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Supplementary Information

For

Supramolecular electrochemical probe based on the system of tetrazole derivative pillar[5]arene/methylene blue

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1. NMR, MALDI TOF MS, IR spectra of the compounds 2, 3, 4, 6, 7, 8 Figure S01. ¹H NMR spectrum of 1,4-bis(2-thiocyanoethoxy)benzene (2), DMSO-d₆, 298 K, 400 MHz.



Figure S02. ¹³C NMR spectrum of 1,4-bis(2-thiocyanoethoxy)benzene (2), DMSO-d₆, 298 K, 100 MHz.





Figure S04. IR spectrum of 1,4-bis(2-thiocyanoethoxy)benzene (2).



Figure S05. ¹H NMR spectrum of 4, 8, 14, 18, 23, 26, 28, 31, 32, 35-deca(2-thiocyanoethoxy)pillar[5]arene (6), DMSO- d₆, 298 K, 400 MHz



Figure S06. ¹³C NMR spectrum of 4, 8, 14, 18, 23, 26, 28, 31, 32, 35-deca(2-thiocyanoethoxy)pillar[5]arene (6), DMSO-d₆, 298 K, 100 MHz.



Figure S07. IR spectrum of 4, 8, 14, 18, 23, 26, 28, 31, 32, 35-deca(2-thiocyanoethoxy)pillar[5]arene (6).



Figure S08. Mass spectrum (MALDI-TOF, 4-nitroaniline matrix) of 4, 8, 14, 18, 23, 26, 28, 31, 32, 35-deca(2-thiocyanoethoxy)pillar[5]arene (6).



Figure S09. ¹H NMR spectrum of 1,4-bis(2-((1H-tetrazol-5-yl)thio)ethoxy)benzene (3), DMSOd₆, 298 K, 400 MHz



Figure S10. ¹³C NMR spectrum of 1,4-bis(2-((1H-tetrazol-5-yl)thio)ethoxy)benzene (3), DMSO-d₆, 298 K, 100 MHz.







Figure S12. IR spectrum of 1,4-bis(2-((1H-tetrazol-5-yl)thio)ethoxy)benzene (3)



Figure S13. ¹H NMR spectrum of 4, 8, 14, 18, 23, 26, 28, 31, 32, 35-deca(2-((1H-tetrazol-5-yl)thio)ethoxy)pillar[5]arene (7), DMSO-d₆, 298 K, 400 MHz.



Figure S14. ¹³C NMR spectrum of 4, 8, 14, 18, 23, 26, 28, 31, 32, 35-deca(2-((1H-tetrazol-5-yl)thio)ethoxy)pillar[5]arene (7), DMSO-d6, 298 K, 100 MHz.



Figure S15. Mass spectrum (MALDI-TOF, 4-nitroaniline matrix) of 4, 8, 14, 18, 23, 26, 28, 31, 32, 35-deca(2-((1H-tetrazol-5-yl)thio)ethoxy)pillar[5]arene (7).



Figure S16. IR spectrum of 4, 8, 14, 18, 23, 26, 28, 31, 32, 35-deca(2-((1H-tetrazol-5-yl)thio)ethoxy)pillar[5]arene (7).



Figure S17. ¹H NMR spectrum of Ammonium salt of compound 4, D₂O, 298 K, 400 MHz.



Figure S18. ¹³C NMR spectrum of Ammonium salt of compound (4), D₂O, 298 K, 100 MHz





Figure S19. Mass spectrum (ESI) of Ammonium salt of compound (4).

Figure S20. IR spectrum of Ammonium salt of compound (4).



Figure S21. ¹H NMR spectrum of Ammonium salt of compound (8), D₂O, 298 K, 400 MHz.



Figure S22. ¹³C NMR spectrum of Ammonium salt of compound (8), D₂O, 298 K, 100 MHz.





Figure S23. Mass spectrum (ESI) of Ammonium salt of compound (8).

Figure S24. IR spectrum of Ammonium salt of compound (8).



2. Thermal Gravimetric Analysis of 3, 4 and 7, 8

Figure S25. TGA (green) and differential scanning calorimetry (DSC) (blue) curves of 3.



Figure S26. TGA (green) and differential scanning calorimetry (DSC) (blue) curves of 4.





Figure S27. TGA (green) and differential scanning calorimetry (DSC) (blue) curves of 7.

Figure S28. TGA (green) and differential scanning calorimetry (DSC) (blue) curves of 8.



3. UV spectra and Bindfit (Fit data to 1:1, 1:2 and 2:1 Host-Guest equilibria)

Figure S29. Bindfit (Fit data to 1:1, 1:2 and 2:1 Host-Guest equilibria) Screenshots taken from the summary window of the **website supramolecular.org**. This screenshots shows the raw data for UV-vis titration of MB / **8** in water, the data fitted to 1:1 binding model (a), 1:2 binding model (b) and 2:1 binding model (c).

