Supporting Information for:

Metallopeptoids

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Materials:
Rink Amide resin was supplied by Novabiochem; (S)-(-)-1-phenylethylamine was supplied by TCI America; 8-hydroxy-2-quinolinecarbonitrile, bromoacetic acid, cobalt acetate tetrahydrate and copper acetate monohydrate were supplied by Sigma-Aldrich; N,N'diisopropylcarbodiimide (DIC) was supplied by Chem-Impex International. Other reagents and solvents were obtained from commercial sources and used without additional purification.

Instrumentation:
Peptoid oligomers were analyzed by reversed-phase HPLC (analytical C18 column, Peeke Scientific, 5 μm, 120 Å, 2.0x50 mm) on a Beckman Coulter System Gold 166 instrument. A linear gradient of 5–95% ACN in water (0.1% TFA) over 10 min was used at a flow rate of 700 μL/min. Preparative HPLC was performed using a Delta-Pak C18 column (Waters, 15μm, 100 Å, 25x100mm). Peaks were eluted with a linear gradient of 5–95% ACN in water (0.1% TFA) over 50 min at a flow rate of 5 mL/min. Mass spectrometry was performed on an Agilent 1100 Series LC/MSD Trap XCT (Agilent Technologies) and on Bruker OmniFLEX MALDI-TOF that contains a pulsed ultraviolet nitrogen laser, 200 μJ at 337 nm and a Time-of-Flight mass analyzer with a 120 cm linear flight path. UV measurements were performed using a Lambda 950 double beam double monochromator, and ratio recording UV/VIS/NIR spectrophotometer with a resolution ≤ 0.05 nm. CD measurements were performed using a circular dichroism spectrometer Model 202SF (AVIV Instruments, Inc.).

Molecular modeling:
Geometry optimization was performed with the Universal Force Field in the Accelrys VS software modeling suite.

Preparation of peptoid oligomers:
Peptoid oligomers were synthesized manually on Rink amide resin using the submonomer approach [1]. All peptoid oligomers were synthesized at room temperature. Typically, 100 mg of resin was swollen in DCM for 40 minutes before initiating oligomer synthesis. Multiple washing steps using DMF were performed between each step described below. Bromoacetylation was completed by adding 20 eq bromoacetic acid (1.2 M in DMF, 8.5 mL g^{-1} resin) and 24 eq of diisopropylcarbodiimide (2 mL g^{-1} resin); this reaction was allowed to shake at room temperature for 20 min. Following the reaction, the bromoacetylation reagents were washed from the resin using DMF (10 mL g^{-1} resin) (3 x 1 min) and 20 equivalents of submonomer amine (1.0 M in DMF, 10 mL g^{-1} resin) were added. The amine displacement reaction was allowed to shake at room temperature for 20 min and was followed by multiple washing steps (DMF, 10 mL g^{-1} resin) (3 x 1 min). This two-step addition cycle was modified as follows: after

incorporation of 8-hydroxy-2-quinolinemethylamine, 0.17 ml of a 0.4 M solution of bromoacetic acid, 0.04 ml of neat *N, N*'-diisopropylcarbodiimide (DIC) and 0.29 ml of DMF were added to the resin and mixed at room temperature for 20 minutes. Bromoacetylations and amine displacement steps were repeated until peptoid oligomers of desired sequence were obtained. To cleave the peptoid oligomers from solid support for analysis, approximately 5 mg of resin was treated with 95% TFA in water (40 mL g^{-1} resin) for 10 minutes. The cleavage cocktail was evaporated under nitrogen gas and the peptoid oligomers were re-suspended in 0.5 mL HPLC solvent (1:1 HPLC grade acetonitrile:HPLC grade water).

Preparation of metal complexes:

Complex of $H_1$5 with Co^{2+}. To a solution of **$H_1$5** (11.5 mg, 0.013 mmol) in methanol (0.5 ml), Co^{2+} acetate (1.8 mg, 0.007 mmol) was added and the mixture was stirred for 30 minutes. An orange solid precipitated after the addition of NH_4PF_6 (0.3 ml of a 25 M aqueous solution). The precipitate was filtered, dried and the (**$H_1$5**)$_2$Co complex (8.4 mg) was isolated.

Complex of $H_2$6 with Co^{2+}. To a solution of **$H_2$6** (10.9 mg, 0.01 mmol) in methanol (0.5 ml), Co^{2+} acetate (2.5 mg, 0.01 mmol) was added and the mixture was stirred for 30 minutes. An orange solid precipitated after the addition of NH_4PF_6 (0.2 ml of a 25 M aqueous solution). The precipitate was filtered, dried and the (**$H_2$6**)Co complex (7.6 mg) was isolated.

