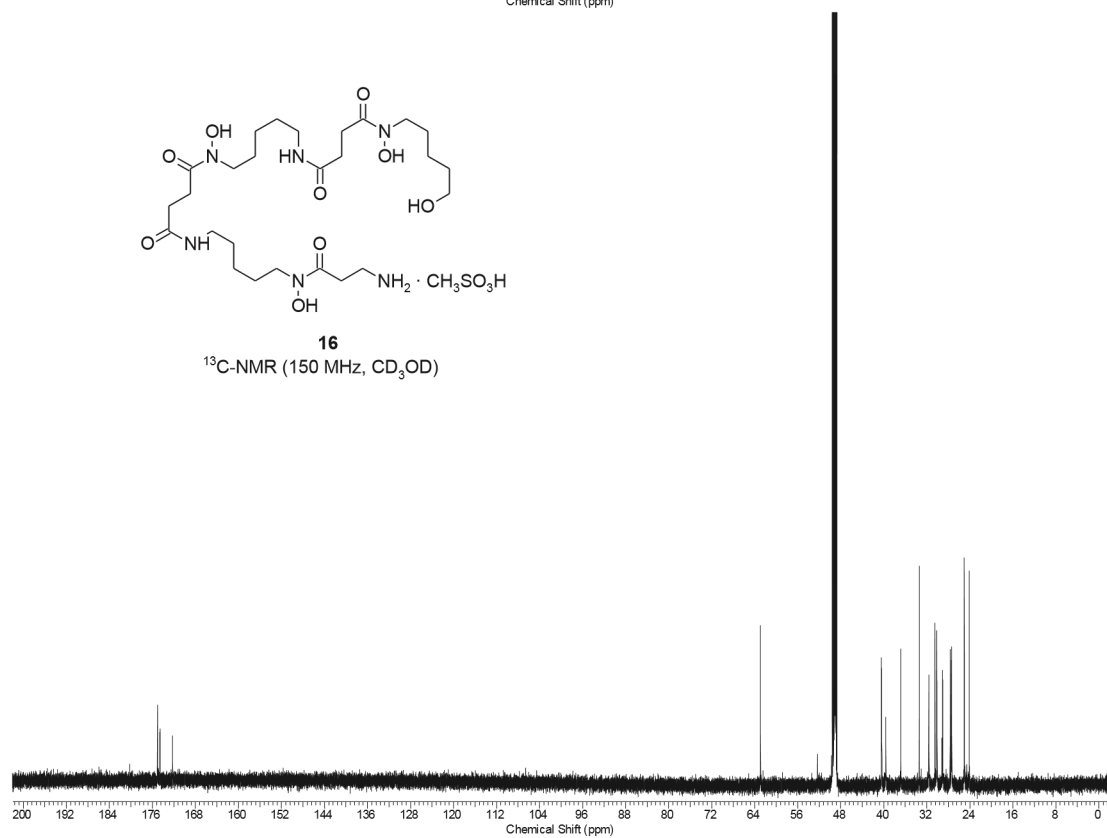
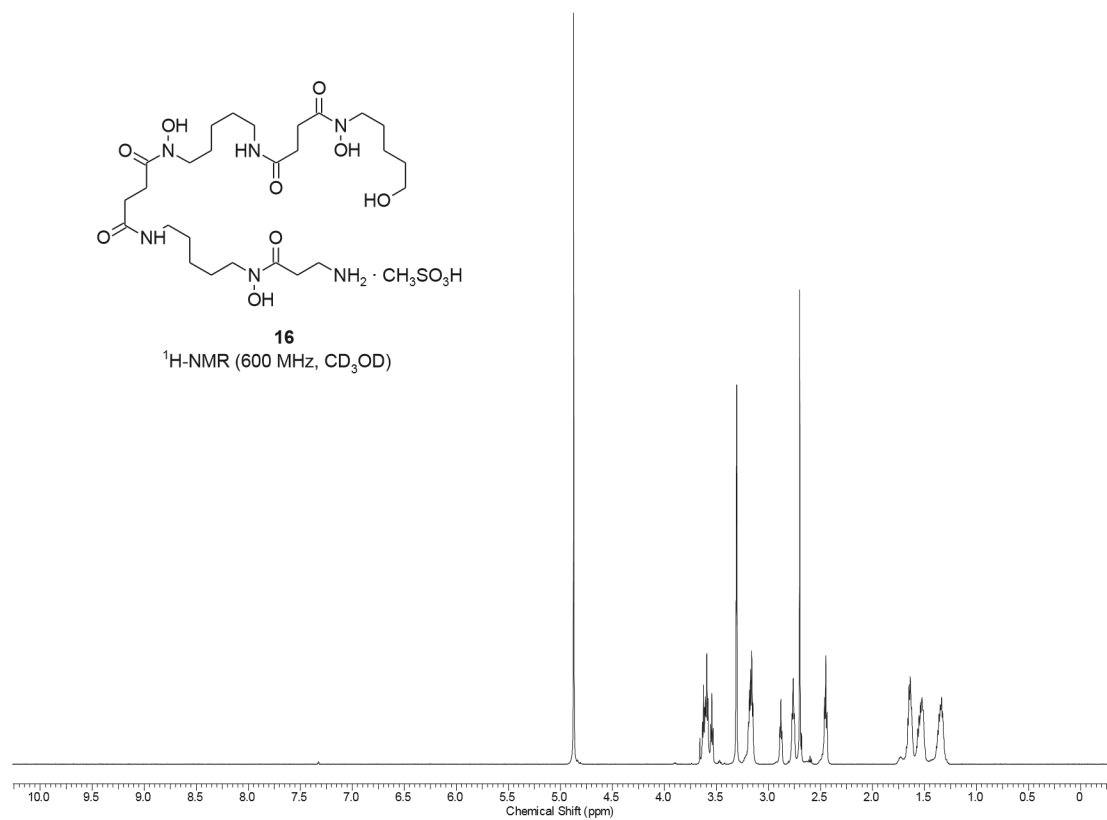
<sup>13</sup>C-NMR (150 MHz, CD<sub>3</sub>OD)

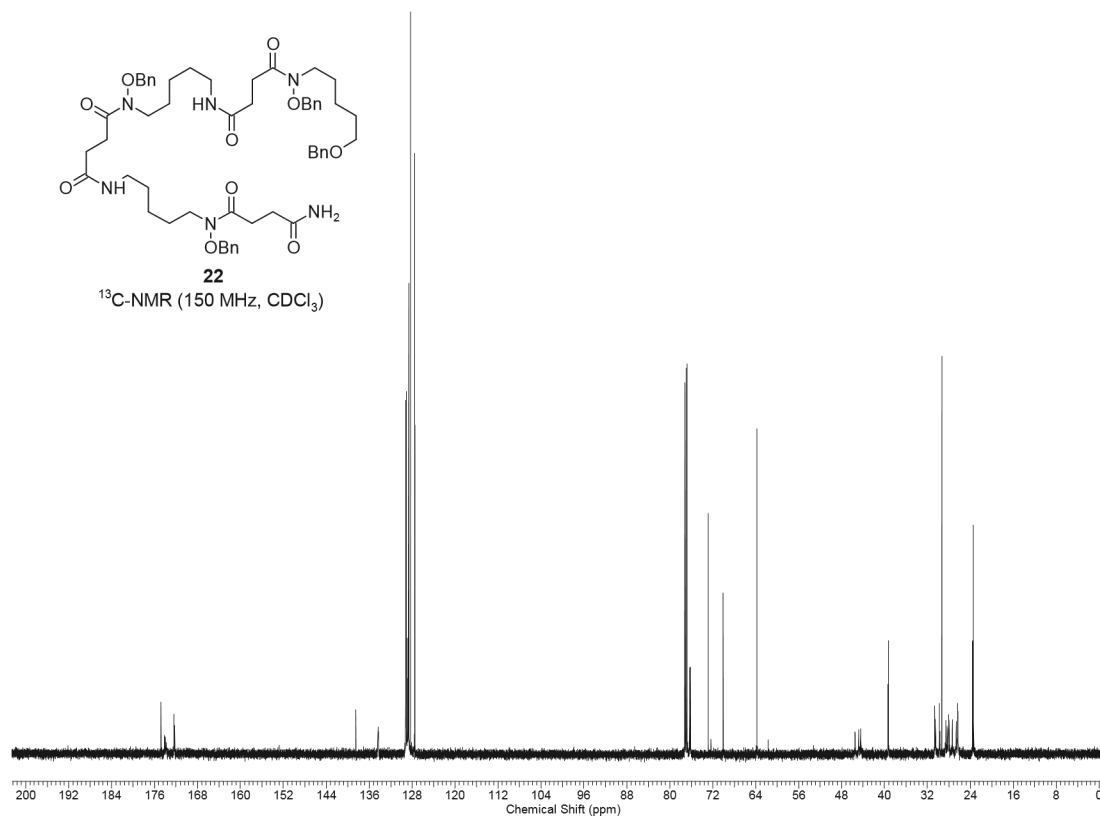
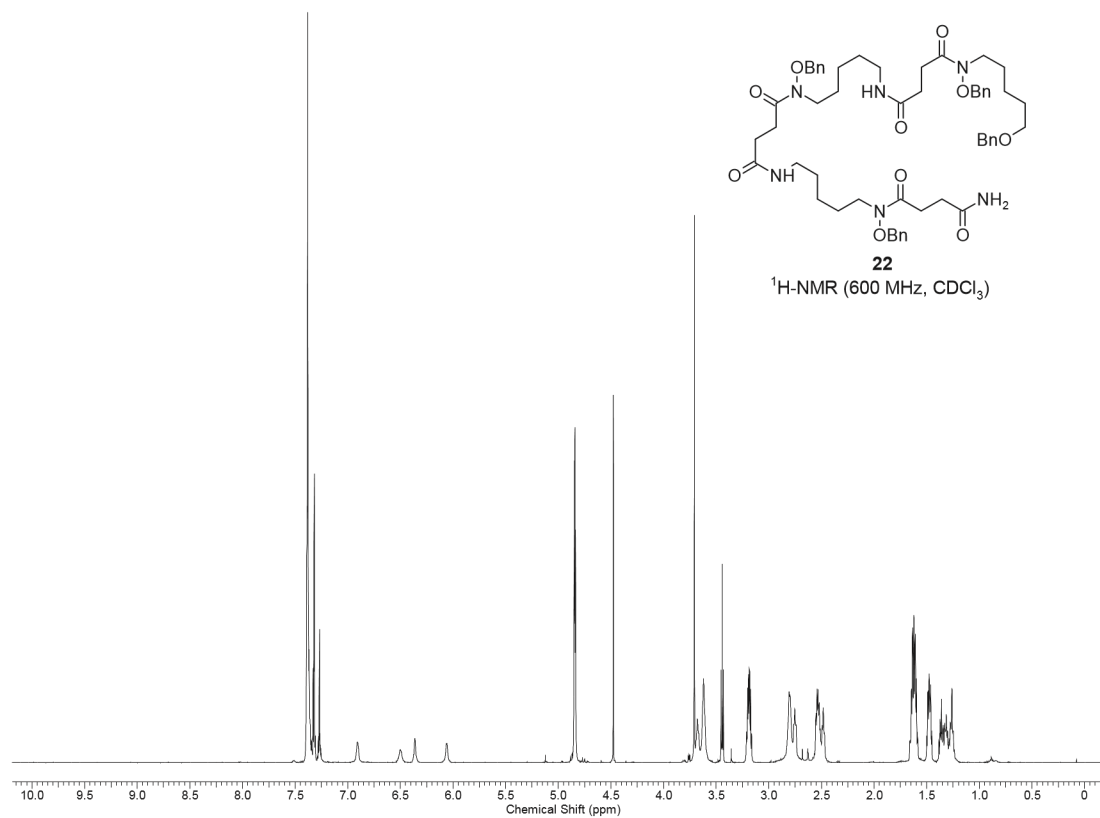
Mixture of rotamers: 1.0 : 0.26

