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New Synthetic Approach for the Incorporation of 3,2-Hydroxypyridinone (HOPO) Ligands: Synthesis of Structurally Diverse Poly HOPO Chelators

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Abstract

The HOPO sulfonamide reagent, **3**, was prepared from commercial 2,3-dihydroxypyridine in four steps in good yields. Sulfonamide **3** readily underwent selective alkylation with dibromides in the presence of base or could be coupled to alcohols using Mitsunobu conditions. The utility of this nucleophilic HOPO reagent was demonstrated by the synthesis some tris and tetraHOPO chelators. This approach for tethering HOPO ligands is unique and flexible as shown by the preparation of HOPO/iminocarboxylic acid chelator **17**.

Keywords

3; 2-Hydroxypyridinone; synthesis; polydentate chelators; mixed ligand chelators; sulfonamide

There has been much recent interest in the synthesis of chelators containing the 3-hydroxy-2-pyridinone (3,2-HOPO) ligand arising from their ability to form strong complexes with hard metal ions.¹ 3,2-HOPO chelators have been examined for a variety of medical applications including the treatment of iron overload diseases², cancer treatment³, and as contrast agents in magnetic resonance imaging (MRI).⁴ Hydroxypyridinone chelators also form strong complexes with actinide(IV) ions leading to possible applications in environmental remediation and biological detoxification.⁵ Recently, the syntheses of HOPO chelators which also carry other ligand groups, such as hydroxamates, have been reported due to their potential biomedical applications.⁶

The direct alkylation of 2,3-dihydroxypyridine can be accomplished but requires harsh conditions and hence is not a convenient method for the preparation of polydentate chelators.⁷ The preferred approach that has been used to introduce the 3,2-HOPO ligand has involved the reaction of an amine in the spacer moiety with an activated carboxylic acid linker attached to the pyridinone ring system followed by removal of protecting groups to expose the free ligand. The HOPO has been linked to the amine spacer either through an activated ester on the ring nitrogen as depicted in **I**, or through an activated carboxylic acid group on the pyridinone ring (see **II**). While the resulting amide linkages may be advantageous in the metal ion complexation process, they can lead to polydentate chelators with reduced organic and aqueous solubility that can limit their efficacy in desired applications.

A while ago, we disclosed a method for introduction of HOPO moieties onto a variety of nucleophilic platforms including polyamines⁸ and polyphenols⁹ that uses novel bicyclic 3,2-

HOPO imidate electrophiles, **III**. While **IIIa** and **IIIb** reacted readily with secondary amines, their reaction with aliphatic primary amines generally gave amidine products, limiting the use of this methodology for the introduction of HOPO ligands onto amine platforms.

A synthon, such as **IV**, that allows the HOPO ligand to serve as a nucleophile rather than as an electrophile would permit access to a diverse array of HOPO ligands including mixed ligand systems and would complement existing methodology. In this paper, we report the preparation and applications of a nucleophilic HOPO sulfonamide reagent, **3**, which can be effectively tethered onto suitable platforms by alkylation reactions. This reagent allows access to new classes of HOPO chelators including mixed ligand systems.

The first challenge was to establish a convenient method for the preparation of HOPO amine **2**. In contrast to alkylation reactions, which require harsh conditions, 2,3-dihydropyridine undergoes Michael additions more readily. Reaction of 2,3-dihydropyridine with acrylonitrile in the presence of cesium fluoride catalyst in acetonitrile at 60 °C gave the corresponding nitrile adduct in good yield and sufficient purity for direct use in the next step. Treatment of the crude nitrile with benzyl bromide and potassium carbonate in refluxing acetonitrile gave the benzyl protected HOPO nitrile **1** which was conveniently purified by washing with ether. The selective reduction of the nitrile in the presence of the pyridinone ring was a major concern.¹⁰ However, selective reduction of the nitrile to the corresponding HOPO amine **2** could be achieved using borane-THF. It was found that the best method for the isolation of the free HOPO amine **2** involved quenching of the reaction mixture with methanol followed by stirring with concentrated HCl for 24 h. After adjusting the pH of the mixture to 8 with sodium hydroxide, the HOPO amine **2** could be extracted into CH₂Cl₂ and isolated in 93% yield in satisfactory purities.¹¹ The desired HOPO amine could be readily prepared in large scale without the need of chromatographic purification.

The amine, **2**, is a useful HOPO synthon, as it can be coupled using standard procedures to various activated acid derivatives like **I** and **II**. However, we were interested in its alkylation reactions that could lead to new chelators without the geometric constraints imposed by amide linkages. In order to achieve selective monoalkylations, it was thought best to use the corresponding *p*-nitrobenzenesulfonyl (nosyl) derivative of **2**. Treatment of HOPO amine **2** with *p*-nitrobenzenesulfonyl chloride in the presence of *N*-methylmorpholine (1.2 eq) in dichloromethane gave the desired 3,2-HOPO sulfonamide reagent **3** as a light yellow solid in 73% yield after chromatographic purification.