Complex of $H_1$5 with Cu^{2+}. To a solution of **$H_1$5** (11.5 mg, 0.013 mmol) in methanol (0.5 ml), Cu^{2+} acetate (2.0 mg, 0.007 mmol) was added and the mixture was stirred for 30 minutes. A yellow solid precipitated after the addition of NH_4PF_6 (0.3 ml of a 25 M aqueous solution). The precipitate was filtered, dried and the (**$H_1$5**)$_2$Cu complex (6.7 mg) was isolated.

Complex of $H_2$6 with Cu^{2+}. To a solution of **$H_2$6** (10.9 mg, 0.01 mmol) in methanol (0.5 ml), Cu^{2+} acetate (2.7 mg, 0.01 mmol) was added and the mixture was stirred for 30 minutes. A yellow solid precipitated after the addition of NH_4PF_6 (0.2 ml of a 25 M aqueous solution). The precipitate was filtered, dried and the (**$H_2$6**)Cu complex (7.3 mg) was isolated.

Characterization of peptoid oligomers and their metal complexes:

For characterization, all compounds were collected following HPLC separation and subjected to LC/MS analysis. Peptoid oligomers were characterized by analytical HPLC using a C18 column. The analysis was done using a solvent gradient conducted from 5% to 95% solvent B (0.1% TFA in HPLC grade acetonitrile) over solvent A (0.1% TFA in HPLC grade water) in 10 minutes with a flow rate of 0.7 mL min^{-1}. Additional characterization of peptoid oligomers and a characterization of their metal complexes were conducted by LC/MS (Table S1). The peptoid oligomers were further purified to >95% by RP-HPLC and lyophilized overnight. HPLC traces of pure peptoid oligomers **$H_1$5** and **$H_2$6** are depicted in Figure S2. The HPLC trace of pure peptoid oligomer achiral-**$H_2$6** is depicted in Figure S3.

Structural characterization of peptoid oligomers and their metal complexes:
UV-VIS Spectroscopy. Titration experiments of the peptoid oligomers **H_15** and **H_26** with the metal ions Co^{+2} and Cu^{+2} were followed by UV-VIS measurements. In a typical experiment, 12-20 μL of a peptoid solution (10 mM in 4:1 MeOH:H_2O) was diluted in 3 ml 4:1 MeOH:H_2O solution (to get 40-67.5 μM) and then sequentially titrated with 2-3 μL aliquots of a metal ion (10 mM in H_2O), in multiple steps, until the binding was completed. Raw data were processed for publication using Microsoft Excel and KaleidaGraph.

Binding constants calculations. The association constants for metal binding were measured by titration of 1.4-80 μL aliquots of a metal ion solution (5 mM in H_2O) into a 3 ml solution of the peptoid (typically 14-35μM). Binding was followed by recording the UV-visible spectrum from 200-800 nm as a function of a total added metal ion.

The metal binding to **H_26** can be described by the following equilibria:

$$L + M \xleftrightarrow{K} LM$$

$$K = \frac{[LM]}{[L][M]}$$

Defining $[L] = [L_0] - [LM]$

$$K = \frac{[LM]}{([M] - [LM]) \times ([L_0] - [LM])}$$

Then

$$[LM] = K[LM]^2 - ([L_0] + [M])K[LM] + [L_0]K[M]$$

$$[LM]^2 - \left([L_0] + [M] + \frac{1}{K}\right)[LM] + [L_0][M] = 0$$

And then the real solution to [LM] is

$$1.\ [LM] = \left([L_0] + [M] + \frac{1}{K}\right) \pm \sqrt{\left([L_0] + [M] + \frac{1}{K}\right)^2 - 4[L_0][M]}$$

Now,

$$A = \frac{A_0}{[L_0]}([L_0] - [LM]) + \frac{A_{\max}}{[L_0]} \times [LM]$$

simplified to

2. $A = A_0 + \frac{1}{[L_0]}(A_{\max} - A_0) \times [LM]$

Where A_0 and A_{max} are the minimum and maximum absorbance measured for the free ligand respectively and L_0 is the initial concentration of the free ligand.

Substitution of [LM] in equation 2 with the expression in equation 1, gives

3. $A = A_0 + \frac{1}{[L_0]}(A_{\max} - A_0) \times \left([L_0] + [M] + \frac{1}{K} \right) \pm \sqrt{\left([L_0] + [M] + \frac{1}{K} \right)^2 - 4[L_0][M]}$

Equation 3 was fit using a non linear regression (curve fit) GraphPad Prism® software (Figure S6, S7, S10 and S11).

The metal binding to **$H_1$5** can be described by the following equilibria:

$$2L + M \xleftarrow{K_1} L + ML \xleftarrow{K_2} ML_2$$

$$[M]_t = [M] + [ML] + [ML_2] = [M] + K_1[L][M] + K_1K_2[L]^2[M]$$

4. $[M]_t = [M]\left(1 + \beta_1[L] + \beta_2[L]^2\right)$

Where $\beta_1 = K_1$ and $\beta_2 = K_1K_2$ and $[M]_t$ = total concentration of the metal ion.