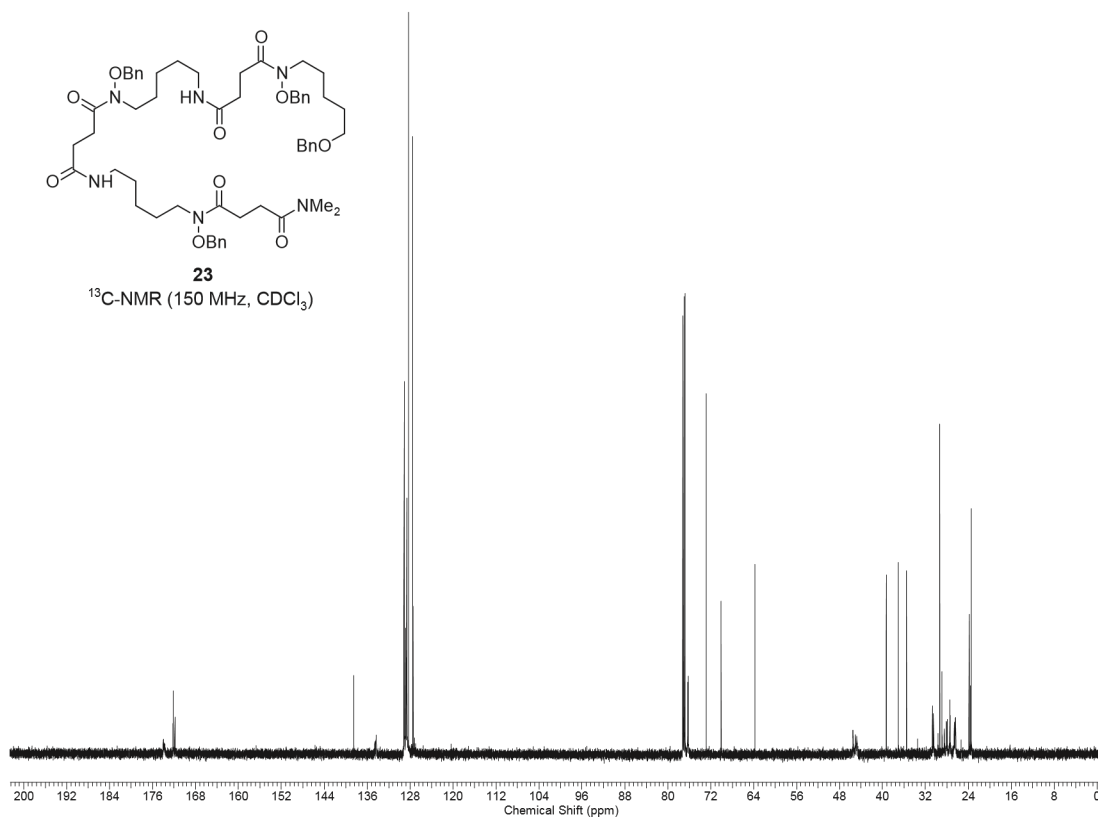
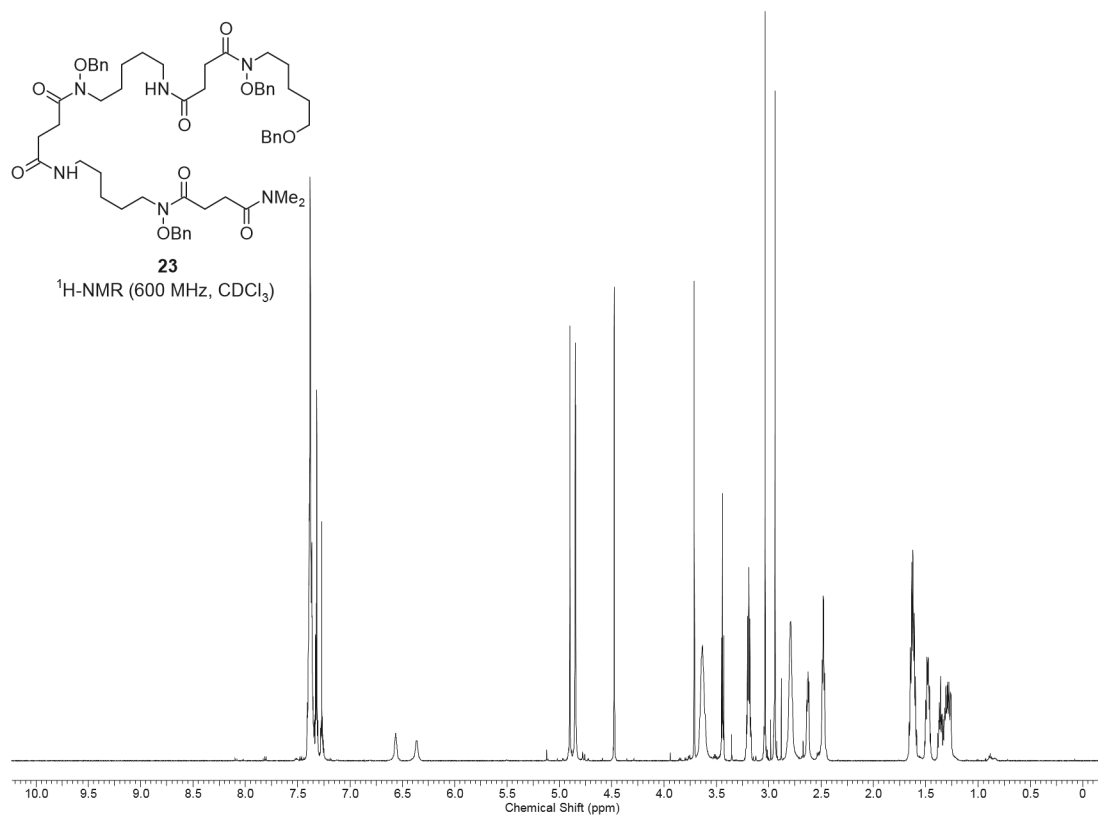


