

Electronic supplementary information

for

Mechanistic insights into the heavy metal ion sensing by NOS₂- macrocyclic fluorosensors via the structure-function relationship: influences of fluorophores, solvents and anions

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Experimental

General. All chemicals and solvents employed in the syntheses were of reagent grade and were used without further purification. NMR spectra were recorded on a Bruker DRX 300 spectrometer. FT-IR spectra were measured with a ThermoFisher Scientific Nicolet iS 10 FT-IR spectrometer. The solid-state excitation and emission spectra were performed on a RF-5301 spectrophotometer. The ESI-mass spectrum was obtained on a Thermo Scientific LCQ Fleet spectrometer. CSI mass spectrum was measured on a JEOL AccuTOF (JMS-T100CS) mass spectrometer with ESI ion source. The elemental analysis was performed on a Thermo-Fisher Scientific Flash 2000 elemental analyser.

Preparation of L¹. The precursor **6** (0.95 g, 3.35 mmol) was added dropwise to a stirred suspension of AlCl₃ (0.51 g, 3.82 mmol) in anhydrous chloroform (50 mL) under nitrogen at the 0 °C. After addition of a 9-(chloromethyl)anthracene (0.85 g, 3.75 mmol), the reaction mixture was refluxed for a further 12 h. After cooling to room temperature, water was added and then the solvent was evaporated. The residue was extracted with dichloromethane and the combined organic phases were dried with anhydrous sodium sulfate and then evaporated to dryness. The flash column chromatography (SiO₂; 5 % ethyl acetate: *n*-hexane) afforded the product **L¹** as a pale-yellow solid in 20% yield. Mp: 192-193 °C. ¹H NMR (300 MHz, CDCl₃): δ 8.42 (*s*, 1 H, anthracene), 8.28-8.25 (*m*, 2 H, anthracene), 8.02-8.05 (*m*, 2 H, anthracene), 7.44-7.50 (*m*, 4 H, anthracene), 6.99 (*d*, 2 H, Ar), 6.52 (*d*, 2 H, Ar), 4.92 (*s*, 4 H, ArCH₂Ar), 3.74 (*t*, 4 H, OCH₂CH₂S), 3.68 (*t*, 4 H, SCH₂CH₂N), 2.85 (*t*, 4 H, OCH₂CH₂S), 2.79 (*t*, 4 H, SCH₂CH₂N). ¹³C NMR (75 MHz, CDCl₃): 146.0, 132.5, 131.7, 130.5, 129.1, 129.1, 128.2, 126.3, 125.8, 125.1, 124.9, 111.3, 73.6, 50.3, 32.4, 32.2, 30.7. IR (KBr pellet): 3046, 2916, 2851, 2787, 1611, 1562, 1519, 1400, 1373, 1354, 1287, 1209, 1185, 1158, 1115, 1019, 919, 872, 824, 727 cm⁻¹. Anal. calcd. for [C₂₉H₃₁NOS₂]: C, 73.53; H, 6.60; N, 2.96; S, 13.54. Found: C, 73.33; H, 6.56; N, 2.89; S, 13.27%. ESI-Mass spectrum: *m/z* = 474.07 [C₂₉H₃₂NOS₂]⁺.

Preparation of L². The mixed ethanol solution (60 mL) of compound **8** (1.0 g, 3.21 mmol), 2-aminobenzenethiol (0.55 mg, 4.39 mmol) and a drop of acetic acid was refluxed with stirring for 4h. After the reaction, the mixture was first concentrated, and 50 mL of water was added. The solution was extracted with chloroform. The extract was dried over NaSO₄ and solvent was evaporated in vacuo. The residue was purified with column chromatography on silica gel using 20% dichloromethane/*n*-hexane led to the isolation of **L²** as a yellow crystalline product in a 63% yield. Mp: 157-158 °C (decomp.). ¹H NMR (300 MHz, CDCl₃): δ 7.9 (*m*, 2H, Ar), 7.8 (*d*, 1H, Ar), 7.4 (*t*,

1H, Ar), 7.3 (t, 1H, Ar), 6.7 (d, 2H, Ar), 3.8 (t, 4H, OCH₂CH₂S), 3.7 (t, 4H, SCH₂CH₂N), 2.9 (t, 4H, OCH₂CH₂S), 2.8 (t, 4H, SCH₂CH₂N). ¹³C NMR (75 MHz, CDCl₃): 168.3, 154.5, 149.1, 135.4, 129.8, 128.0, 126.6, 124.8, 123.3, 112.0, 73.9, 51.5, 33.4, 31.4. IR (KBr pellet): 3447, 2922, 2844, 2367, 2343, 1604, 1485, 1437, 1185, 1111, 762 cm⁻¹. Anal. calcd. for [C₂₁H₂₄N₂OS₃]: C, 60.54.; H, 5.81; N, 6.72; S, 23.09. Found: C, 60.93; H, 6.12; N, 6.47; S, 22.90. ESI-Mass spectrum: *m/z* = 417.10 [C₂₁H₂₅N₂OS₃]⁺.

CAUTION! Perchlorate salts of metal complexes are potentially explosive and should be handled with great care.