With the nosyl reagent **3** in hand, it was decided that the best way to demonstrate its synthetic utility was by the synthesis of polydentate chelators such as the tripodal TREN-based trisHOPO chelator **8** shown in Scheme 2. TREN-based HOPO and hydroxamate chelators have been of much interest for the binding of hard metal ions.¹² Reaction of the sulfonamide reagent **3** using triphenylphosphine and diisopropyl azodicarboxylate (DIAD) in THF at 50 °C with HOPO alcohol **4**^{8a} gave the nosyl-bis-3,2-HOPO amine **5** in 64% yield after silica gel chromatography. Attempts to remove the nosyl group using DBU and thioglycerol in DMF gave complex mixtures that were difficult to purify. However, the

nosyl group could be easily cleaved with thioacetic acid and lithium hydroxide in DMF at room temperature to furnish the bis-3,2-HOPO amine **6** in 75% yield after purification.^{13,14} Alkylation of the secondary amine, **6**, with 3,2-HOPO imidate salt, **IIIb**, in the presence of excess of triethylamine in acetonitrile at 60°C gave **7** in good yield after purification.^{8a} Removal of the benzyl groups using 1:1 HBr/AcOH gave the target tris-3,2-HOPO chelator **8** as a hydrobromide salt in 81% yield.

The availability of structurally diverse diamines has made them a well-exploited spacer group for the introduction of ligand moieties. Hence, the trisHOPO **12** and tetraHOPO **14** assembled on a propane diamine platform were attractive targets to demonstrate the usefulness of our methodology. To our satisfaction, the monoalkylation of **3** could be achieved in high yields using excess 1,3-dibromopropane in the presence of potassium carbonate in refluxing acetonitrile to give **9** in excellent yield (Scheme 3). Further alkylation of **9** with bis-3,2-HOPO amine **6** followed by removal of the nosyl group gave **11**. Debenzylation using standard reaction conditions gave the tris-3,2-HOPO chelator **12** as the hydrobromide salt. It is important to point out that the diamine platform of **12** can be easily modified by the use of other dibromides in the initial alkylation of **3**.

The above synthetic scheme was readily modified to prepare tetraHOPO chelators, a class of compounds which have been of interest for their actinide ion binding (Scheme 4).⁵ The alkylation of 1,3-dibromopropane (1 eq.) with two equivalents of bis-3,2-HOPO amine **6** using potassium carbonate in refluxing acetonitrile gave the corresponding tetrabenzyl protected HOPO **13** in good yield after purification. Removal of the benzyl protecting groups gave the tetraHOPO chelator **14** as the hydrobromide salt.

Finally, it was felt relevant to demonstrate the use of this methodology to make a mixed HOPO ligand system. The ability to access mixed HOPO ligand chelators, carrying other metal binding ligands such as iminocarboxylic acids and hydroxamic acids would provide new classes of molecules with interesting coordination chemistry and biological activity.⁶ The iminoacetic acid/HOPO chelator **17** was chosen as a target for this synthetic study. Dialkylation of 1,3-dibromopropane with 2 equivalents of sulfonamide HOPO **3** gave the desired bisHOPO **15** in 74% yield along with 6% of the monoalkylated product **9**. The nosyl groups of **15** were removed using standard conditions and the resulting diamine was used without purification. Dialkylation of the crude diamine with *t*-butyl bromoacetate in acetonitrile in the presence of potassium carbonate gave **16** in 59% yield for two steps after purification. The concurrent removal of both benzyl and *t*-butyl protecting groups in the last step using 1:1 concentrated HBr/glacial acetic acid is a matter of convenience and gave the desired mixed ligand chelator **17** as a light brown solid (Scheme 5). The chelator, **17**, is water soluble as might be expected.

In conclusion, the development of strategies and synthetic methodologies for the incorporation of the 3,2-HOPO moiety into a diverse array of platforms is of great importance due to the potential environmental and biomedical applications of these metal ion chelators. The chemistry presented in this paper is applicable to the synthesis of a broad array of polydentate 3,2-HOPO targets and related analogs. The amine **2** can be prepared from commercially available material in three steps and is a useful reagent for direct coupling with carboxylic acids to tether the HOPO moiety. It can be converted to its nosyl derivative **3** in good yields. The sulfonamide **3** can undergo selective alkylation reactions with dibromides or can participate in Mitsunobu reactions with alcohols under mild conditions. The usefulness of this synthetic approach has been demonstrated by the synthesis of some novel tris and tetra HOPO chelators. The synthesis of mixed HOPO-iminoacetic acid chelator **17**, highlights the use of this method to access desired mixed ligand targets. Also, the approach is amenable to the preparation of hexadentate chelators such as

trisHOPO **11** which have a functional handle for the introduction of a fluorescent reporter group¹⁵ or attachment to other biomolecules¹⁶ of interest.

¹H NMR (200 MHz) and ¹³C NMR (50 MHz) spectra were recorded on a Varian XL 200. ¹H NMR (300 MHz) and ¹³C NMR (75 MHz) spectra were recorded on a 300 MHz Varian NMR. ¹H NMR (400 MHz) spectra were recorded on a Varian Unity 400 spectrometer. NMR spectral samples were prepared in CDCl₃ or D₂O as noted. Chemical shifts are given in parts per million (ppm) relative to tetramethylsilane (TMS) reference. For ¹H spectra obtained in D₂O, the HOD peak was set to 4.72 ppm and used as an internal reference. For ¹³C spectra obtained in D₂O, 1,4-dioxane (66.5 ppm) was used as an internal reference. Analytical and preparative thin layer chromatography was performed on silica 60/F254 plastic plates (EM Science). Column chromatography was performed on silica gel (Merck 60–200 mesh) or basic alumina (Aldrich 150 mesh). Chromatography solvents were reagent grade and were obtained from either Fisher Scientific or VWR Scientific. Reagents were obtained from Aldrich or Lancaster chemical companies and were used as received. Anhydrous THF was collected from a GlassContourTM solvent purification system. Other dry solvents (CH₃CN, CH₂Cl₂, etc) were obtained from Acros. Desert Analytics, Tucson, Arizona performed elemental analyses.