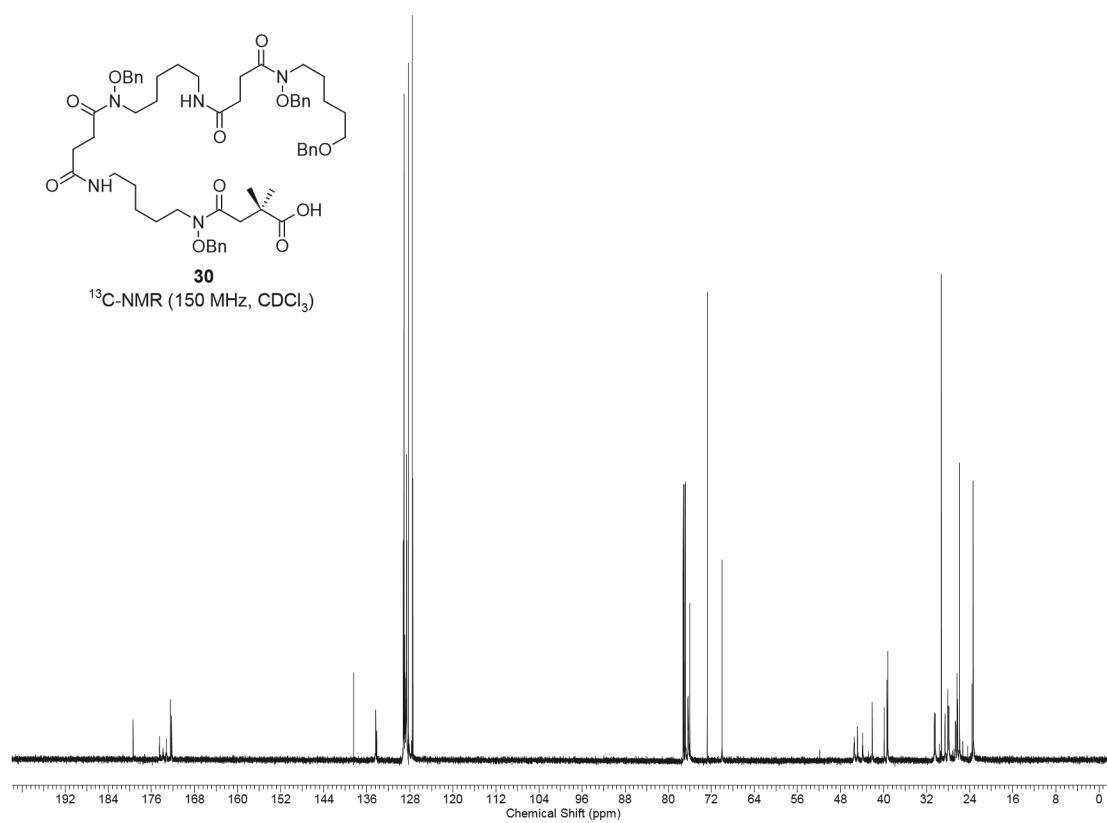
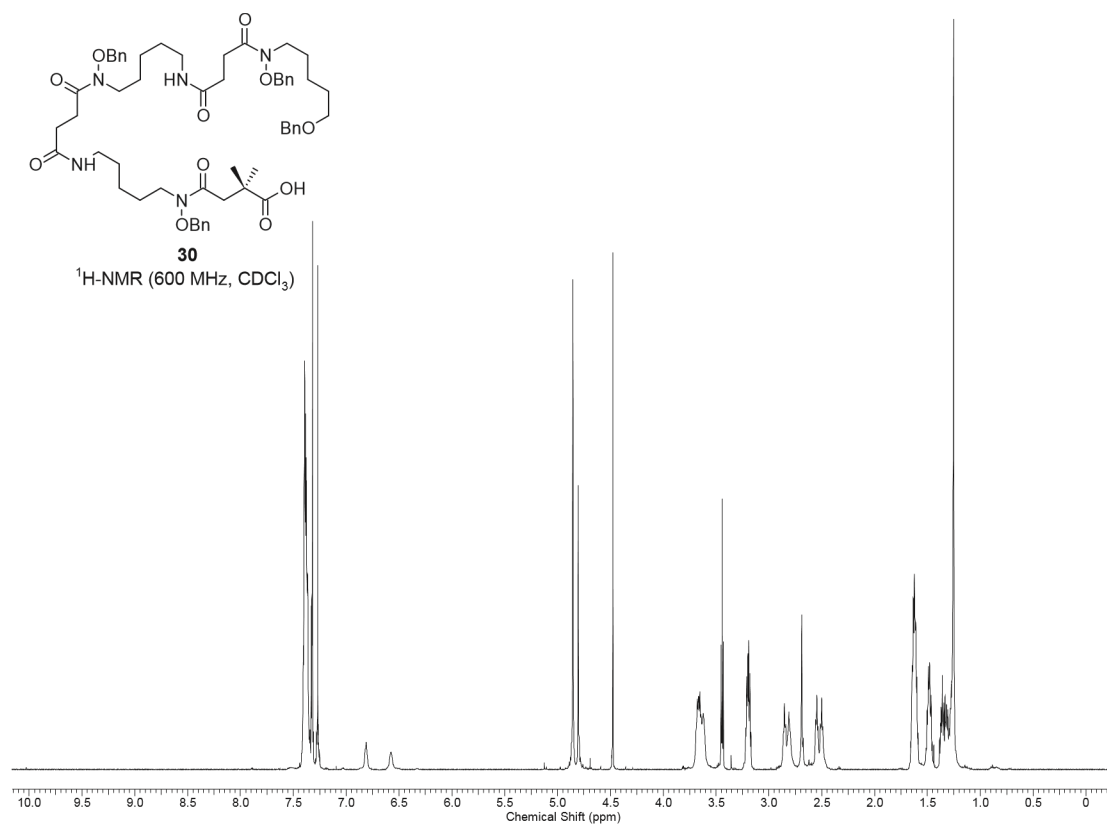


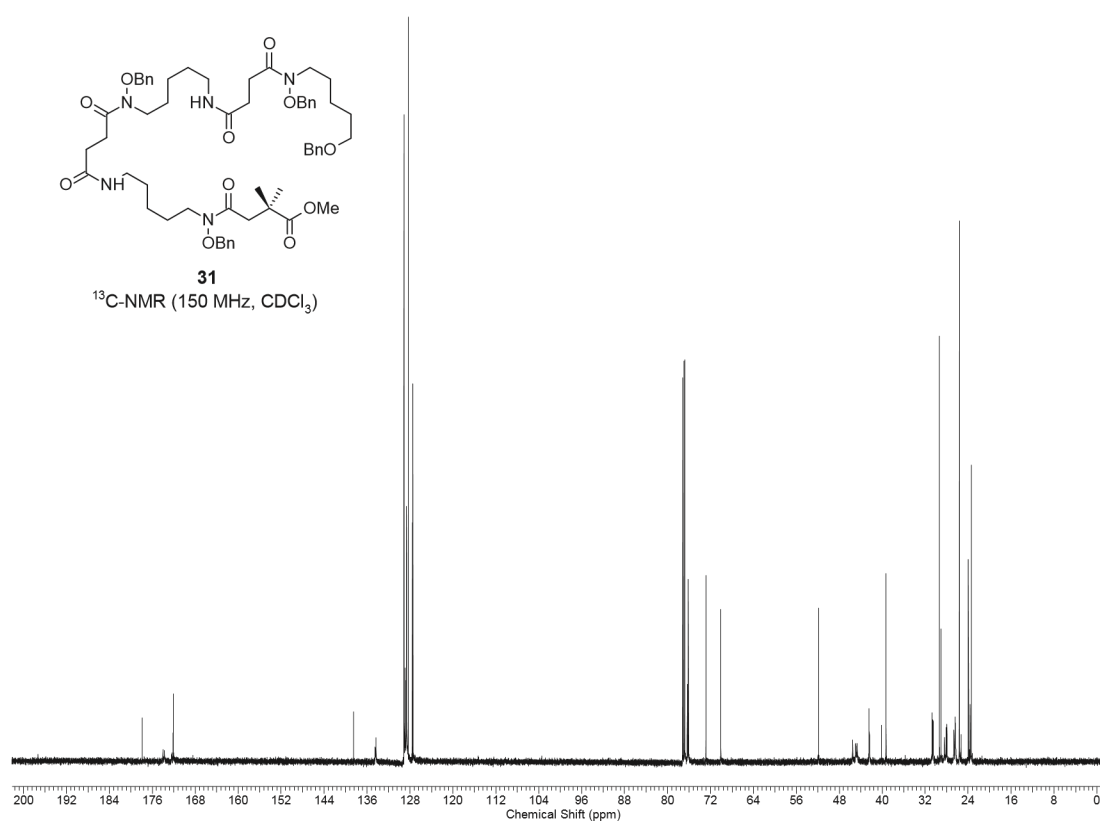
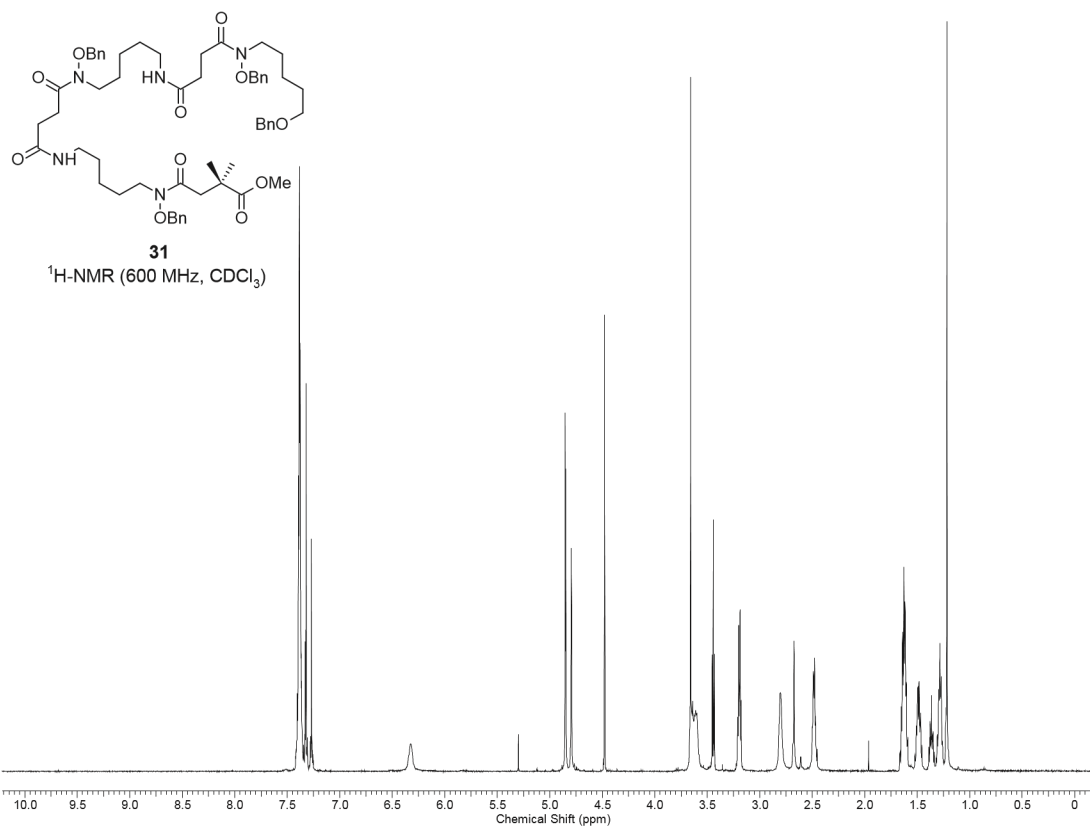