Then,

$$[L]_t = [L] + [ML] + [ML_2] = [L] + K_1[L][M] + 2K_1K_2[L]^2[M]$$

where $[L]_t$ is the total concentration of the free ligand

5. $[L]_t = [L] + [M]\left(\beta_1[L] + 2\beta_2[L]^2\right)$

Substitution of [M] from 4 into 5 gives

$$[L]_t = [L] + \frac{[M]_t\left(\beta_1[L] + 2\beta_2[L]^2\right)}{\left(1 + \beta_1[L] + \beta_2[L]^2\right)}$$

The fraction of the bound ligand can be defined as

6. $f_B = \frac{[L]_t - [L]}{[L]_t}$

and since

7. $[L]_t - [L] = \frac{[M]_t\left(\beta_1[L] + 2\beta_2[L]^2\right)}{\left(1 + \beta_1[L] + \beta_2[L]^2\right)}$

Substitution of equation 7 into 6 gives

$$f_B = \frac{[M]_t}{[L]_t}\frac{[M]_t\left(\beta_1[L] + 2\beta_2[L]^2\right)}{\left(1 + \beta_1[L] + \beta_2[L]^2\right)}$$

Also $f_B = 1 - \frac{A}{A_{max}}$ so

8. $1 - \frac{A}{A_{max}} = \frac{[M]_t}{[L]_t}\frac{[M]_t\left(\beta_1[L] + 2\beta_2[L]^2\right)}{\left(1 + \beta_1[L] + \beta_2[L]^2\right)}$

Now, since $[L] = [L]_0 - [M]$ we get

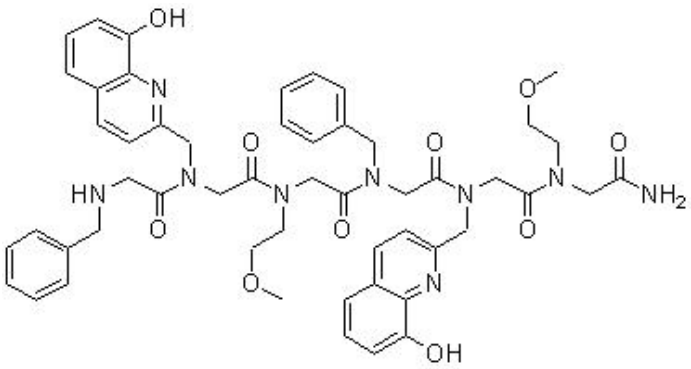
9. $\frac{A}{A_{max}} = 1 - \frac{[M]_t}{[L]_t}\frac{[M]_t\left(\beta_1([L]_0 - [M]) + 2\beta_2([L]_0 - [M])^2\right)}{\left(1 + \beta_1([L]_0 - [M]) + \beta_2([L]_0 - [M])^2\right)}$

Equation 9 was fit using a non linear regression (curve fit) GraphPad Prism® software (Figures S8 and S9).

Circular Dichroism (CD) Spectroscopy. Approximately 200 µL solutions (10 mM in 4:1 MeOH:H_2O) of lyophilized peptoid powders were prepared immediately before CD measurements. The typical concentration of each measured sample (free ligand peptoids and metal-peptoid complexes) was 100 µM. CD scans were performed at 25˚C. Spectra were obtained by averaging 8 scans per sample in a fused quartz cell (Starna, Inc.) (path length=0.1 cm). Scans were performed over the 360 to 180 nm region at a step of 0.5 or 1nm (scan rate=1 sec/step). Raw data were processed for publication using Microsoft Excel and KaleidaGraph.

References

1. Zuckermann, R. N.; Kerr J. M.; Kent S. B. W.; Moos W. H. *J. Am. Chem. Soc.* 1992, **114**, 10646-10647.



Scheme S1. Achiral-**$H_2$6**.

Peptoid and Peptoid -Metal complex	Molecular weight	Abs. Maxima
	Calc: Found	Wavelength (nm)
*N*spe-*N*spe-*N*spe-*N*spe-*N*hq ($H_1$5*)*	876.1: 876.5	245, 307
$(H_1 5)_2$Co	1809.1: 1809.0	262, 365
$(H_1 5)_2$Cu	1813.5: 1815.4	260, 366
*N*spe-*N*hq-*N*spe-*N*spe-*N*hq-*N*spe ($H_2$6)	1090.3: 1090.6	246, 308
($H_2$6)Co	1147.2: 1180.1(M^+ + MeOH)	266, 375
($H_2$6)Cu	1153.6: 1152.9	263, 384
*N*pm-*N*hq-*N*sme-*N*pm-*N*hq-*N*me (achiral-$H_2$6)	970.08: 970.5	248, 310

Table S1. Peptoid oligomer sequences and their isolated metal complexes. Compounds **$H_1$5, $H_2$6** and achiral-**$H_2$6** were subsequently purified by preparative HPLC to >95%. *N*spe = S)-(-)-1-phenylethylamine, *N*hq = 8-hydroxy-2-quinolinemethylamine, *N*pm = N-benzylglycine and *N*me = N-methoxyethylglycine.