Nitrile 1

To a solution of 2,3-dihydroxypyridine (10.0 g, 90 mmol) in CH₃CN (110 mL) was added cesium fluoride (1.37 g, 9 mmol) and acrylonitrile (29 mL, 450 mmol) and the solution heated at reflux for 3 d. The solvent and excess acrylonitrile were removed *in vacuo*. The product was dissolved in hot CH₃CN (2 × 100 mL) and the solids filtered off. The solvent was removed under reduced pressure and the resulting solid washed with cold water (50 mL) at 0 °C for 1 h. The product was collected *via* vacuum filtration and then dried under reduced pressure to give the crude nitrile as a solid (10.7 g, 72%) which was used without purification.: IR (neat) 3217, 2247, 1656, 1605 cm⁻¹; ¹H NMR (200 MHz, CDCl₃): δ = 2.93 (t, *J* = 6.2 Hz, 2H), 4.23 (t, *J* = 6.2 Hz, 2H), 6.24 (t, *J* = 7.3 Hz, 1H), 6.76–6.94 (m, 3H); ¹³C NMR (50 MHz, CDCl₃): δ = 17.3, 46.4, 107.5, 114.7, 116.8, 126.9, 146.7, 158.4. To a solution of the crude nitrile (7.0 g, 42.6 mmol) in CH₃CN (70 mL) was added potassium carbonate (6.48 g, 46.9 mmol) and benzyl bromide (5.6 mL, 46.9 mmol) and the mixture was heated at reflux for 19 h. The solvent and excess benzyl bromide were removed *in vacuo*. The residue was dissolved CH₂Cl₂ (100 mL) and washed with saturated NaHCO₃ (2 × 60 mL). The aqueous layer was extracted once more with CH₂Cl₂ (40 mL). The combined organic layers were then washed with saturated NaCl (2 × 100 mL), dried (MgSO₄), and filtered. The solid product was washed with diethyl ether (2 × 50 mL) and collected *via* vacuum filtration. The white crystalline product was dried under reduced pressure to give nitrile **1** (10.3 g, 95.3%). \

Mp 138–140°C;

IR (KBr) 2946, 2246, 1652, 1611, 1457 cm⁻¹;

¹H NMR (200 MHz, CDCl₃): δ = 2.94 (t, *J* = 6.6 Hz, 2H), 4.19 (t, *J* = 6.6 Hz, 2H), 5.12 (s, 2H), 6.11 (t, *J* = 7.0 Hz, 1H), 6.70 (dd, *J* = 1.5, 7.7 Hz, 1H), 6.98 (dd, *J* = 1.5, 7.0 Hz, 1H), 7.30–7.46 (m, 5H);

¹³C NMR (50 MHz, CDCl₃): δ = 16.9, 46.2, 70.7, 105.2, 115.9, 117.1, 127.2, 128.0, 128.4, 128.8, 135.9, 148.8, 157.7;

Anal. Calcd. for C₁₅H₁₄N₂O₂: C, 70.85; H, 5.55; N, 11.02. Found: C, 70.46; H, 5.25; N, 10.89.

Amine 2

To a solution of nitrile **1** (2.00 g, 7.87 mmol) in anhydrous THF (40 mL) at 0 °C was added $\text{BH}_3 \cdot \text{THF}$ (11.6 mL, 11.7 mmol) and the solution stirred at rt for 39 h. The reaction mixture was cooled to 0 °C, quenched with MeOH and the solvent removed *in vacuo*. The crude product was dissolved in MeOH (40 mL), cooled to 0 °C and the pH adjusted to 2 with concentrated HCl. After stirring at rt for 24 h, the solvent was removed *in vacuo* and the resulting thick oil was washed with EtOAc (3 × 5 mL). The remaining residue was dissolved in water (15 mL) and cooled to 0 °C. The pH was adjusted to 8 with 20% aqueous NaOH and the product extracted into CH_2Cl_2 (3 × 100 mL), dried (Na_2SO_4), filtered and solvent was removed *in vacuo*. The crude amine **2** (1.88 g, 93%) was dried under reduced pressure and used without purification.¹⁰

IR (neat) 3367, 2944, 1652, 1601 cm^{-1} ;

^1H NMR (400 MHz, CDCl_3): δ = 1.86–1.93 (m, 2H), 2.71 (t, J = 6.6 Hz, 2H), 4.09 (t, J = 6.8 Hz, 2H), 5.12 (s, 2H), 6.03 (t, J = 7.0 Hz, 1H), 6.64 (dd, J = 1.7, 7.2 Hz, 1H), 6.92 (dd, J = 1.8, 6.8 Hz, 1H), 7.30–7.45 (m, 5H).