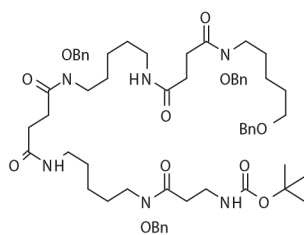






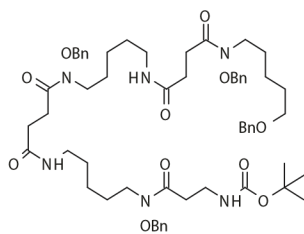
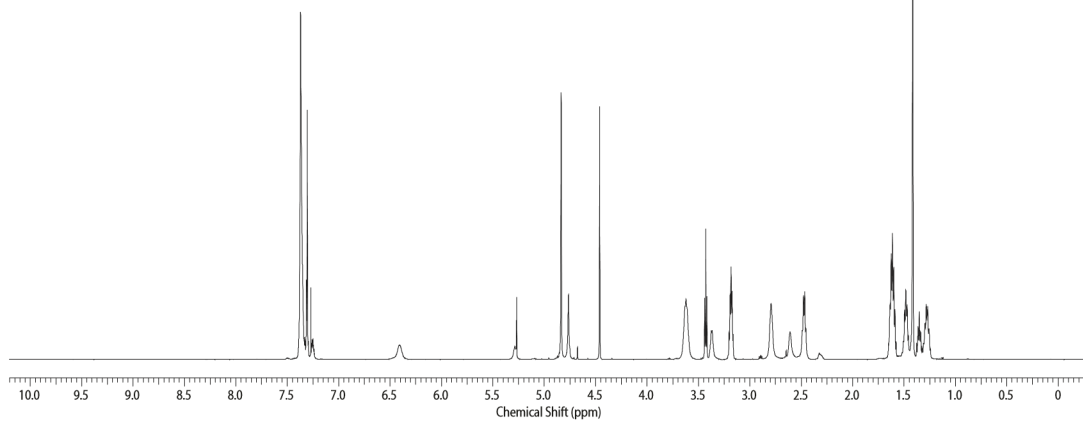






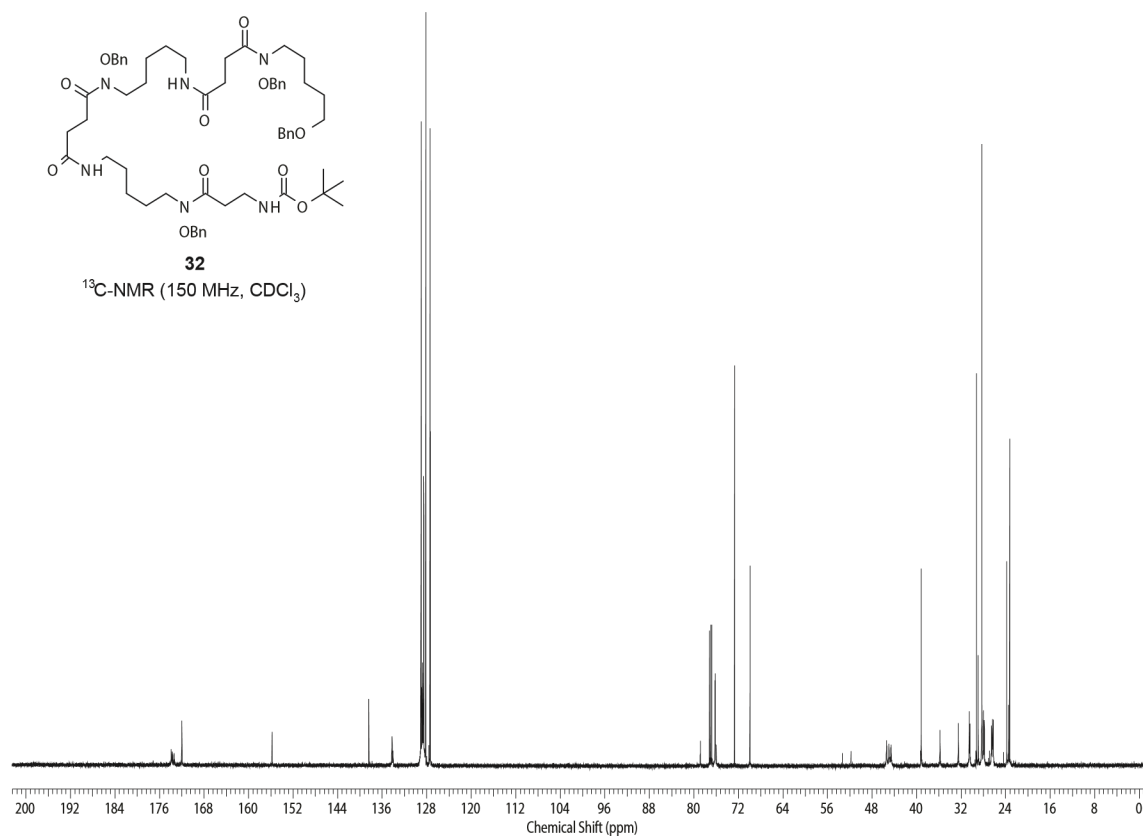
**32**

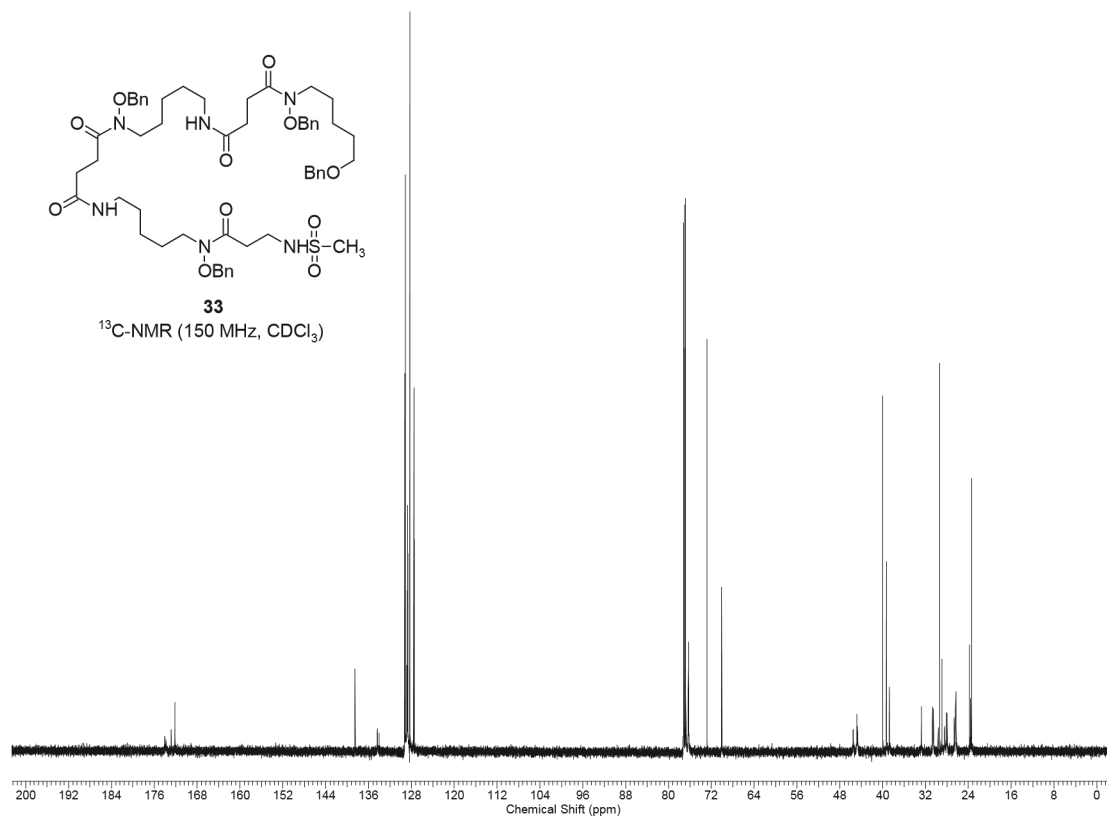
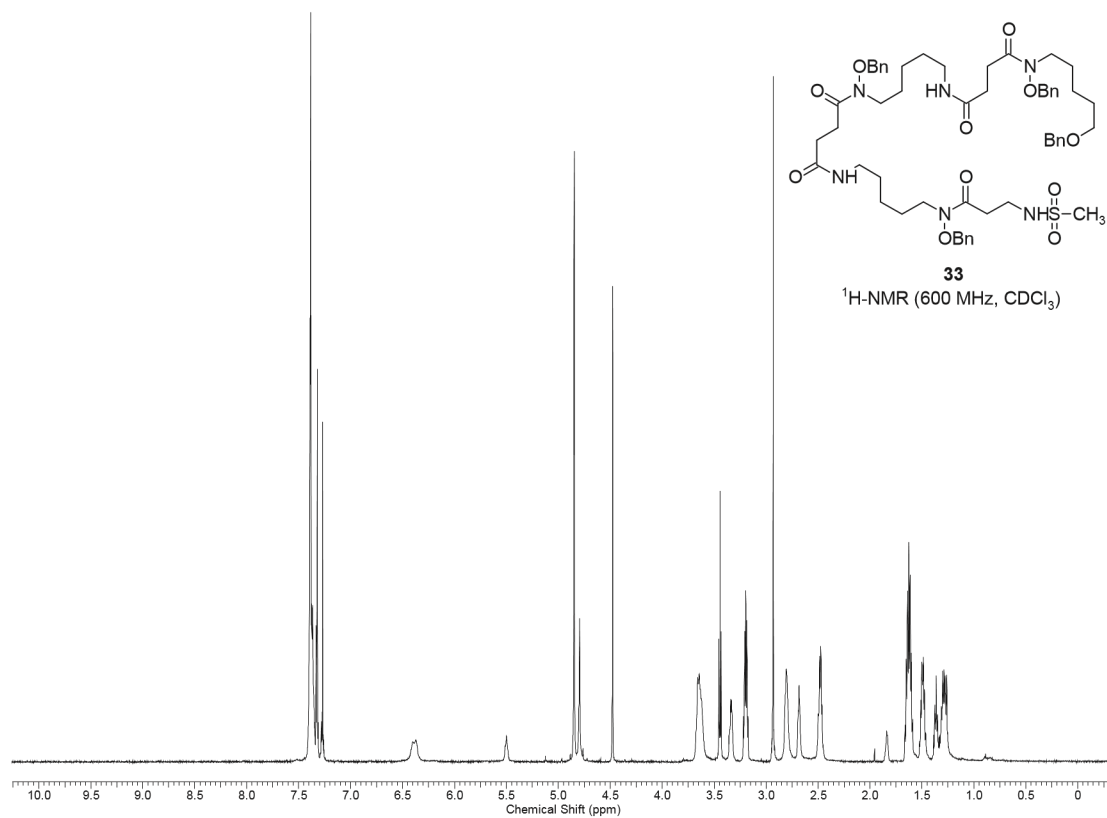
$^1\text{H-NMR}$  (600 MHz,  $\text{CDCl}_3$ )