Compound	First maximum		Second maximum	
	λ, nm	ε, $M^{-1}cm^{-1}$	λ, nm	ε, $M^{-1}cm^{-1}$
*N*spe-*N*spe-*N*spe-*N*spe-*N*hq (H_15)	245	31,500 ± 100	307	10000 ± 100
$(H_15)_2Co$	262	18,500 ± 100	365	1700 ± 10
$(H_15)_2Cu$	260	25,500 ± 200	366	8500 ± 70
*N*spe-*N*hq-*N*spe-*N*spe-*N*hq-*N*spe (H_26)	246	40,500 ± 100	308	5700 ± 10
(H_26)Co	266	31,000 ± 400	375	4100 ± 40
(H_26)Cu	263	32,000 ± 100	384	4700 ± 60
Npm-Nhq-Nsme-Npm-Nhq-Nme (achiral-H_26)	246	40,500 ± 100	308	5700 ± 10
(achiral-H_26)Co	264	27500 ± 200	383	2100 ± 50
(achiral-H_26)Cu	266	28,500 ± 200	388	1750 ± 10

Table S2. UV-Vis absorbance data of peptoid oligomer sequences and their metal complexes. *N*spe = S)-(-)-1-phenylethylamine, *N*hq = 8-hydroxy-2 quinolinemethylamine.

Compound	K_1 (M^{-1})	K_2 (M^{-1})	β_2 (M^{-2})
$(H_15)_2Co$	$7.4x10^5$	$1.3x10^4$	$9.45x10^9$
$(H_15)_2Cu$	$3.75x10^5$	$1.45x10^4$	$5.45x10^9$
(H_26)Co	$7.1x10^{14}$		
(H_26)Cu	$8.5x10^{14}$		
(achiral-H_26)Co	$6.4x10^{11}$		
(achiral-H_26)Cu	$2.5x10^{14}$		

Table S3. Association constants for peptoid-metal complexes. Binding was followed by recording the UV-visible spectrum of the peptoid ligand, at typical concentrations of 23-35μM, as a function of a total added metal ion. The association constants were obtained from a curve fitting using a non linear regression GraphPad Prism® software (Figures S6-S9).

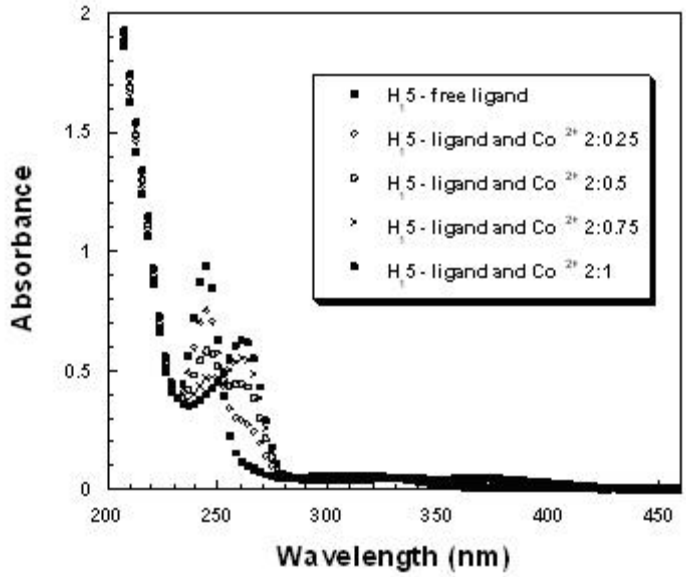


Figure S1. UV-VIS spectra of $\mathbf{H_15}$ and the formation of $\mathbf{(H_15)_2Co}$ complex. The spectra were recorded at room temperature, in a MeOH:H_2O (4:1) solution. Initial concentration of $\mathbf{H_15}$ was 54μM.