Sulfonamide 3

A solution of p-nitrobenzenesulfonyl chloride (0.96 g, 4.33 mmol) in 10 mL CH_2Cl_2 was added to a solution of amine **2** (1.12 g, 4.34 mmol) and N-methylmorpholine (0.57 mL, 5.19 mmol) in CH_2Cl_2 at 0 °C. After stirring at rt for 4 h, the solvent was removed *in vacuo* and the residue dissolved in 15% $\text{CH}_2\text{Cl}_2/\text{EtOAc}$ (80 mL). The organic layer was washed with water (2 × 50 mL), dried (Na_2SO_4) and the solvent removed *in vacuo*. The crude product was purified by silica gel chromatography (30% EtOAc/hexane to 2% MeOH/ CHCl_3) to give sulfonamide HOPO **3** as a pale yellow solid (1.41 g, 73%).

Mp 165–167 °C;

IR (KBr) 3103, 1652, 1599, 1521 cm^{-1} ;

^1H NMR (400 MHz, CDCl_3): δ = 1.92–1.98 (m, 2H), 2.85–2.89 (m, 2H), 4.12 (t, J = 6.3 Hz, 2H), 5.09 (s, 2H), 6.12 (t, J = 7.0 Hz, 1H), 6.67 (dd, J = 1.4, 7.4 Hz, 1H), 6.84 (dd, J = 1.6, 6.8 Hz, 1H), 6.92 (unres t, 1H), 7.33–7.43 (m, 5H), 8.08 (d, J = 8.8 Hz, 2H), 8.26 (d, J = 9.0 Hz, 2H);

^{13}C NMR (50 MHz, CDCl_3 with few drops of CD_3OD): δ = 29.6, 39.4, 46.2, 70.8, 106.3, 115.9, 124.1, 127.3, 128.2, 128.4, 128.6, 135.7, 146.2, 148.6, 149.8, 158.6.

Anal. Calcd. for $\text{C}_{21}\text{H}_{21}\text{N}_3\text{O}_6\text{S}$, C: 56.87, H: 4.77; N: 9.48. Found, C: 56.57, H: 4.94, N: 9.18.

Sulfonamide 5

A solution of HOPO alcohol **4** (0.591 g, 2.28 mmol) in anhydrous THF (10 mL) was added to a suspension of sulfonamide HOPO **3** (1.01 g, 2.28 mmol) and triphenylphosphine (2.09 g, 7.97 mmol) in THF (15 mL) and the mixture stirred at rt for 5 min.

Diisopropylazodicarboxylate (0.88 mL, 4.54 mmol) was slowly added and the reaction mixture heated at 50 °C for 24 h. The solvent was removed under reduced pressure and the residue purified by silica gel chromatography (60–90% EtOAc/hexane) to give sulfonamide **5** as a light red solid (0.99 g, 64%).

Mp 45–46 °C;

IR (KBr) 1654, 1603, 1528 cm^{-1} ;

^1H NMR (300 MHz, CDCl_3): δ = 2.02–2.12 (m, 4H), 3.25 (t, J = 7.6 Hz, 4H), 4.01 (t, J = 7.3 Hz, 4H), 5.10 (s, 4H), 6.04 (t, J = 7.3 Hz, 2H), 6.65 (dd, J = 1.8, 7.3 Hz, 2H), 6.95 (dd, J = 1.7, 7.0 Hz, 2H), 7.32–7.44 (m, 10H), 7.92 (d, J = 9.1 Hz, 2H), 8.33 (d, J = 8.8 Hz, 2H);

^{13}C NMR (50 MHz, CDCl_3): δ = 28.4, 46.4, 47.3, 70.8, 105.0, 115.5, 124.5, 127.3, 128.0, 128.3, 128.5, 128.9, 136.2, 144.6, 148.8, 150.0, 158.1.

Anal. Calcd. for $\text{C}_{36}\text{H}_{36}\text{N}_4\text{O}_8\text{S}\cdot 0.5\text{H}_2\text{O}$, C: 62.32, H: 5.38, N: 8.08. Found, C: 62.03, H: 5.18, N: 8.19.

Amine 6

Mercaptoacetic acid (0.24 mL, 3.45 mmol) and lithium hydroxide monohydrate (0.303 g, 7.22 mmol) were added to a solution of sulfonamide HOPO **5** (1.17 g, 1.71 mmol) in dimethylformamide (5 mL) and the mixture stirred at rt for 5.5 h. The solvent was removed under reduced pressure. The residue was dissolved in CH_2Cl_2 (80 mL) and washed with saturated NaHCO_3 (3×60 mL) dried (Na_2SO_4) and the solvent removed under reduced pressure. The crude product was purified by column chromatography on basic alumina (0–10% MeOH/EtOAc) to give amine **6** as a viscous red oil (0.64 g, 75%).

IR (neat) 3302, 1652, 1602 cm^{-1} ;

^1H NMR (400 MHz, CDCl_3): δ = 1.90–1.97 (m, 4H), 2.59 (t, J = 6.6 Hz, 4H), 4.08 (t, J = 6.8 Hz, 4H), 5.11 (s, 4H), 6.00 (t, J = 7.2 Hz, 2H), 6.62 (dd, J = 1.8, 7.4 Hz, 2H), 6.96 (dd, J = 1.6, 6.8 Hz, 2H), 7.29–7.44 (m, 10H);

^{13}C NMR (50 MHz, CDCl_3): δ = 29.4, 46.0, 47.3, 70.8, 104.5, 115.7, 127.3, 127.9, 128.5, 129.3, 136.4, 148.8, 158.2;

Anal. Calcd. for $\text{C}_{30}\text{H}_{33}\text{N}_3\text{O}_4\cdot 0.12\text{CHCl}_3$, C: 70.39, H: 6.50, N: 8.18. Found: C, 70.08, H: 6.44, N: 8.37.