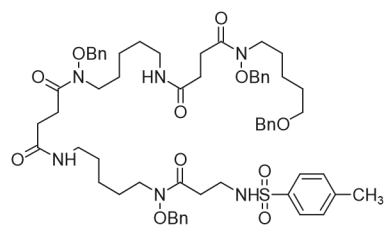
**32**

$^{13}\text{C-NMR}$  (150 MHz,  $\text{CDCl}_3$ )

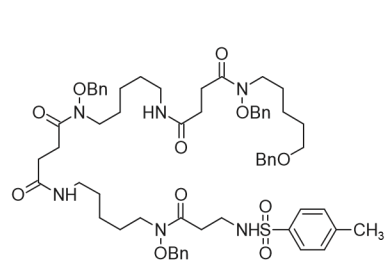
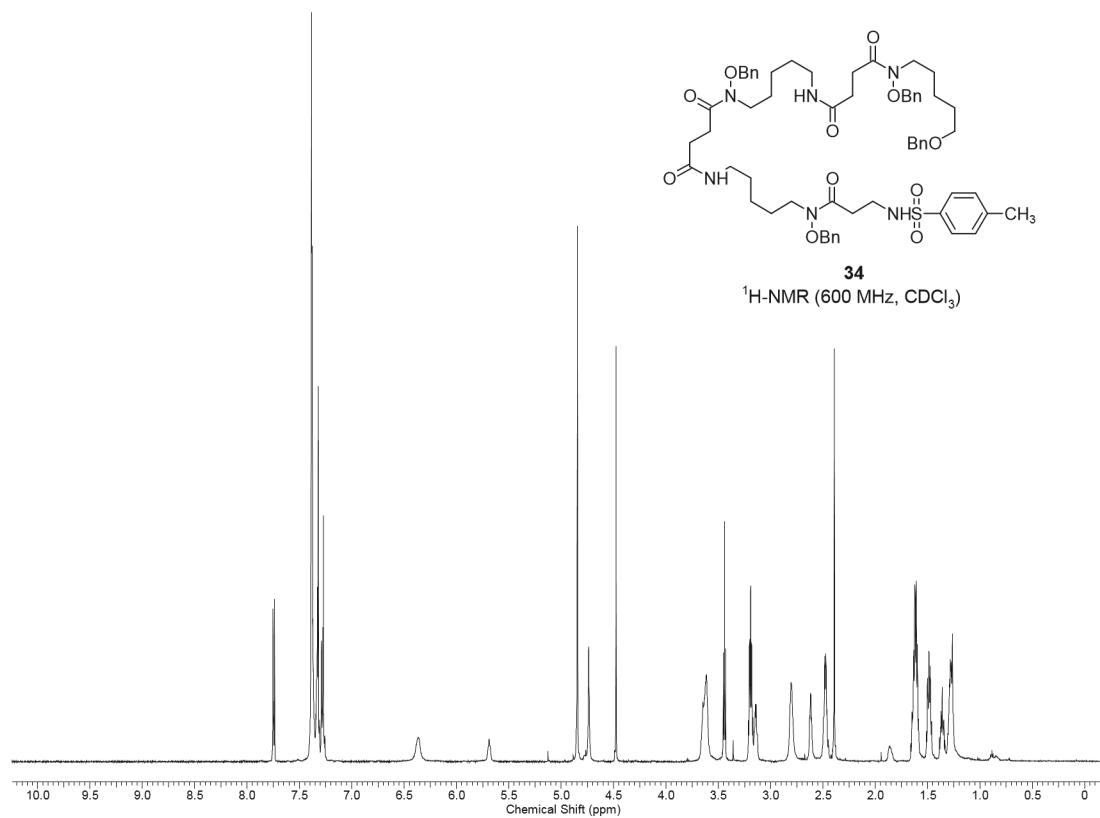