Preparation of [Ag₆(L¹)₆](ClO₄)₆ (1). AgClO₄ (4.36 mg, 21.6 mmol) in methanol (2 mL) was added to a solution of L¹ (10.2 mg, 21.5 mmol) in chloroform. Slow evaporation of the solution afforded the colourless crystalline product **1** suitable for X-ray analysis. IR (KBr pellet): 3045, 2911, 2829, 2799, 1611, 1566, 1503, 1416, 1351, 1356, 1184, 1138, 1119, 1045, 921, 881, 829, 757 cm⁻¹. Anal. calcd. for [C₂₉H₃₁Ag₁Cl₁N₁O₅S₂]: C, 51.15; H, 4.59; N, 2.06; S, 9.42. Found: C, 51.21; H, 4.60; N, 2.22; S, 9.19.

Preparation of [Ag₂(L²)₂](PF₆)₂·2CH₂Cl₂ (2). AgPF₆ (7.28 mg, 28.8 mmol) in methanol (2 mL) was added to a solution of L (10.0 mg, 24.0 mmol) in dichloromethane. Slow evaporation of the solution afforded the colourless crystalline product **2** suitable for X-ray analysis. Mp 193-194 °C (decomp.). IR (KBr pellet): 3430, 3001, 2902, 2814, 1662, 1485, 1456, 1185, 1105, 1085, 1042, 841, 775 cm⁻¹. Anal. calcd. for [C₂₁H₂₄Ag₁P₁F₆N₂OS₃]: C, 37.68; H, 3.61; N, 14.37; S, 14.37. Found: C, 37.84; H, 3.72; N, 4.30; S, 14.49.

Preparation of {[Hg(L²)(acetone)](ClO₄)₂}_n (3). Reaction of L (10.0 mg, 24.0 mmol) with Hg(ClO₄)₂ (11.5 mg, 28.8 mmol) in dichloromethane/acetone afforded colourless precipitate. The vapour diffusion of diethyl ether to acetonitrile solution gave to crystalline product **3**. IR (KBr pellet): 3421, 2931, 1601, 1514, 1452, 1339, 1244, 1132, 1117, 1105, 955, 858, 756 cm⁻¹. Anal. calcd. for [C₂₄H₃₀Cl₂Hg₁N₂O₁₀S₃]: C, 32.98; H, 3.46; N, 3.20; S, 11.00. Found: C, 33.16; H, 3.42; N, 3.37; S, 11.19.

Preparation of [Hg(L²)I₂]_n (4). HgI₂ (25.1 mg, 0.063 mmol) in methanol (2 mL) was added to a solution of L² (20.0 mg, 0.021 mmol) in dichloromethane (2 mL). Slow evaporation of the solution afforded the colourless crystalline product **4** suitable for X-ray analysis. Mp 196-197 °C (decomp.). IR (KBr pellet): 3445, 3003, 2978, 2825, 2248, 1602, 1522, 1454, 1336, 1245, 858, 756

4

cm⁻¹. Anal. calcd. for [C₂₁H₂₄HgI₂N₂OS₃]: C, 28.96; H, 2.78; N, 3.22; S, 11.04. Found: C, 29.21; H, 2.92; N, 4.30; S, 10.88.

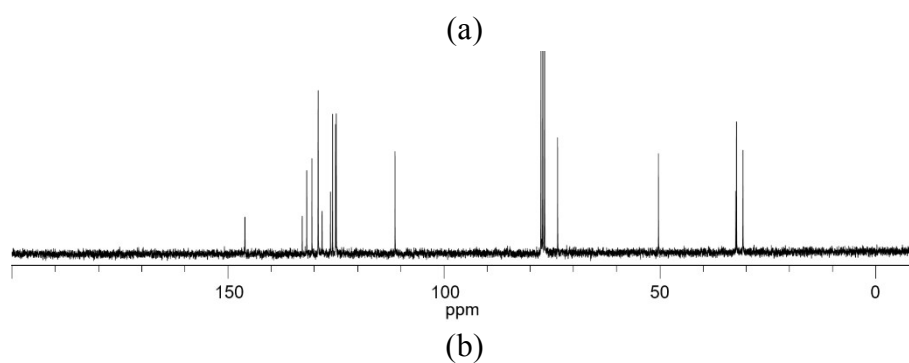
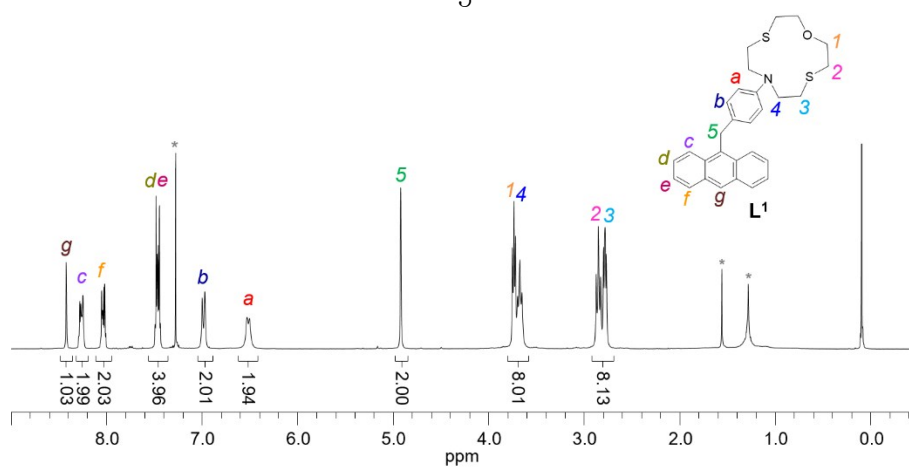


Fig. S1 (a) ^1H -NMR and (b) ^{13}C -NMR spectra of L^1 in CDCl_3 .