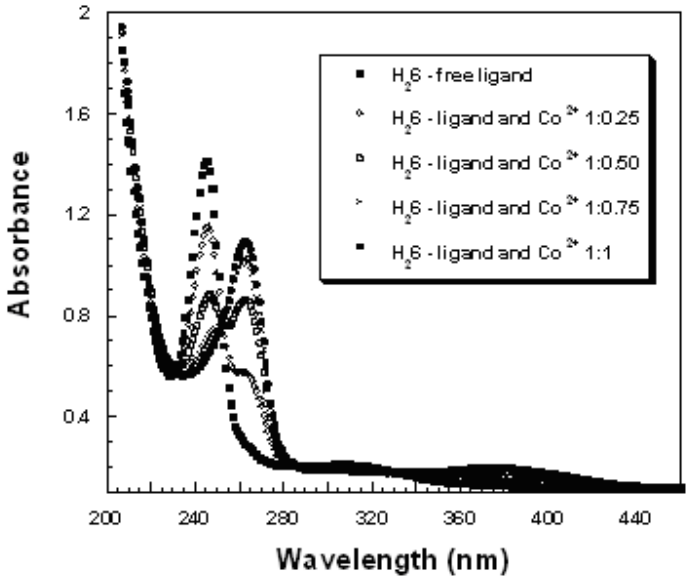


Figure S2. UV-VIS spectra of $\mathbf{H_26}$ and the formation of $\mathbf{(H_26)Co}$ complex. The spectra were recorded at room temperature, in a MeOH:H_2O (4:1) solution. Initial concentration of $\mathbf{H_26}$ was 40μM.

HPLC data

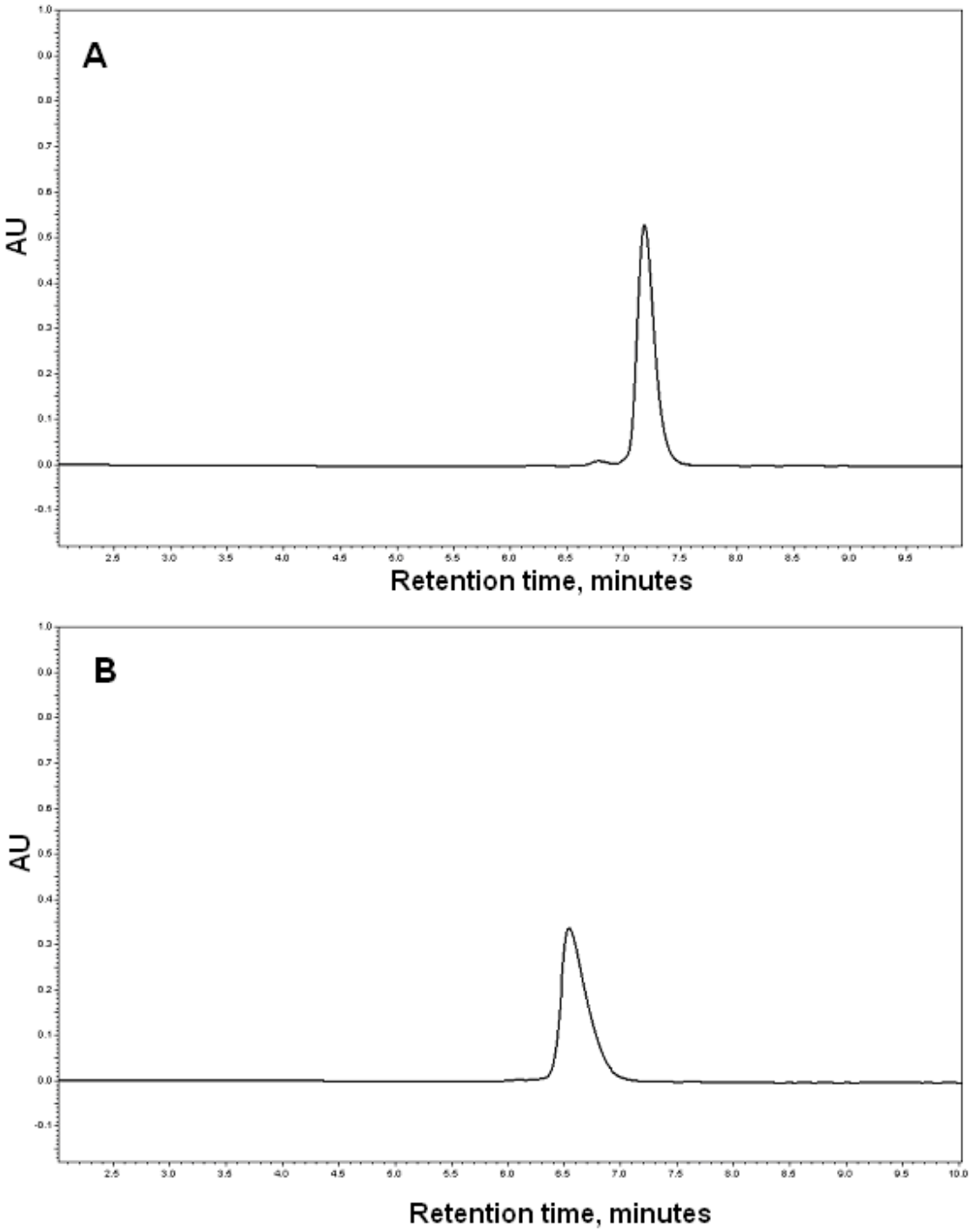


Figure S3. HPLC traces of purified peptoid oligomers $\mathbf{H_1 5}$ (A) and $\mathbf{H_2 6}$ (B) at 214 nm.