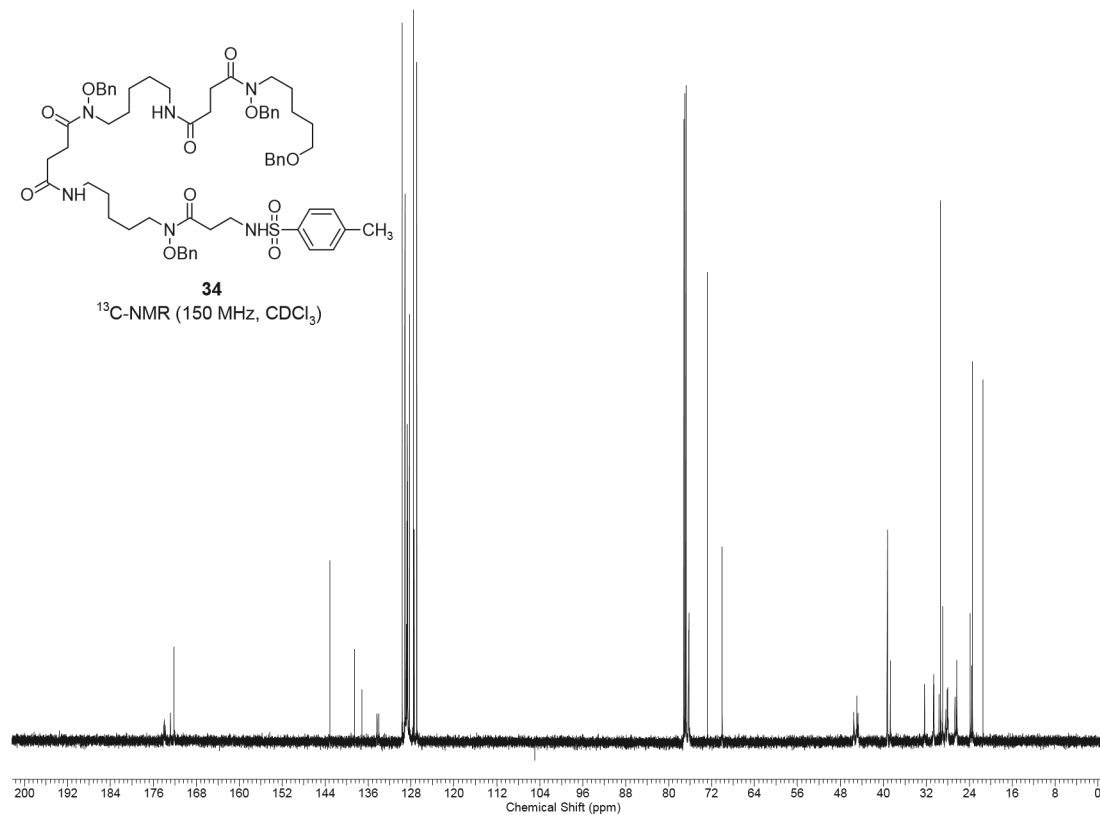


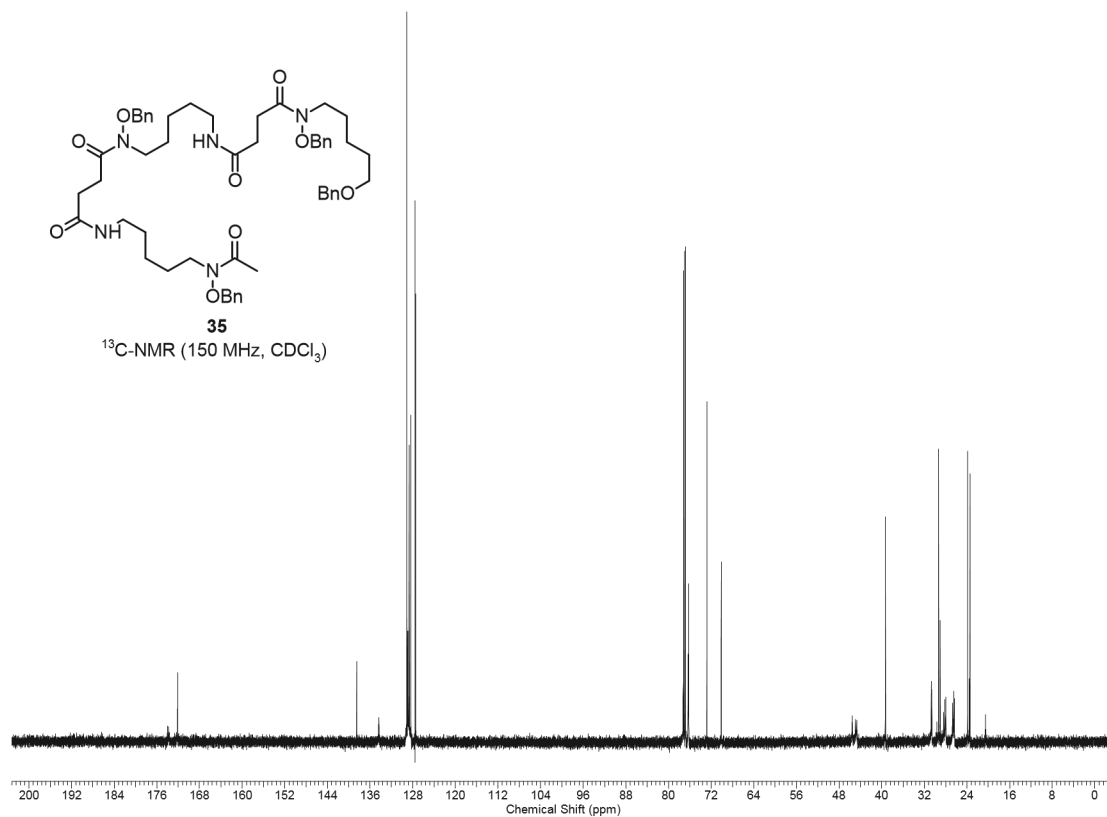
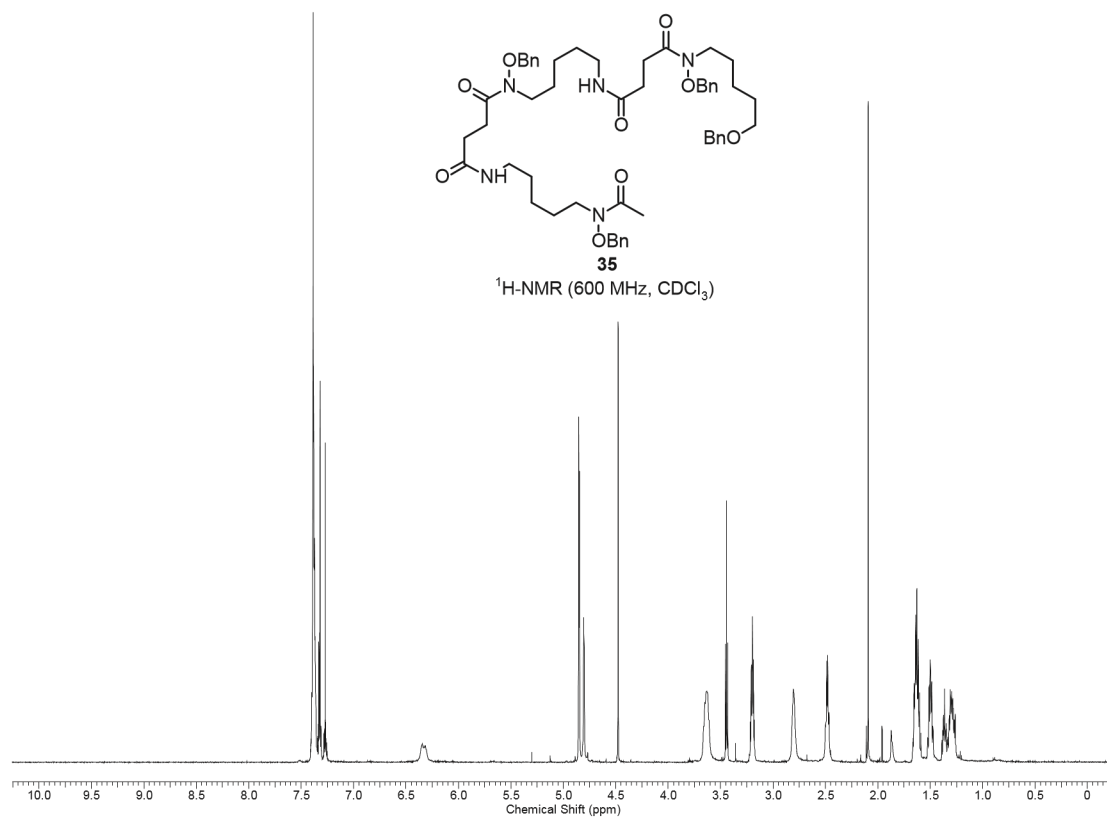


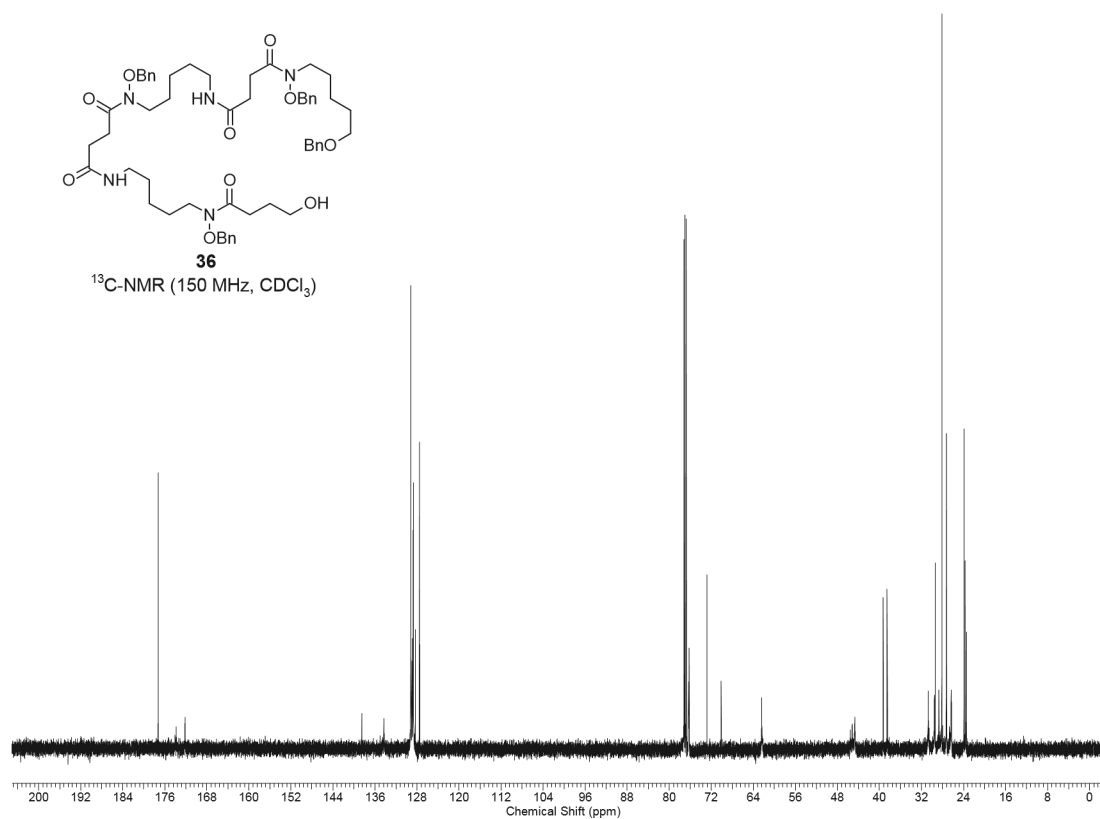
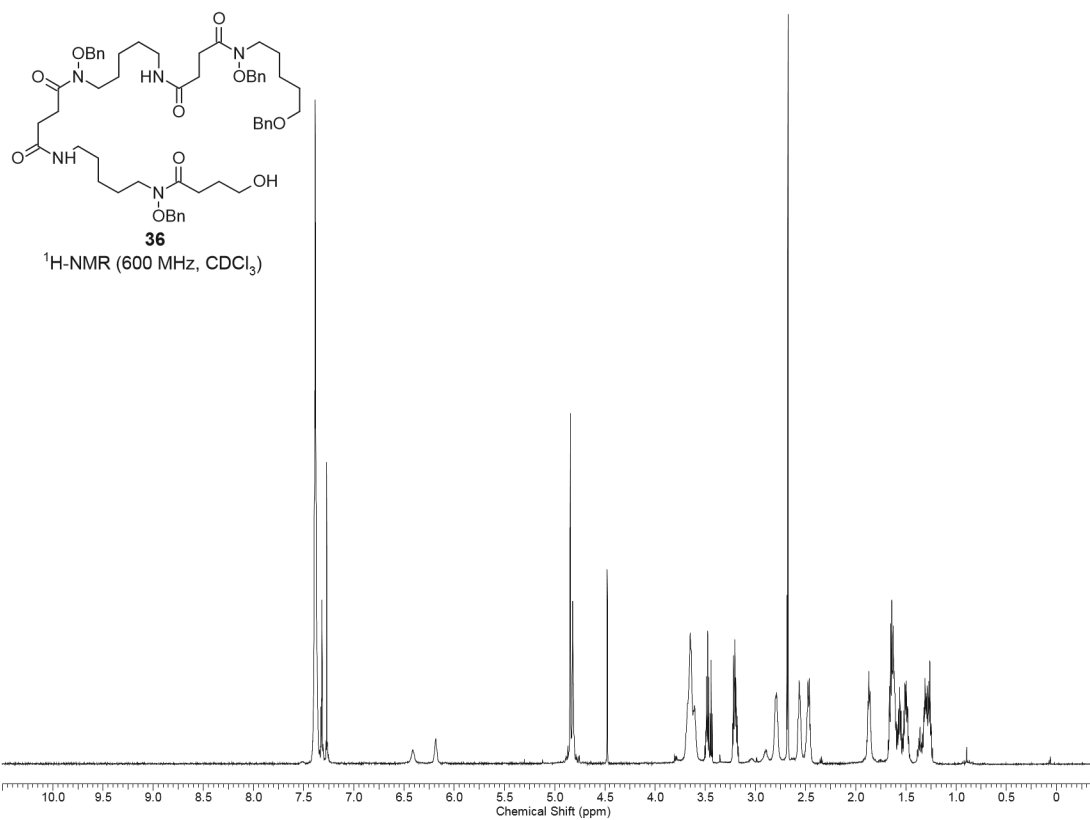
**34**  
<sup>1</sup>H-NMR (600 MHz, CDCl<sub>3</sub>)