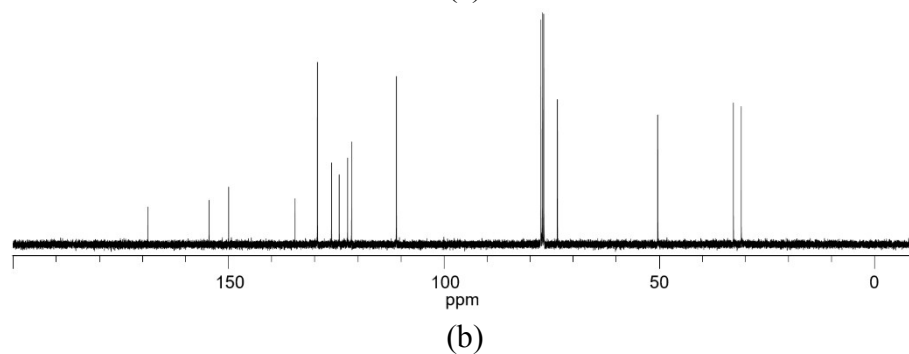
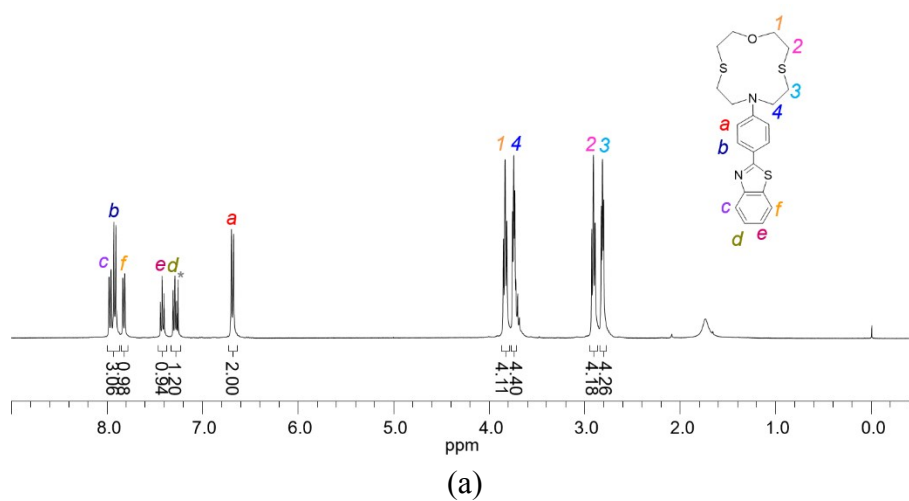
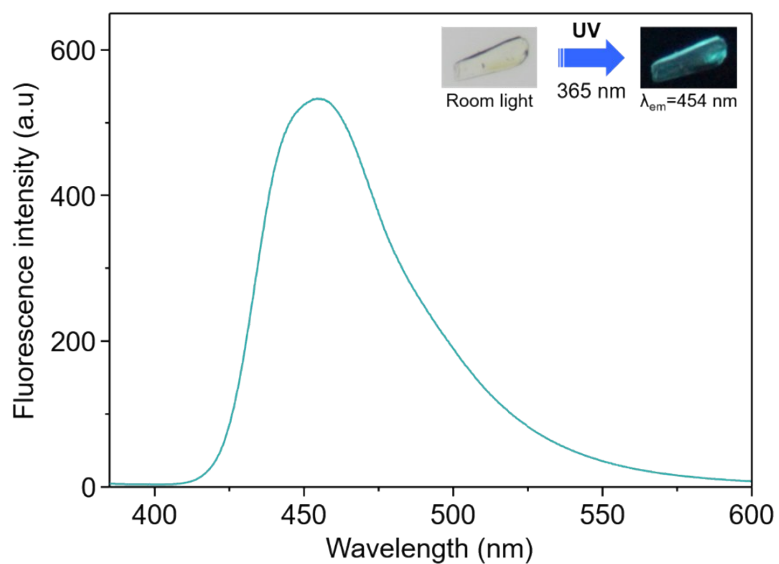
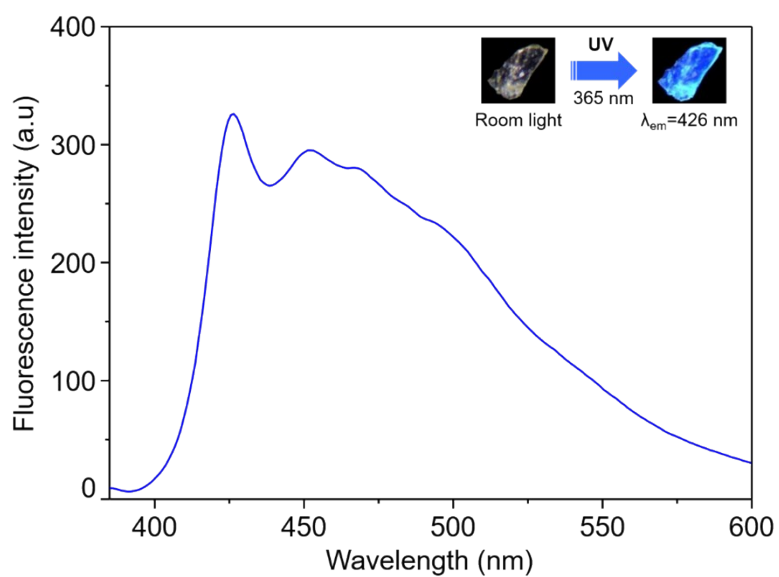