Amine 7

A solution of triethylamine (0.17 mL, 1.22 mmol) in dry CH_3CN (1 mL) was added to a solution of benzyl protected HOPO amine **6** (0.30 g, 0.6 mmol) in dry CH_3CN (2 mL). A solution of HOPO iminium salt **IIIb**¹¹ (0.30 g, 0.90 mmol) in CH_3CN (6 mL) was added and the reaction mixture was heated at 60 °C for 22 h. The volatiles were removed under reduced pressure and the residue purified on basic alumina (0–2% MeOH/EtOAc). The benzyl protected trisHOPO **7** was obtained as a viscous brown oil (0.32 g, 72%).

IR (neat) 1651, 1603, 1555 cm^{-1} ;

^1H NMR (400 MHz, CDCl_3): δ = 1.87–1.95 (m, 6H), 2.52 (t, J = 6.6 Hz, 6H), 4.06 (t, J = 7.6 Hz, 6H), 5.09 (s, 6H), 6.00 (t, J = 7.0 Hz, 3H), 6.62 (dd, J = 1.4, 7.4 Hz, 3H), 6.99 (dd, J = 1.2, 7.0 Hz, 3H), 7.27–7.44 (m, 15H);

^{13}C NMR (50 MHz, CDCl_3): δ = 26.4, 48.4, 50.4, 70.6, 104.5, 115.3, 127.2, 127.9, 128.5, 129.4, 136.3, 148.7, 158.0.

Anal. Calcd. for $\text{C}_{45}\text{H}_{48}\text{N}_4\text{O}_6$, C: 72.95, H: 6.53, N: 7.56. Found: C: 73.17, H: 6.66, N: 7.42.

Chelator 8

Benzyl protected tris-HOPO amine **7** (0.30 g, 0.412 mmol) was dissolved 1:1 conc. HBr/AcOH (20 mL) and stirred at rt for 67 h. The volatiles were removed under reduced pressure. The residue was washed with EtOAc and ether and dried under high vacuum for 24 h. The product was lyophilized to yield trisHOPO chelator **8** (0.264 g, 81%) as a light brown tetrahydrobromide salt.

IR (KBr) 3408, 1645, 1543 cm^{-1} ;

^1H NMR (D_2O , 300 MHz): δ = 1.96–2.06 (m, 6H), 3.09–3.14 (m, 6H), 3.98 (t, J = 7.0 Hz, 6H), 6.30 (t, J = 7.0 Hz, 3H), 6.89 (dd, J = 1.8, 7.6 Hz, 3H), 7.07 (dd, J = 1.8, 7.0 Hz, 3H);

^{13}C NMR (75 MHz, D_2O): δ = 23.2, 46.7, 49.7, 109.0, 118.8, 128.8, 145.4, 158.3.

Anal. Calcd. for $\text{C}_{24}\text{H}_{30}\text{N}_4\text{O}_6 \cdot 4\text{HBr}$, C: 36.30, H: 4.32, N: 7.05. Found: C: 36.05, H: 4.22, N: 6.89.

Sulfonamide 9

Sulfonamide HOPO **3** (0.60 g, 1.35 mmol) and 1,3-dibromopropane (1.5 mL, 14.8 mmol) were dissolved in anhydrous CH_3CN (12 mL) followed by the addition of potassium carbonate (0.37 g, 2.70 mmol) and the mixture was heated at reflux for 21 h. The solvent was removed under reduced pressure. The residue was diluted with EtOAc (60 mL), washed with water (2×20 mL), dried (Na_2SO_4), and the solvent removed in vacuo. The crude product was purified by silica gel chromatography (50–60% EtOAc-hexane) to give **9** as a viscous yellow oil (0.712 g, 93%).

IR (neat) 1652, 1603, 1531 cm^{-1} ;

^1H NMR (400 MHz, CDCl_3): δ = 2.08–2.16 (m, 4H), 3.25–3.33 (m, 4H), 3.40 (t, J = 6.5 Hz, 2H), 4.02 (t, J = 7.0 Hz, 2H), 5.11 (s, 2H), 6.08 (t, J = 6.8 Hz, 1H), 6.67 (d, J = 7.0 Hz, 1H), 6.94 (d, J = 6.6 Hz, 1H), 7.31–7.44 (m, 5H), 7.98 (d, J = 8.0 Hz, 2H), 8.37 (d, J = 8.8 Hz, 2H);

^{13}C NMR (50 MHz, CDCl_3): δ = 30.5, 31.9, 33.9, 48.9, 49.4, 49.5, 72.8, 107.1, 117.5, 126.5, 129.4, 130.1, 130.4, 130.6, 130.8, 138.2, 146.8, 151.0, 152.2, 160.2.

Anal. Calcd. for $\text{C}_{24}\text{H}_{26}\text{BrN}_3\text{O}_6\text{S} \cdot 0.8\text{CHCl}_3$ C: 45.13; H: 4.09; N: 6.37. Found, C: 45.40; H: 4.44; N: 6.67.