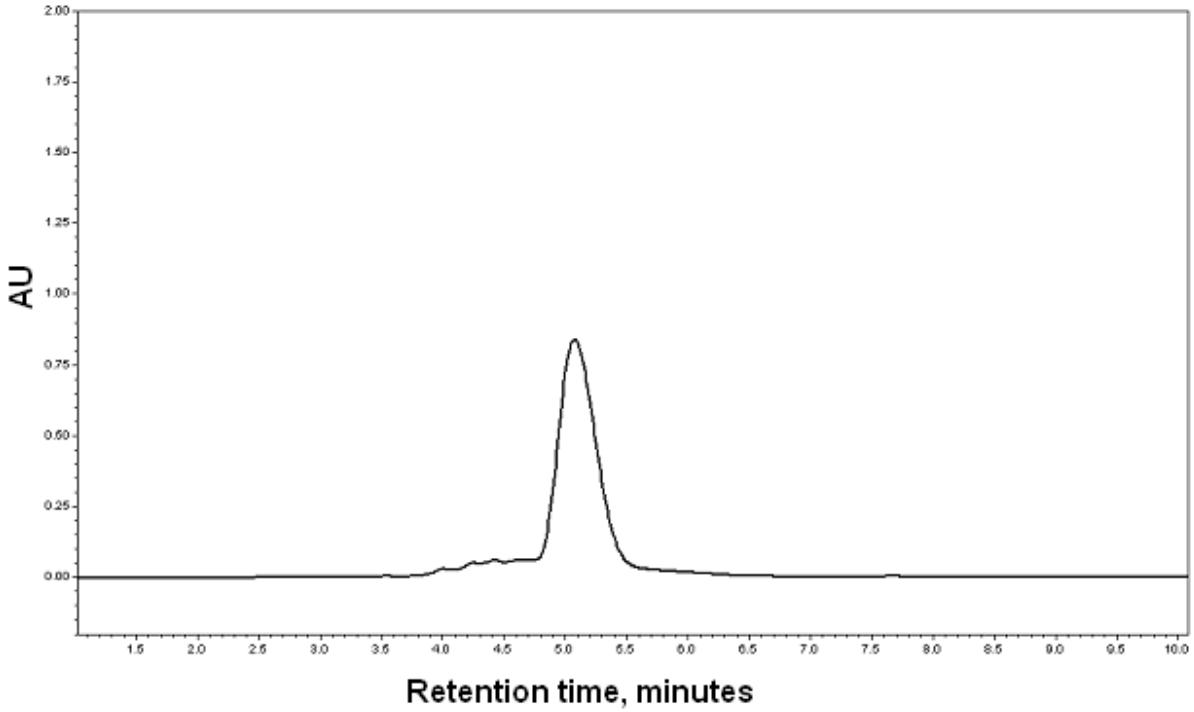


Figure S4. HPLC traces of purified peptoid oligomer achiral-$\mathbf{H_2}$**6** at 214nm.