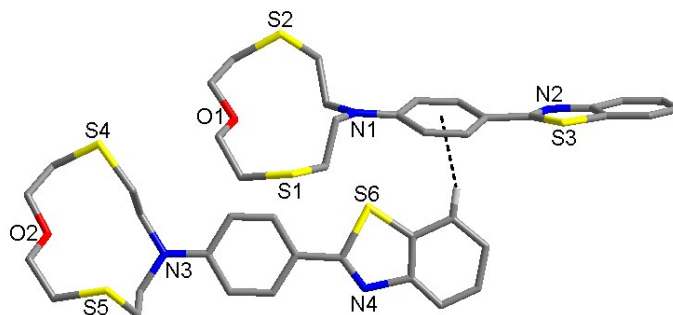
Fig. S2 (a) ^1H -NMR and (b) ^{13}C -NMR spectra of L^2 in CDCl_3 .



(a)



(b)

Fig. S3 Solid-state photoluminescence spectra of (a) L^1 and (b) L^2 ($\lambda_{ex} = 365$ nm).**Fig. S4** Crystal structure of L^2 showing the edge-to-face type $CH \cdots \pi$ interaction (2.958 Å).

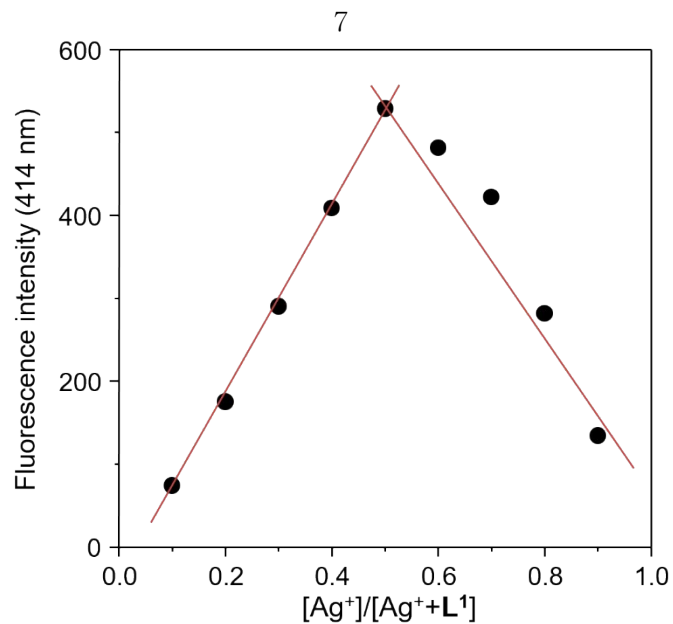


Fig. S5 Job plot of L^1 with silver(I) perchlorate, showing a 1:1 ratio (metal-to-ligand) in ethanol.

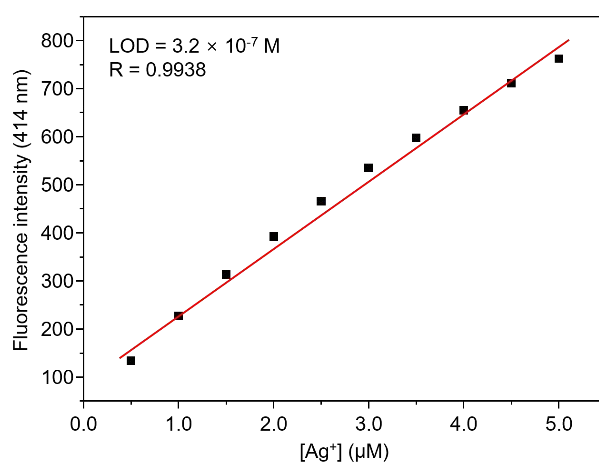


Fig. S6 Linear fitting of fluorescence intensity of L^1 with silver(I).