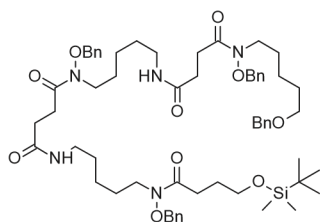


**34**  
<sup>13</sup>C-NMR (150 MHz, CDCl<sub>3</sub>)



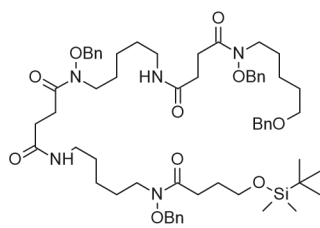
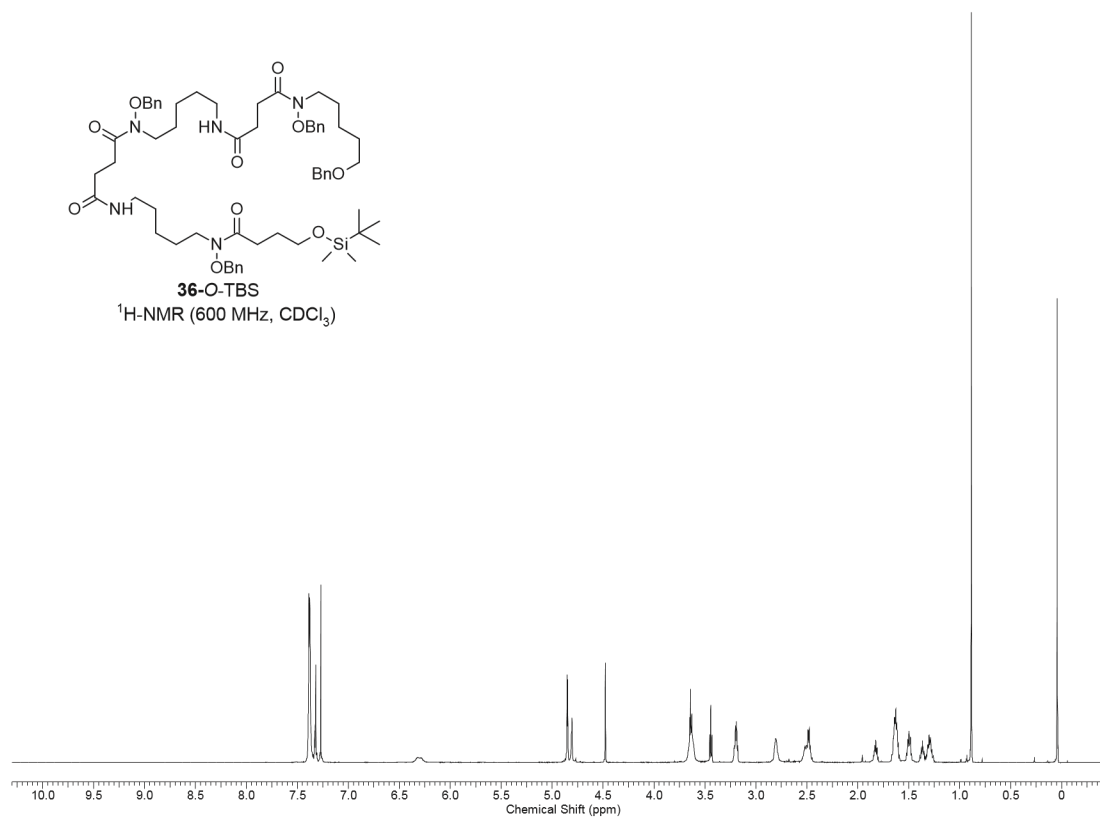