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(c)	. ,	Details Time to fit SSR Fitted datapoints Fitted params	0.2840 s 0.3945 252 22			84 835 83					0.226 0.226 0.241 0.236
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(b) $\frac{K(0-\pi)}{M^{2}} = \frac{393555}{5} = \frac{443772}{M^{2}} = \frac{1000}{M^{2}}$ Back Net $\frac{14235}{5} = \frac{14235}{5} = \frac$		Parameter (bounds)	Optimised	Error	Initial	0.2 1 2		6 7 Equivalent tot	a 9 tó 1 al[G]∞(H]o	1 12 13 14	15
Back Net Back Net Piter UV 12 Pit Summary Swe Details Net Prameters Net Back Net		$\mathbf{K} (0 \to \infty)$	393455.69 M ⁻¹	± 49.7772 %	100.00 M ⁻¹	0.15 0.1 0.05 0					0.286 residuals 0.284 residuals 0.241 residuals 0.236 residuals
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Providence Laboration Provid		Back		Next		-0.051	2 3 4	5 8	7 8 9	10 11 12 13	14 15

Figure S30. Bindfit (Fit data to 1:1, 1:2 and 2:1 Host-Guest equilibria) Screenshots taken from the summary window of the **website supramolecular.org**. This screenshots shows the raw data for UV-vis titration of MB / 7 in methanol, the data fitted to 1:1 binding model (a), 1:2 binding model (b) and 2:1 binding model (c).



Figure S31. Bindfit (Fit data to 1:1, 1:2 and 2:1 Host-Guest equilibria) Screenshots taken from the summary window of the **website supramolecular.org**. This screenshots shows the raw data for UV-vis titration of $7/Zn^{2+}$ in methanol, the data fitted to 1:1 binding model (a), 1:2 binding model (b) and 2:1 binding model (c).



Figure S32. Bindfit (Fit data to 1:1, 1:2 and 2:1 Host-Guest equilibria) Screenshots taken from the summary window of the **website supramolecular.org**. This screenshots shows the raw data for UV-vis titration of $7 / \text{Co}^{2+}$ in methanol, the data fitted to 1:1 binding model (a), 1:2 binding model (b) and 2:1 binding model (c).



Figure S33. Bindfit (Fit data to 1:1, 1:2 and 2:1 Host-Guest equilibria) Screenshots taken from the summary window of the **website supramolecular.org**. This screenshots shows the raw data for fluorescence titration of **8** / Co^{2+} in water, the data fitted to 1:1 binding model (a), 1:2 binding model (b) and 2:1 binding model (c).



Figure S34. Bindfit (Fit data to 1:1, 1:2 and 2:1 Host-Guest equilibria) Screenshots taken from the summary window of the **website supramolecular.org**. This screenshots shows the raw data for fluorescence titration of **8** / Zn^{2+} in water, the data fitted to 1:1 binding model (a), 1:2 binding model (b) and 2:1 binding model (c).



4. Dynamic light scattering.

Table S1. DLS study on the aggregation for 7 with Zn^{2+} and Co^{2+} in the methanol

Ratio	С7, М	C Zn2+, M Z average (d) , nm		PDI
7/Zn ²⁺	(CH ₃ OH)	(CH ₃ OH)		
1:0	1×10-3	0	1216±100	$0.42{\pm}0.22$
1:0	1×10 ⁻⁴	0	916±42	0.64±0.12
1:0	1×10-5	0	1015±131	0.54±0.11
<mark>1:1</mark>	1×10 ⁻³	1×10 ⁻³	<mark>981.6±10</mark>	0.22 ± 0.09
1:1	1×10-4	1×10 ⁻⁴	842±32	0.39±0.12
1:1	1×10-5	1×10 ⁻⁵	798±25	$0.44{\pm}0.20$
1:2	1×10-3	2×10-3	-	0.9
1:2	1×10-4	2×10-4	531±15	0.32±0.09
1:2	1×10-5	2×10-5	512±32	0.49±0.19
2:1	2×10-3	1×10-3	1411±281	0.34±0.09
2:1	2×10-4	1×10 ⁻⁴	1215±171	0.51±0.21
2:1	2×10 ⁻⁵	1×10 ⁻⁵	1221±315	0.54±0.19
7/Co ²⁺	С7, М	С со2+, М	Z average (d) , nm	PDI
<mark>1:1</mark>	1×10 ⁻³	1×10 ⁻³	231 ± 2	0.10 ± 0.01
1:1	1×10-4	1×10 ⁻⁴	341±17	0.36±0.09
1:1	1×10-5	1×10-5	389±19	0.41±0.12
1:2	1×10-3	2×10-3	412±21	0.41±0.11
1:2	1×10-4	2×10-4	473±22	0.44±0.19
1:2	1×10-5	2×10-5	477±34	0.49±0.23
2:1	2×10-3	1×10-3	421±71	0.63±0.35
2:1	2×10-4	1×10 ⁻⁴	573±105	0.9
2:1	2×10-5	1×10-5	-	-

 Table S2. DLS study on the aggregation for 7 / MB in MeOH.