CD data

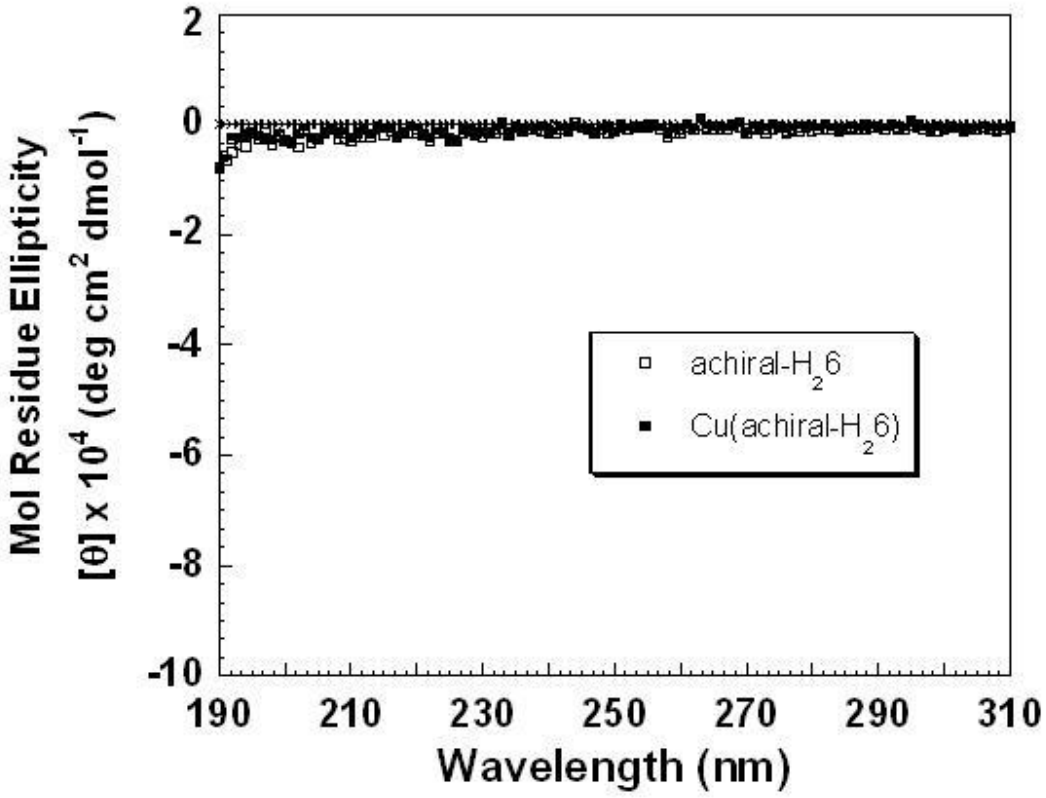


Figure S5. CD spectra of achiral-$H_2$6 and its Cu complex. The spectra were recorded at room temperature, in a MeOH/H_2O (4:1) solution and **$H_2$6** concentration of 100μM.

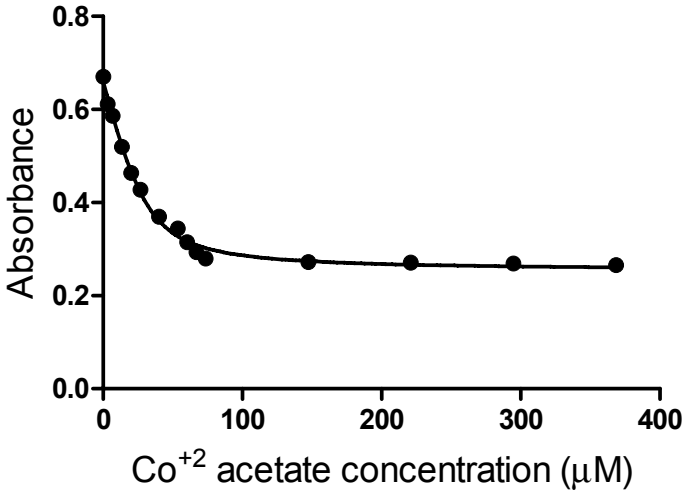


Figure S6. Association curve (dots) and fit (line) for the formation of ($H_2$6)Co complex. The fitting was done using a nonlinear regression (curve fit) GraphPad Prism® software. The regression coefficient factor for this fit is 0.995.

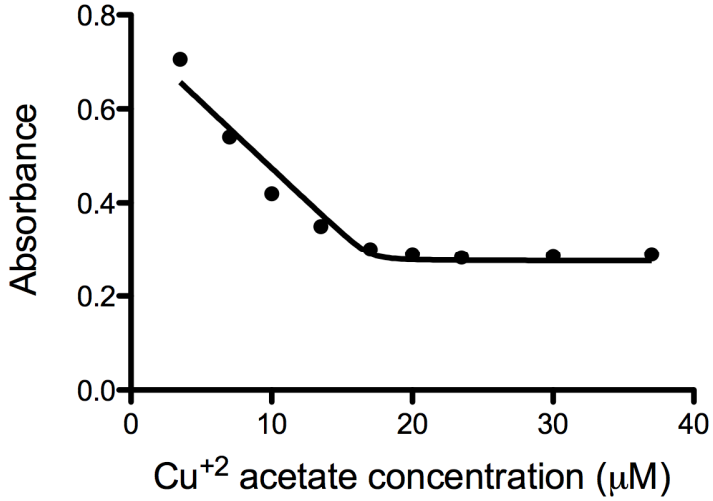


Figure S7. Association curve (dots) and fit (line) for the formation of ($H_2$6)Cu complex. The fitting was done using a nonlinear regression (curve fit) GraphPad Prism® software. The regression coefficient factor for this fit is 0.965.

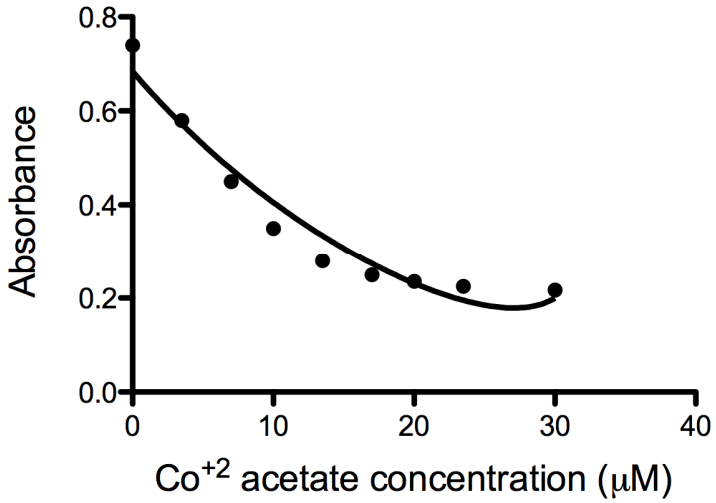


Figure S8. Association curve (dots) and fit (line) for the formation of $(H_15)_2Co$ complex. The fitting was done using a nonlinear regression (curve fit) GraphPad Prism® software. The regression coefficient factor for this fit is 0.927.