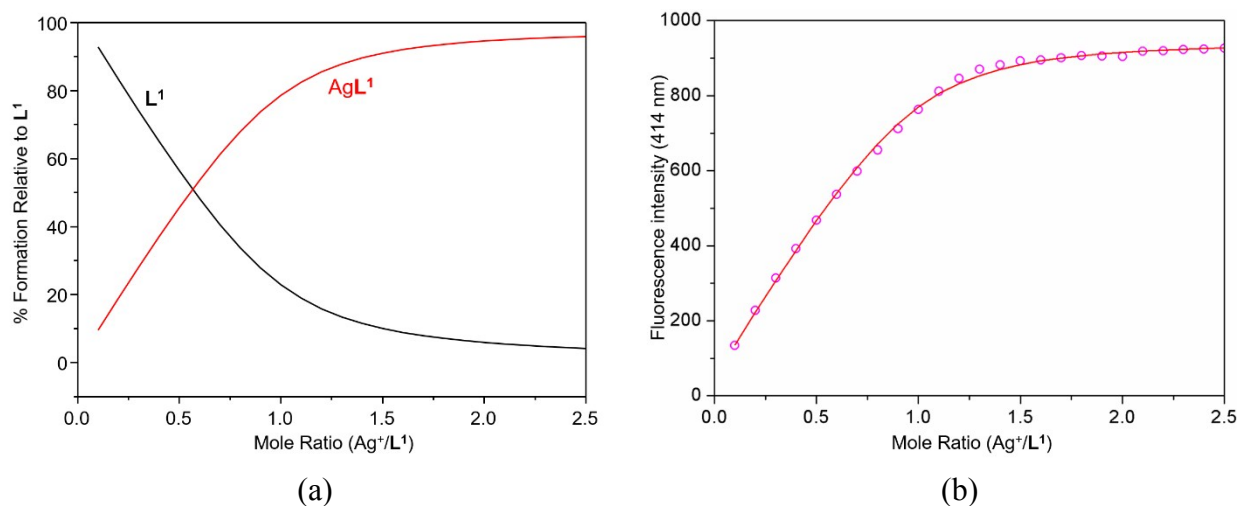


Fig. S7 Fitting of fluorescence titration data to determine the stability constants of the silver(I)- L^1 complexation with HyperSpec^{S1} software by employing the multiple binding model including 1:1 ratio: (a) species distribution diagram for L^1 and its silver(I) complexes as a function of the mole ratio (Ag^+/L^1) and (b) HyperSpec output (\circ : experimental points, solid line: theoretical fit).

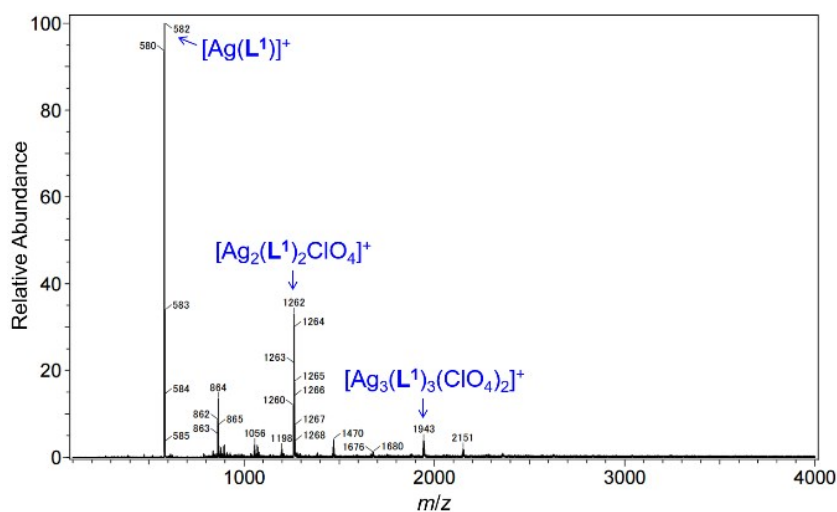


Fig. S8 CSI-MS spectrum of L^1 upon the addition of silver(I) perchlorate.

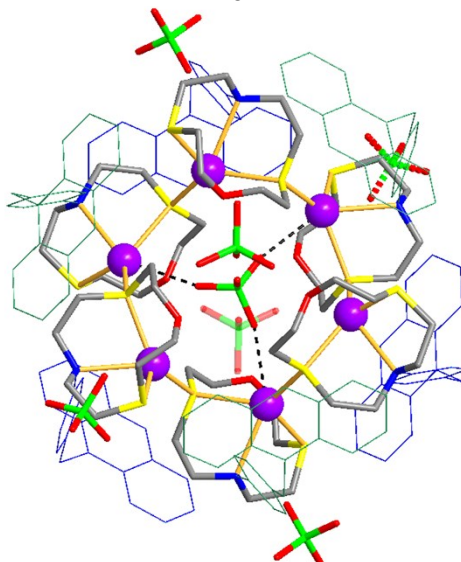


Fig. S9 Crystal structure of silver(I) perchlorate complex with L^1 , $[Ag_6(L^1)_6](ClO_4)_6$ (**1**).