Sulfonamide 10

A solution of bromide **9** (0.64 g, 1.14 mmol) in dry CH_3CN (15 mL) was added to a suspension of benzyl protected HOPO amine **6** (0.53 g, 1.06 mmol) and potassium carbonate (0.59 g, 4.2 mmol) in anhydrous CH_3CN (15 mL) and the mixture heated at reflux for 48 h. The solvent was removed under reduced pressure and the residue was diluted with CHCl_3 (60 mL) and washed with saturated NaHCO_3 (1×40 mL). The aqueous layer was again extracted with CHCl_3 (1×40 mL) and the combined organic layer was dried (Na_2SO_4) and the solvent removed in vacuo. The crude product was purified on basic alumina (60–70 % EtOAc/hexane to 2% MeOH–EtOAc) to give **10** as a yellow solid (0.749 g, 72%).

Mp: 42–43 $^\circ\text{C}$;

IR (neat) 1652, 1603, 1528 cm^{-1} ;

^1H NMR (400 MHz, CDCl_3): δ = 1.71–1.80 (m, 2H), 1.83–1.92 (m, 4H), 2.00–2.10 (m, 2H), 2.40–2.49 (m, 6H), 3.22 (t, J = 7.3 Hz, 2H), 3.33 (t, J = 9.1 Hz, 2H), 3.97–4.03 (m, 6H), 5.08 (s, 2H), 5.09 (s, 4H), 5.97–6.03 (m, 3H), 6.63 (dd, J = 1.5, 7.6 Hz, 3H), 6.97 (d, J = 6.7 Hz, 3H), 7.26–7.43 (m, 15H), 8.01 (d, J = 8.8 Hz, 2H), 8.32 (d, J = 8.8 Hz, 2H);

^{13}C NMR (50 MHz, CDCl_3): δ = 26.4, 26.9, 28.4, 46.4, 47.3, 47.6, 48.3, 50.6, 50.8, 70.6, 104.4, 104.7, 115.2, 115.4, 124.4, 127.2, 127.8, 128.3, 129.1, 129.2, 136.1, 136.2, 144.8, 148.7, 149.8, 157.8, 157.9;

Anal. Calcd. for $\text{C}_{54}\text{H}_{58}\text{N}_6\text{O}_{10}\text{S}\cdot 0.4\text{CHCl}_3$, C: 63.38; H: 5.71; N: 8.15. Found, C: 63.47; H: 5.64; N: 8.41.

Diamine 11

Mercaptoacetic acid (0.132 mL, 1.9 mmol) and lithium hydroxide monohydrate (0.16 g, 3.8 mmol) were added to a solution of sulfonamide **10** (0.94 g, 0.95 mmol) in N,N' -dimethylformamide (6 mL) and the solution stirred at rt for 3 h. The solvent was removed under reduced pressure. The residue was diluted with CHCl_3 (80 mL) and washed with saturated NaHCO_3 (40 mL). The aqueous layer was again extracted with CHCl_3 (50 mL). The combined organic layer was washed with saturated NaHCO_3 (60 mL), dried (Na_2SO_4) and the solvent removed under reduced pressure. The crude product was purified on basic alumina (2–10% MeOH/EtOAc) to give amine **11** as a red oil (0.590 g, 78%).

IR (neat) 3302, 1652, 1602 cm^{-1} ;

^1H NMR (300 MHz, CDCl_3): δ = 1.56–1.65 (m, 2H), 1.85–1.97 (m, 6H), 2.47 (t, J = 6.7 Hz, 6H), 2.58–2.64 (m, 4H), 3.97–4.06 (m, 6H), 5.09 (s, 6H), 5.95–6.02 (m, 3H), 6.62 (dd, J = 1.5, 7.3 Hz, 3H), 6.92–6.96 (m, 3H), 7.28–7.44 (m, 15H);

^{13}C NMR (75 MHz, CDCl_3): δ = 26.4, 27.0, 29.3, 46.3, 47.3, 48.0, 48.3, 50.7, 51.5, 70.6, 104.4, 115.4, 127.2, 127.8, 128.4, 129.3, 136.3, 148.8, 158.0;

Anal. Calcd. for $\text{C}_{48}\text{H}_{55}\text{N}_5\text{O}_6\cdot 0.7\text{CHCl}_3$, C: 66.35; H: 6.37; N: 7.94. Found, C: 66.73; H: 6.45; N: 8.31.

Chelator 12

Benzyl protected HOPO **11** (0.60 g, 0.745 mmol) was dissolved in 1:1 conc HBr/AcOH (28 mL) and the solution stirred at rt for 66 h. The volatiles were removed under reduced pressure. The residue was washed with EtOAc and ether and dried under high vacuum for 24 h. The product was lyophilized to yield trisHOPO chelator **12** (0.63 g, 90%) as a tetrahydrobromide salt.

IR (KBr) 3400, 2960, 1640, 1547 cm^{-1} ;

^1H NMR (400 MHz, D_2O): δ = 2.03–2.16 (m, 8H), 3.01 (t, J = 7.4 Hz, 2H), 3.08 (t, J = 7.6 Hz, 2H), 3.17–3.27 (m, 6H), 4.04–4.11 (m, 6H), 6.35–6.40 (m, 3H), 6.95–6.99 (m, 3H), 7.12–7.16 (m, 3H);

^{13}C NMR (50 MHz, D_2O): δ = 20.5, 23.2, 25.5, 44.2, 44.6, 46.6, 46.7, 49.9, 109.0, 118.9, 119.0, 129.0, 145.2, 145.3, 158.4, 158.6.