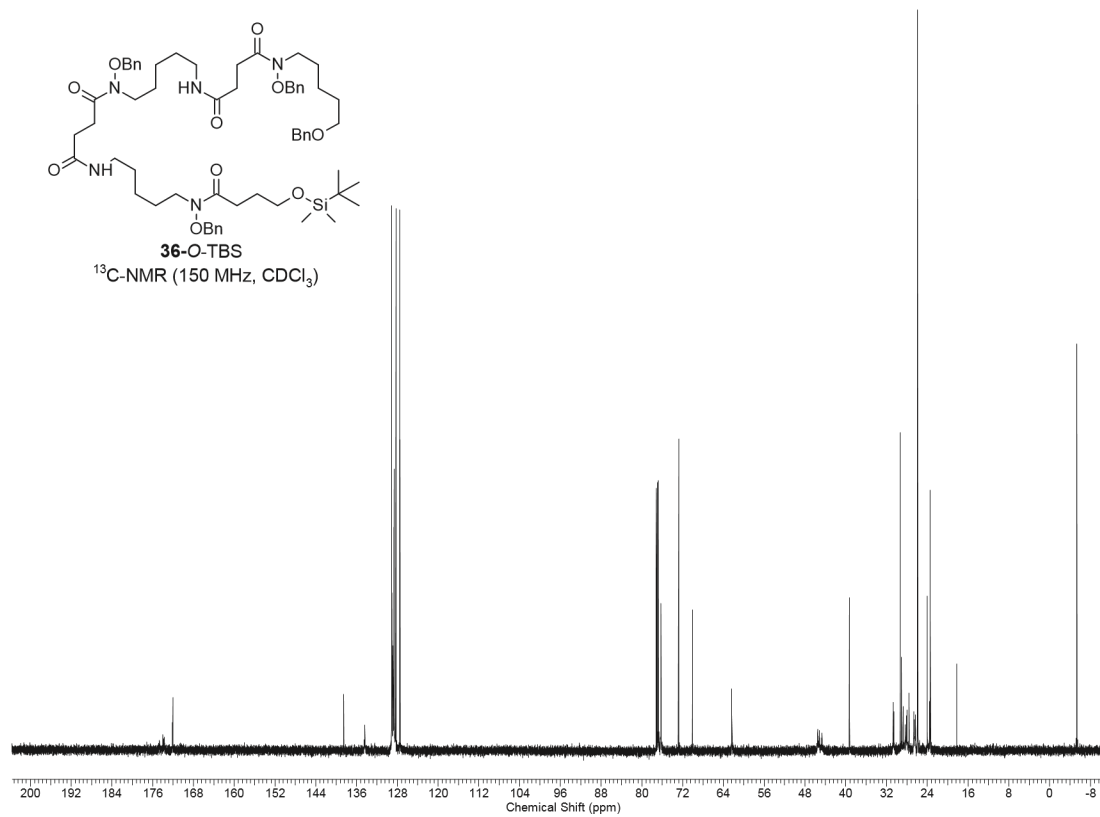
**36-O-TBS**

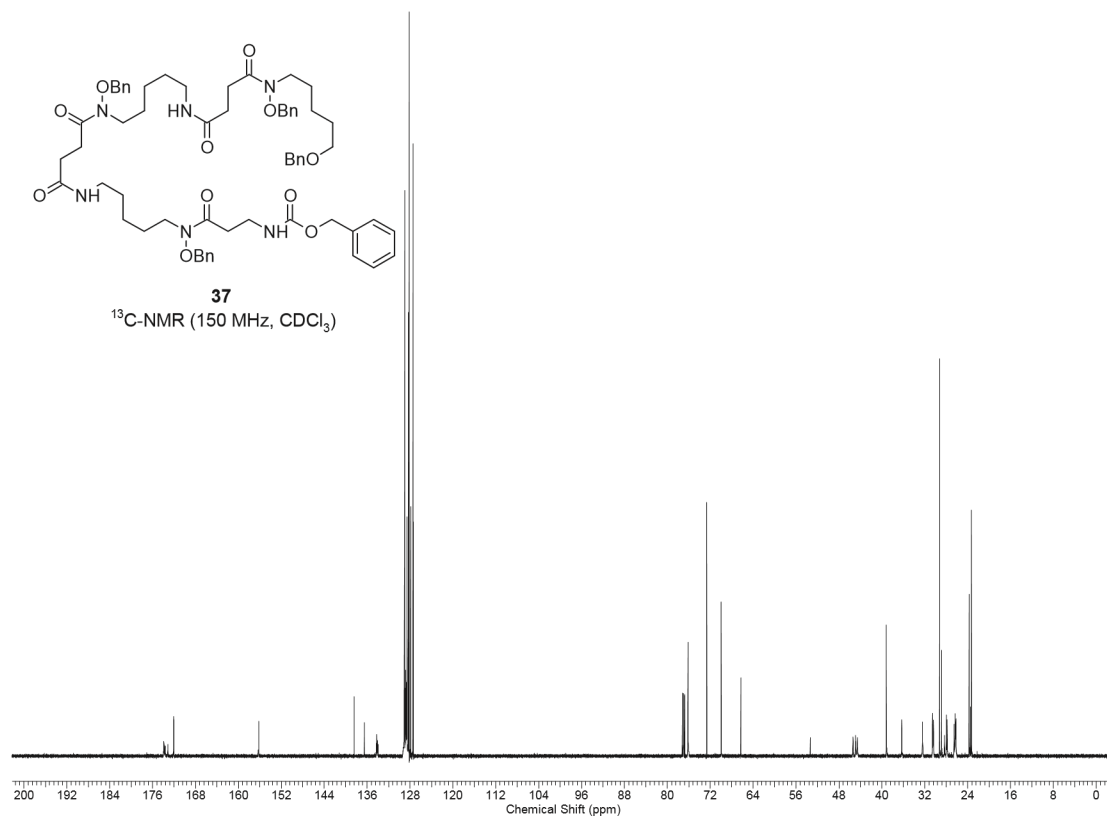
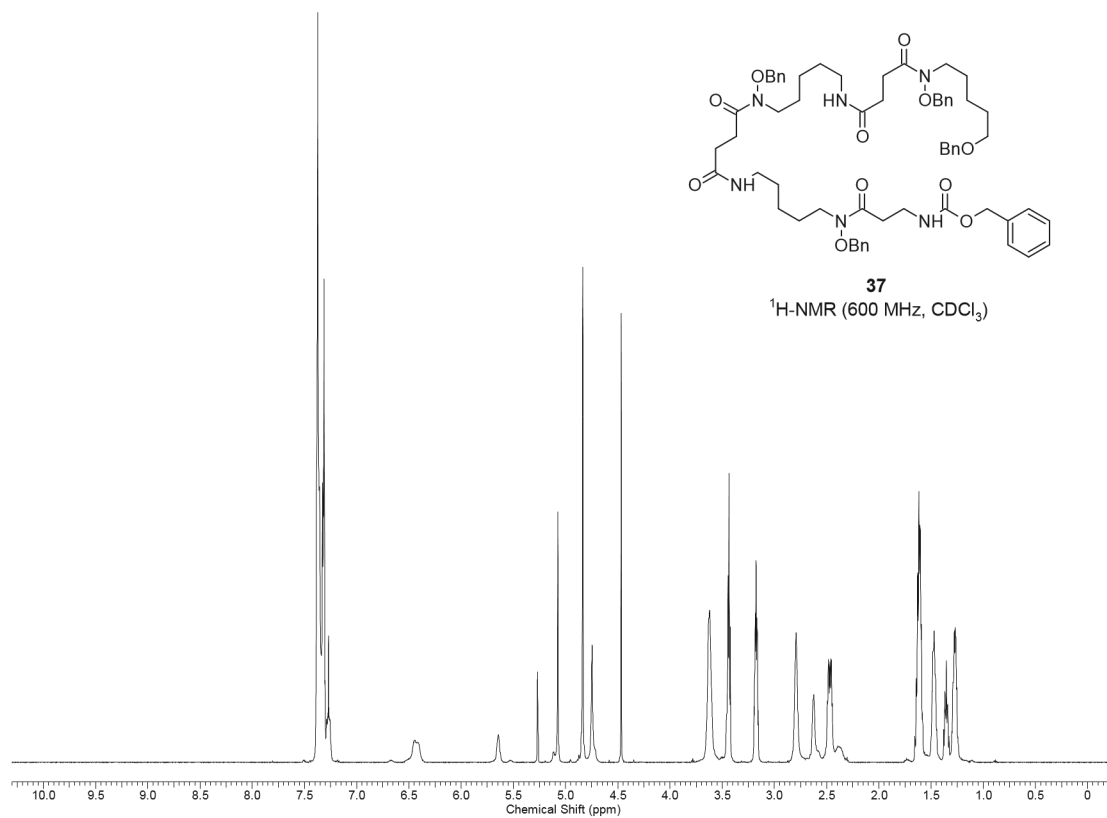
$^1\text{H-NMR}$  (600 MHz,  $\text{CDCl}_3$ )

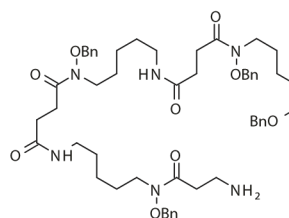


**36-O-TBS**

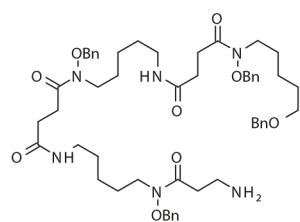
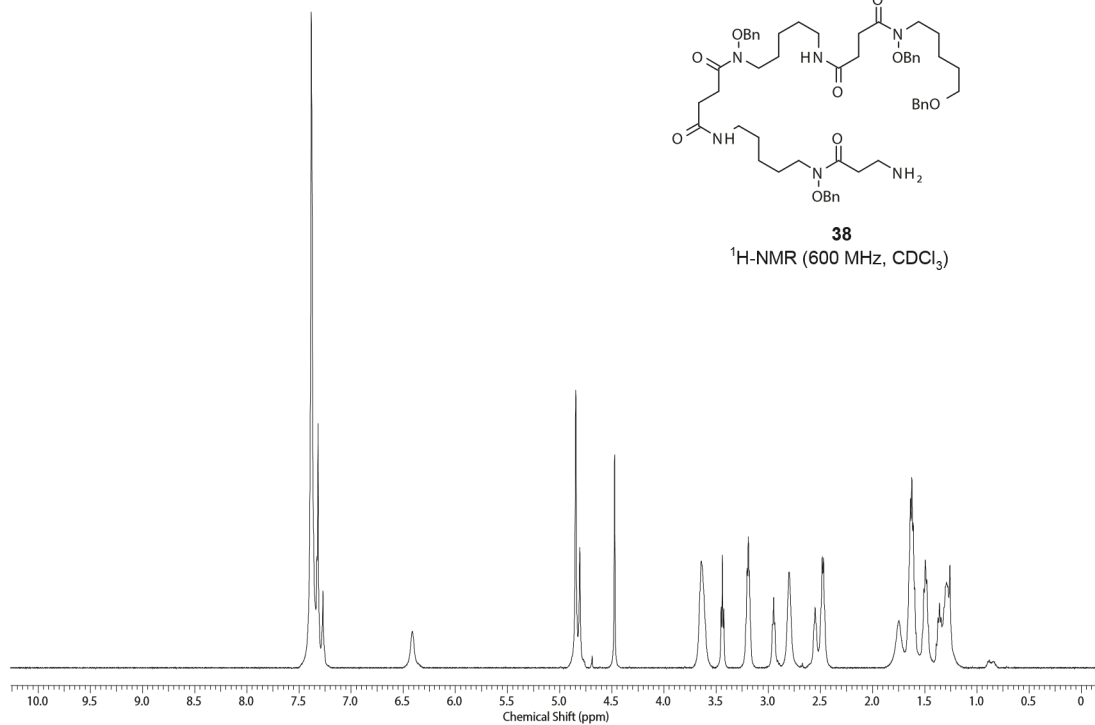
$^{13}\text{C-NMR}$  (150 MHz,  $\text{CDCl}_3$ )



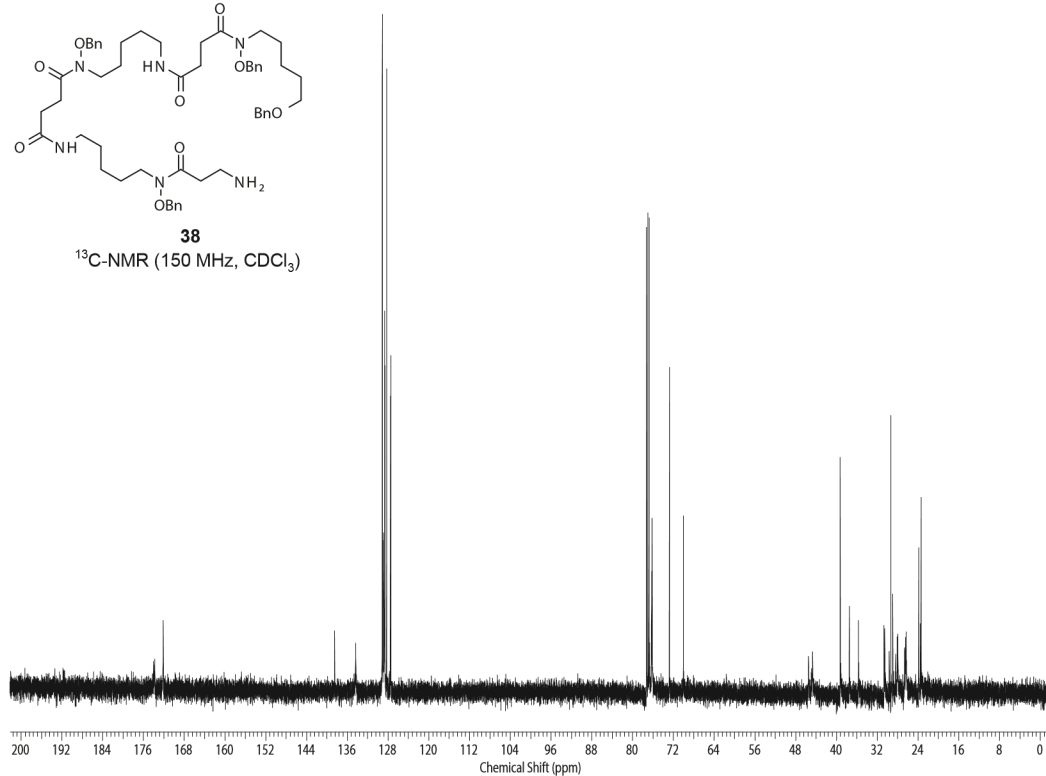




**38**  
<sup>1</sup>H-NMR (600 MHz, CDCl<sub>3</sub>)



**38**  
<sup>13</sup>C-NMR (150 MHz, CDCl<sub>3</sub>)



## VI. Acknowledgments

We thank Prof. John-Stephen Taylor (WUSTL, Dept. of Chemistry) for assistance with fluorescence quenching assays. We thank the Lizzadro Magnetic Resonance Research Center at the University of Notre Dame for access to NMR instrumentation. We thank the University of Notre Dame Mass Spectrometry and Proteomics Facility for access to MS instrumentation. We thank Prof. Marvin J. Miller and the University of Notre Dame for providing siderophore samples through a material transfer agreement.

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