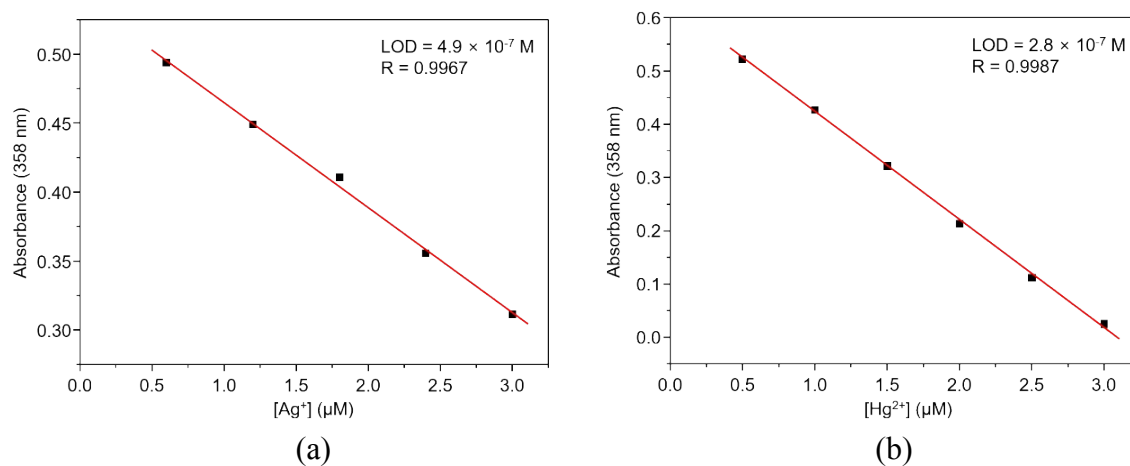


Fig. S10 Linear fitting of fluorescence intensity of L^2 with (a) silver(I) and (b) mercury (II).

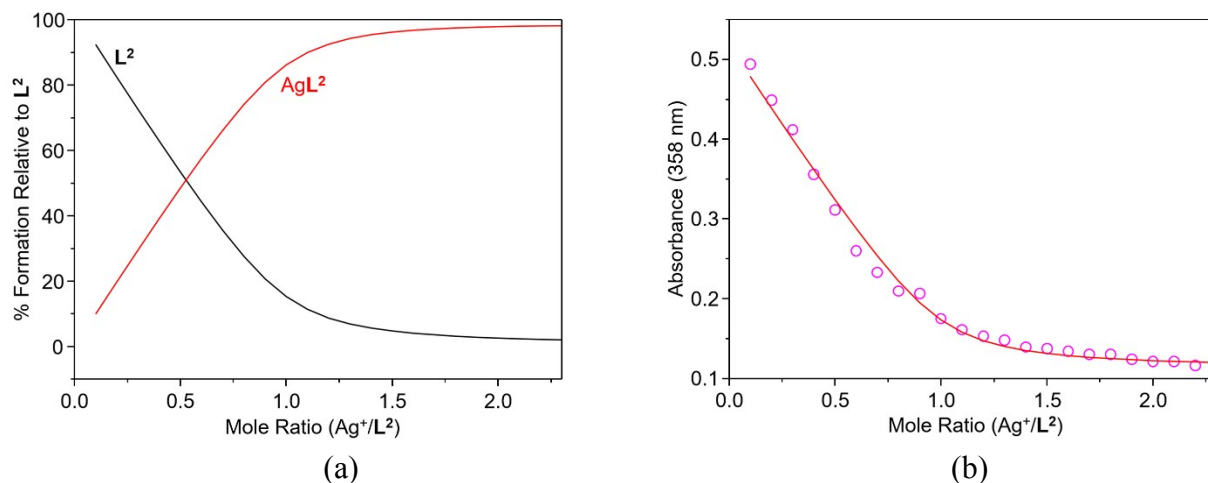


Fig. S11 Fitting of UV-vis titration data to determine the stability constants of the silver(I)- L^2 complexation with HyperSpec software^{S1} by employing the multiple binding model including 1:1 ratio: (a) species distribution diagram for L^2 and its silver(I) complexes as a function of the mole ratio (Ag^+/L^2) and (b) HyperSpec output (\circ : experimental points, solid line: theoretical fit).

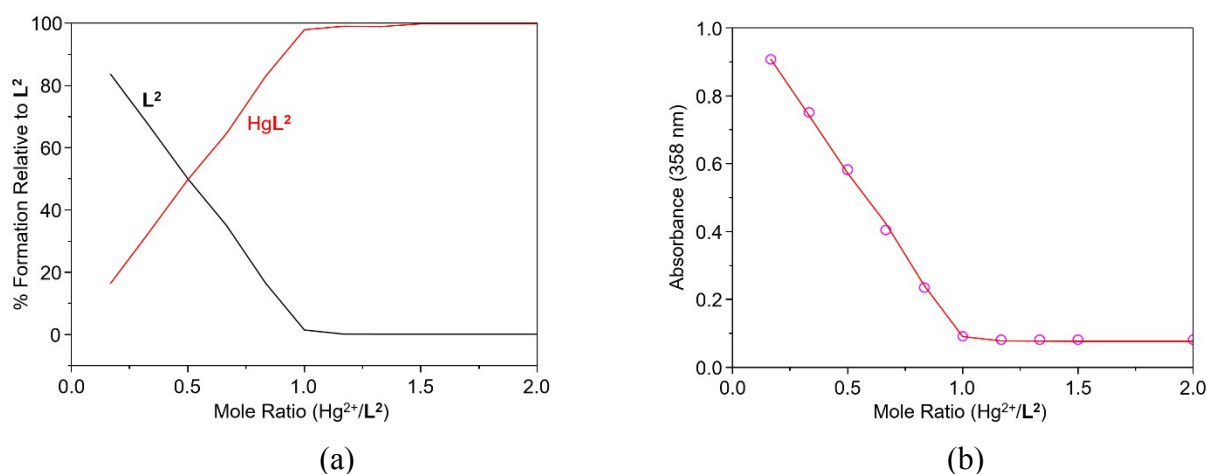


Fig. S12 Fitting of UV-vis titration data to determine the stability constants of the mercury(II)- L^2 complexation with HyperSpec software^{S1} by employing the multiple binding model including 1:1 ratio: (a) species distribution diagram for L^2 and its mercury(II) complexes as a function of the mole ratio (Hg^{2+}/L^2) and (b) HyperSpec output (\circ : experimental points, solid line: theoretical fit).