Anal. Calcd. for $\text{C}_{27}\text{H}_{37}\text{N}_5\text{O}_6\cdot 4\text{HBr}\cdot \text{H}_2\text{O}$, C: 37.31; H: 4.99; N: 8.06. Found, C: 37.53; H: 4.69; N: 8.38.

Diamine 13

1,3-Dibromopropane (30.6 μ L, 0.30 mmol) was added to a suspension of benzyl protected bisHOPO **6** (0.30 g, 0.60 mmol) and potassium carbonate (0.21 g, 1.5 mmol) in anhydrous CH_3CN (6 mL) and the mixture was heated at reflux for 2 d. The solvent was removed under reduced pressure. The residue was diluted with CHCl_3 (50 mL) and washed with NaHCO_3 (30 mL). The aqueous layer was again extracted with CHCl_3 (30 mL). The combined organic layer was dried (Na_2SO_4) and the solvent removed under reduced pressure. The crude product was purified on basic alumina (2% MeOH-EtOAc) to provide benzyl protected HOPO **13** (0.21 g, 67%) as a viscous oil.

IR (neat) 2949, 2811, 1651, 1602, 1555 cm^{-1} ;

^1H NMR (300 MHz, CDCl_3): δ = 1.46–1.60 (m, 2H), 1.84–1.93 (m, 8H), 2.42–2.49 (m, 12H), 3.99 (t, J = 7.6 Hz, 8H), 5.07 (s, 8H), 5.97 (t, J = 7.0 Hz, 4H), 6.60 (dd, J = 1.7, 7.3 Hz, 4H), 6.98 (dd, J = 1.7, 6.7 Hz, 4H), 7.27–7.43 (m, 20H);

^{13}C NMR (75 MHz, CDCl_3): δ = 23.7, 26.3, 48.2, 50.6, 51.5, 70.5, 104.4, 115.3, 127.2, 127.8, 128.4, 129.4, 136.2, 148.6, 157.9.

Anal. Calcd. for $\text{C}_{63}\text{H}_{70}\text{N}_6\text{O}_8 \cdot 0.5\text{CHCl}_3$, C: 69.40; H: 6.47; N: 7.65. Found, C: 69.33; H: 6.54; N: 7.68.

Chelator 14

Benzyl protected HOPO **13** (0.13 g, 0.13 mmol) was dissolved in 1:1 conc HBr/glacial AcOH (10 mL) and stirred at rt for 4 d. The volatiles were removed under reduced pressure. The residue was washed with EtOAc and ether and dried under high vacuum for 24 h. The product was lyophilized from water/ CH_3CN to yield tetraHOPO chelator **14** (0.11 g, 74%) as a reddish hydrobromide salt.

IR (KBr) 2963, 1640, 1600, 1544 cm^{-1} ;

^1H NMR (D_2O , 300 MHz): δ = 2.07–2.16 (m, 10H), 3.17–3.26 (m, 12H), 4.04 (t, J = 6.7 Hz, 8H), 6.35 (t, J = 7.3 Hz, 4H), 6.96 (dd, J = 1.7, 7.6 Hz, 4H), 7.15 (dd, J = 1.7, 6.7 Hz, 4H);

^{13}C NMR (50 MHz, D_2O): δ = 18.6, 23.3, 46.7, 49.8, 50.0, 109.0, 118.8, 128.9, 145.4, 158.4.

Anal. Calcd. for $\text{C}_{35}\text{H}_{46}\text{N}_6\text{O}_8 \cdot 5.7\text{HBr}$, C: 36.88; H: 4.57; N: 7.37; Found, C: 36.54; H: 4.77; N: 7.76.

Sulfonamide 15

Potassium carbonate (0.60 g, 4.4 mmol) was added to a solution of sulfonamide HOPO **3** (0.78 g, 1.75 mmol) and 1,3-dibromopropane (0.18 g, 0.87 mmol) in anhydrous CH_3CN (20 mL) and the mixture heated at reflux for 20 h. The solvent was removed under reduced pressure and the residue was diluted with CH_2Cl_2 (40 mL) and washed with water (3×20 mL), dried (Na_2SO_4) and the solvent removed in vacuo. The crude product was purified by silica gel chromatography (50–80% EtOAc/hexane) to give dialkylated compound **15** as a yellow solid (0.60 g, 74%) and monoalkylated compound **9** (0.064 g, 6%).

Mp 76–78 $^{\circ}\text{C}$;

IR (KBr) 1654, 1604, 1528 cm^{-1} ;

^1H NMR (400 MHz, CDCl_3): δ = 2.01–2.07 (m, 6H), 3.22–3.26 (m, 8H), 3.98 (t, J = 7.2 Hz, 4H), 5.07 (s, 4H), 6.04 (t, J = 7.2 Hz, 2H), 6.66 (dd, J = 1.6, 7.6 Hz, 2H), 6.94 (dd, J = 1.6, 6.8 Hz, 2H), 7.30–7.41 (m, 10H), 7.97 (d, J = 9.0 Hz, 4H), 8.36 (d, J = 9.0 Hz, 4H);

^{13}C NMR (50 MHz, CDCl_3): δ = 28.2, 28.7, 46.8, 47.6, 70.8, 105.0, 115.3, 124.5, 127.4, 128.1, 128.5, 129.0, 136.0, 144.5, 148.9, 150.1, 158.0;

Anal. Calcd. for $\text{C}_{45}\text{H}_{46}\text{N}_6\text{O}_{12}\text{S}_2$, C: 58.30; H: 5.00; N: 9.07. Found, C: 58.46; H: 5.03; N: 9.28.