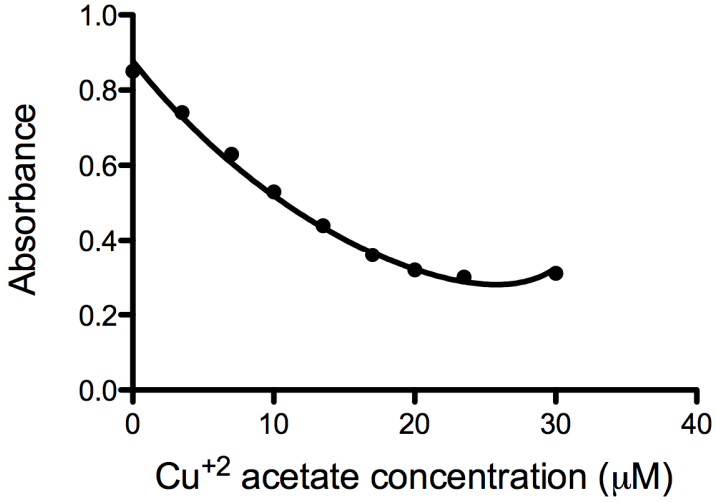


Figure S9. Association curve (dots) and fit (line) for the formation of $(H_15)_2Cu$ complex. The fitting was done using a nonlinear regression (curve fit) GraphPad Prism® software. The regression coefficient factor for this fit is 0.994.

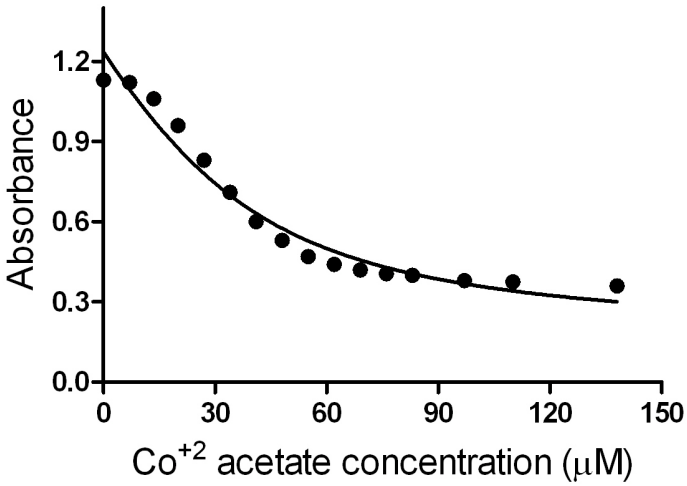


Figure S10. Association curve (dots) and fit (line) for the formation of (achiral-$H_2$6)Co complex. The fitting was done using a nonlinear regression (curve fit) GraphPad Prism® software. The regression coefficient factor for this fit is 0.965.

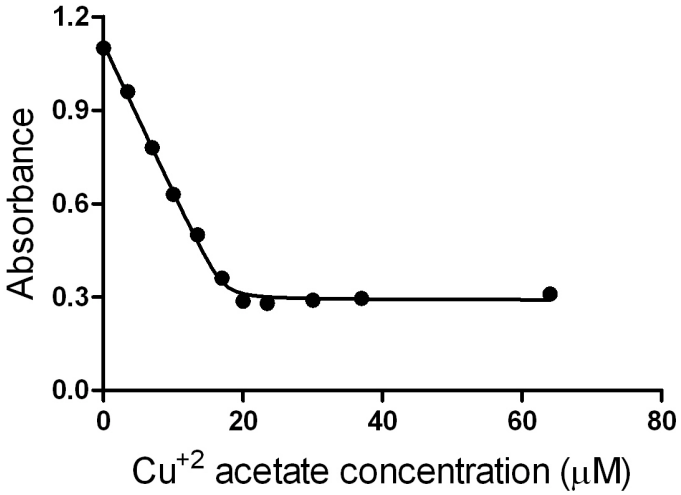


Figure S11. Association curve (dots) and fit (line) for the formation of (achiral-$H_2$6)Cu complex. The fitting was done using a nonlinear regression (curve fit) GraphPad Prism® software. The regression coefficient factor for this fit is 0.997.