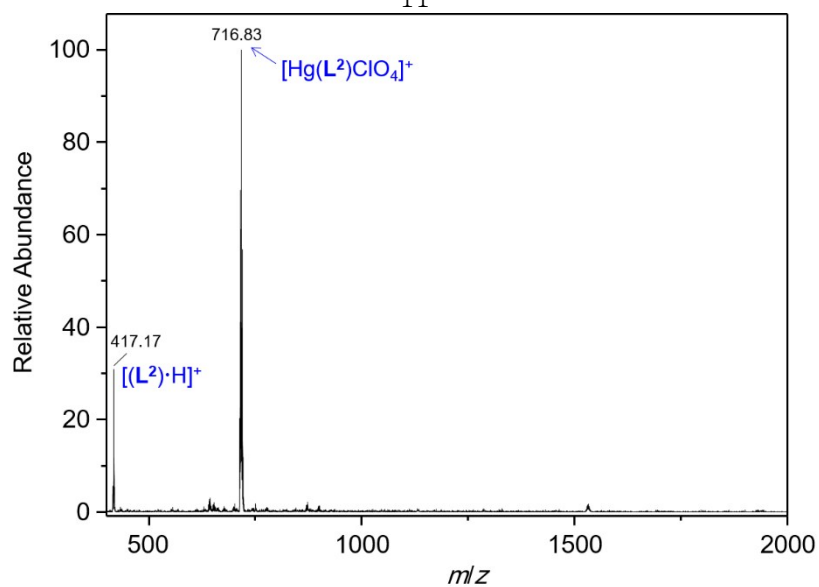


Fig. S13 ESI-MS spectrum of L^2 upon the addition of mercury(II) perchlorate.

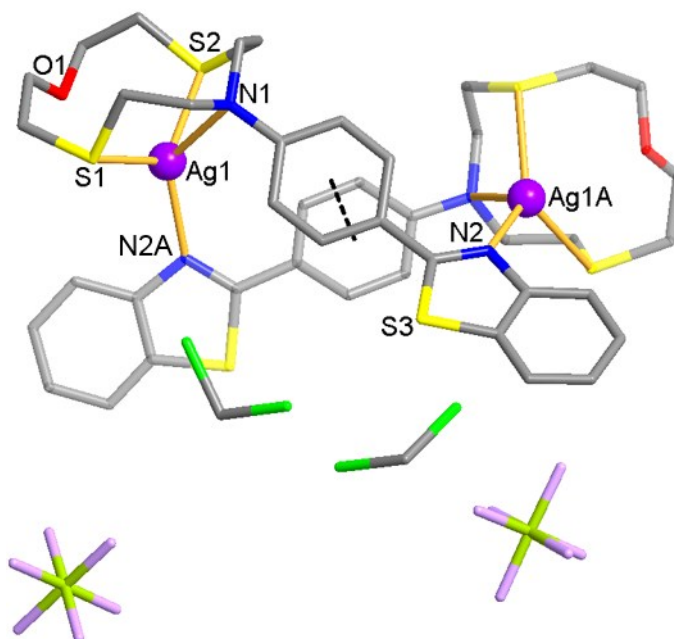


Fig. S14 Crystal structure of silver(I) hexafluorophosphate complex with L^2 , $[Ag_2(L^2)_2](PF_6)_2 \cdot 2CH_2Cl_2$ (**2**).

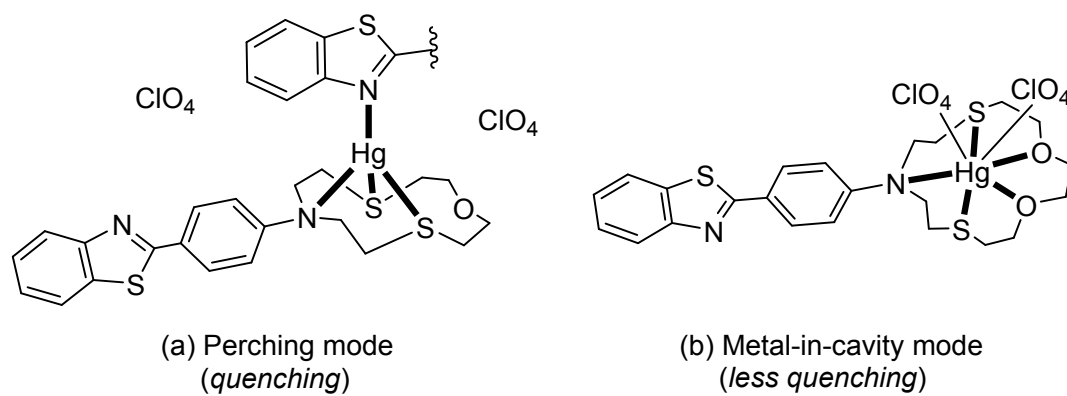


Fig. S15 Comparison of the coordination modes of mercury(II) perchlorate complexes with fluorosensors incorporating different macrocyclic receptors: (a) 12-membered NOS₂-macrocyclic (L² in this work) and (b) 15-membered NO₂S₂-macrocyclic (*see Ref. S2*).