Ester 16

Thioacetic acid (0.06 mL, 0.86 mmol) and lithium hydroxide hydrate (76 mg, 1.81 mmol) were added to a solution of disulfonamide **15** (0.20 g, 0.22 mmol) in dimethylformamide (3 mL) and the reaction mixture stirred at rt for 1.5 h. The reaction mixture was then diluted with CH_2Cl_2 (40 mL) and washed with saturated NaHCO_3 (15 mL). The aqueous layer was again extracted with CH_2Cl_2 (30 mL). The combined organic layer was washed with saturated sodium bicarbonate (20 mL), dried (Na_2SO_4) and the solvent removed under reduced pressure. The crude diamine was used without purification. IR (CHCl_3) 3300, 1652, 1602 cm^{-1} ; ^1H NMR (300 MHz, CDCl_3) δ 1.61–1.70 (m, 2H), 1.88–1.98 (m, 4H), 2.59–2.68 (m, 8H), 4.05 (t, J = 7.0 Hz, 4H), 5.11 (s, 4H), 6.00 (t, J = 7.0 Hz), 6.62 (dd, J = 1.4, 7.6 Hz), 6.93 (dd, J = 1.5, 7.0 Hz, 2H), 7.31–7.44 (m, 10H); ^{13}C NMR (75 MHz, CDCl_3) δ 29.3, 30.2, 46.2, 47.3, 48.2, 70.6, 104.5, 115.4, 127.2, 127.8, 128.4, 129.1, 136.3, 148.7, 158.1. Potassium carbonate (0.2 g, 1.45 mmol) and t-butyl bromoacetate (0.17 mL, 1.1 mmol) were added to a solution of the crude diamine (0.16 g, 0.29 mmol) in dry CH_3CN (5 mL) and the mixture stirred at rt for 24 h. The solvent was removed under reduced pressure and the residue was diluted with 30 mL of CH_2Cl_2 and washed with water (10 mL). The aqueous layer was again extracted with CH_2Cl_2 (15 mL) and the combined organic layer was dried (Na_2SO_4) and the solvent removed under reduced pressure. The crude product was purified by silica gel column chromatography (EtOAc) to provide **16** as a viscous yellow oil (0.10 g, 59% from **15**).

IR (neat) 2931, 1731, 1653, 1605, 1554 cm^{-1} ;

^1H NMR (300 MHz, CDCl_3): δ = 1.44 (s, 18H), 1.57–1.65 (m, 2H), 1.87–1.96 (m, 4H), 2.58–2.62 (m, 8H), 3.21 (s, 4H), 4.03 (t, J = 7.0 Hz, 4H), 5.09 (s, 4H), 5.98 (t, J = 7.0 Hz, 2H), 6.62 (dd, J = 1.8, 7.3 Hz, 2H), 7.01 (dd, J = 1.5, 6.7 Hz, 2H), 7.27–7.44 (m, 10H);

^{13}C NMR (75 MHz, CDCl_3): δ = 25.4, 26.7, 28.1, 48.0, 51.2, 52.2, 55.5, 70.7, 80.8, 104.3, 115.5, 127.3, 127.8, 128.4, 129.7, 136.4, 148.8, 158.1, 170.7.

Anal. Calcd. for $\text{C}_{45}\text{H}_{60}\text{N}_4\text{O}_8 \cdot 0.25\text{CHCl}_3$: C, 66.91; H, 7.47; N, 6.90. Found C, 67.11; H, 7.28; N, 6.74.

Chelator 17

Compound **16** (0.28 g, 0.35 mmol) was dissolved in 1:1 conc HBr/glacial AcOH (30 mL) and stirred at rt for 5 d. The volatiles were removed under reduced pressure. The residue was washed with EtOAc and ether and dried under high vacuum for 24 h to yield mixed ligand chelator **16** (0.23 g, 81%) as a light brown hydrobromide salt.

IR (KBr) 3413, 2961, 1741, 1641, 1597, 1547 cm^{-1} ;

^1H NMR (300 MHz, D_2O): δ = 2.12–2.25 (m, 6H), 3.27–3.36 (m, 8H), 4.05–4.10 (m, 8H), 6.37 (t, J = 7.02 Hz, 2H), 6.96 (dd, J = 1.5, 7.6 Hz, 2H), 7.16 (dd, J = 1.5, 6.7 Hz, 2H);

^{13}C NMR (50 MHz, D_2O): δ = 19.1, 23.6, 46.5, 52.0, 52.1, 53.7, 109.0, 119.0, 129.0, 145.3, 158.6, 168.2.

Anal. Calcd. for $\text{C}_{23}\text{H}_{32}\text{N}_4\text{O}_8 \cdot 5\text{HBr} \cdot 2\text{H}_2\text{O}$: C, 29.60; H, 4.43; N, 6.00: Found C, 29.47; H, 4.38; N, 5.88.

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Scheme 1.
Synthesis of HOPO sulfonamide **3**

Scheme 2.
Synthesis of tris-3,2-HOPO chelator **8**

Scheme 3.
Synthesis of tris-3,2-HOPO chelator **12**

Scheme 4.
 Synthesis of tetra-3,2-HOPO chelator **14**

Scheme 5. Synthesis of mixed 3,2-HOPO/iminoacetic acid chelator **17**