Ratio 7/MB	С7, М	С мв, М	Z average (d) , nm	PDI	ζ- potential, mV
1:0	1×10-5	0	388±66	0.31±0.01	
2:1	2×10 ⁻⁵	1×10 ⁻⁵	340±24	$0.39{\pm}0.07$	-
1:1	1×10-5	1×10 ⁻⁵	174±33	0.27±0.15	-
1:2	1×10-5	2×10 ⁻⁵	167±44	0.23 ± 0.02	-
0:1	0	10-5	-	-	-
1:0	1×10 ⁻⁴	0	524±96	0.53±0.25	
2:1	2×10 ⁻⁴	1×10 ⁻⁴	226±34	0.42±0.13	
1:1	1×10 ⁻⁴	1×10 ⁻⁴	226±46	0.43±0.17	
<mark>1:2</mark>	1×10^{-4}	2×10^{-4}	142±5	0.21 ± 0.04	18.42 ± 0.04
0:1	0	1×10 ⁻⁴	-	-	-
1:0	1×10-3	0	216±18	0.62 ± 0.23	
2:1	2×10-3	1×10-3	-	-	-
1:1	1×10 ⁻³	1×10 ⁻³	816±37	$0.52{\pm}0.10$	-
1:2	1×10-3	2×10 ⁻³	-	-	-
0:1	0	1×10 ⁻³	-	-	-

		66 6						
Ratio 8/MB	C ₈ , M	С мв, М	Z average (d) , nm	PDI	ζ- potential, mV			
1:0	1×10 ⁻⁵	0	87±3	$0.39{\pm}0.05$				
2:1	2×10 ⁻⁵	1×10 ⁻⁵	157.7±18.89	0.36±0.06	-			
1:1	1×10-5	1×10 ⁻⁵	133.0±16.92	0.43±0.10	-			
1:2	1×10-5	2×10-5	176.0±40.71	0.41 ± 0.09	-			
0:1	0	1×10-5	-	-	-			
1:0	1×10-4	0	107±26	0.51±0.10				
2:1	2×10 ⁻⁴	1×10 ⁻⁴	98±3	0.42 ± 0.06	-			
1:1	1×10 ⁻⁴	1×10 ⁻⁴	80±14	0.41±0.07	-			
1:2	1×10 ⁻⁴	2×10^{-4}	72±1	0.30 ± 0.03	21.10 ± 0.03			
0:1	0	1×10 ⁻⁴	-	-	-			
1:0	1×10-3	0	87±2	0.38 ± 0.04				
2:1	2×10-3	1×10 ⁻³	118±5	0.36±0.10	-			
1:1	1×10 ⁻³	1×10 ⁻³	108±4	0.79±0.24	-			
1:2	1×10 ⁻³	2×10 ⁻³	-	-	-			
0:1	0	1×10-3	-	-	-			

Table S3. Aggregation of the particles for 8 / MB in H_2O .

Figure S35. Size distribution of the particles by intensity for 8 (1×10^{-4} M) / MB (2×10^{-4} M) in H₂O







6. Electrochemistry





Figure S 38. Cyclic voltammograms recorded on GCE in the solutions of MB (0.2 mM), compounds **3** and **4** (0.1 mM) and metal salts (0.1 mM), HEPES buffer, pH 7.0, 50 mV/s.



7. Transmission electron microscopy

Figure S39. TEM images of self-associates 7 (1×10⁻⁴M)/MB (2×10⁻⁴M)



Figure S40. TEM images of self-associates 7 (1×10⁻⁴M)/MB (2×10⁻⁴M)





Figure S41. TEM images of self-associates 8 (1×10⁻⁴M)/MB (2×10⁻⁴M)

Figure S42. TEM images of self-associates 8 (1×10⁻⁴M)/MB (2×10⁻⁴M)



File name=LM212TET-N+MB_006.tif Image date=2023/11/1 08:15:19 Image number=7307 Calibration=1 974nm/pixel at x10.0k Magnification=x100k Lens mode=2com-1 HC-1 Camera name=XR81-DIR

Spot number=1 Image rotation=0° Acc. voltage=100.0kV Emission=15.0µA Stage X=-701 Y=-294 Tilt=-0.1 Azim=0.0 Camera size=3296x2464pixel

200nm