X-ray crystallographic analysis

All data were collected on a Bruker SMART APEX2 ULTRA diffractometer equipped with graphite monochromated Mo K α radiation ($\lambda = 0.71073 \text{ \AA}$) generated by a rotating anode. Data collection, data reduction, and semiempirical absorption correction were carried out using the software package APEX2.^{S3} All of the calculations for the structure determination were carried out using the SHELXTL package.^{S4} In all cases, all nonhydrogen atoms were refined anisotropically and all hydrogen atoms were placed in idealised positions and refined isotropically in a riding manner along with their respective parent atoms. Relevant crystal data collection and refinement data for the crystal structures are summarised in Table S1. CCDC 1945425 (**L**¹), 1945426 (**L**²), 1945427 (**1**), 1945428 (**2**), 1945429 (**3**) and 1945430 (**4**) contain the supplementary crystallographic data for this paper. These data can be obtained free of charge from The Cambridge Crystallographic Data Centre via www.ccdc.cam.ac.uk/data_request/cif.

Details of Structural Refinements. In **L**², the DFIX restraints in the structural model were applied during the refinement due to the large variation of some bond geometries. For the refinement of disordered atoms, the ISOR command has been used.

In **1**, the *R* values for this structure are higher than expected due to single crystal quality and the large amounts of disordered whole backbone and perchlorate anions in the lattice. It seems that disorder of the molecules contributes to the high *R* values reported for this structure. For the refinement of disordered atoms, the commands (ISOR, SADI, SIMU, etc.) have been used. The “Alert A” in the CheckCIF: for **1**, The *R* values for this structure are higher than expected due to single crystal quality and the large amounts of disordered whole backbone and perchlorate anions in the lattice. Attempts were made to best resolve all solvent positions accurately without squeezing the data. It is likely that disorder of the molecules contributes to the high *R* values reported for this structure.

In **2**, The unit cell involves large region of solvent molecules in which thermal parameters were not satisfactory. The contribution of solvent electron density was removed by the SQUEEZE routine in PLATON.^{S5}

References

- S1. P. Gans, A. Sabatini and A. Vacca, *Talanta*, 1996, **43**, 1739–1753.
- S2. H. Ju, D. J. Chang, S. Kim, H. Ryu, E. Lee, I.-H. Park, J. H. Jung, M. Ikeda, Y. Habata, S. S. Lee, *Inorg. Chem.*, 2016, **55**, 7448-7456.
- S3. Bruker, *APEX2 Version 2009.1-0 Data collection and Processing Software*, Bruker AXS Inc., Madison, Wisconsin, U.S.A., 2008.
- S4. Bruker, *SHELXTL-PC Version 6.22 Program for Solution and Refinement of Crystal Structures*, Bruker AXS Inc., Madison, Wisconsin, U.S.A., 2008.
- S5. A. L. Spek, PLATON SQUEEZE: a tool for the calculation of the disordered solvent contribution to the calculated structure factors. *Acta Cryst.* 2015, **C71**, 9–18.

Table S2 Selected bond lengths (Å) and bond angles (°) for **1**

Ag1-N1	2.702(3)	Ag1-S1	2.569(4)
Ag1-S2	2.525(6)	Ag1-S1A	2.541(5)
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N1-Ag1-S1	74.7(5)	N1-Ag1-S2	70.0(7)
N1-Ag1-S1A	132.0(4)	S1-Ag1-S2	119.7(2)
S2-Ag1-S1A	122.6(2)	S1-Ag1-S1A	122.6(2)

Symmetry operation: (A) $x, 1.5-y, 0.5+z$ **Table S3** Selected bond lengths (Å) and bond angles (°) for **2**

Ag1-N1	2.693(7)	Ag1-N2A	2.268(7)
Ag1-S1	2.566(2)	Ag1-S2	2.504(2)
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N1-Ag1-S1	76.7(2)	N1-Ag1-S2	79.1(2)
N1-Ag1-N2A	121.8(2)	S1-Ag1-S2	123.9(1)
N2A-Ag1-S1	108.9(2)	N2A-Ag1-S2	126.9(2)

Symmetry operation: (A) $-x+1, y, -z+2$ **Table S4** Selected bond lengths (Å) and bond angles (°) for **3**

Hg1-N1	2.679(6)	Hg1-O2	2.525(4)
Hg1-S1	2.504(2)	Hg1-S2	2.527(2)
Hg1-N2A	2.315(5)		
<hr/>			
N1-Hg1-S1	78.3(1)	N1-Hg1-S2	79.4(1)
N1-Hg1-N2A	104.4(2)	N1-Hg1-O2	163.2(2)
S1-Hg1-O2	107.3(1)	S2-Hg1-O2	85.2(1)
N2A-Hg1-O2	88.5(2)	S1-Hg1-S2	130.6(1)
N2A-Hg1-S1	111.3(1)	N2A-Hg1-S2	116.8(1)

Symmetry operation: (A) $x, -y+0.5, z-0.5$ **Table S5** Selected bond lengths (Å) and bond angles (°) for **4**

Hg1-S1	2.763(3)	Hg1-I1	2.648(1)
Hg1-I2	2.656(1)		
<hr/>			
S1-Hg1-I1	107.02(6)	S1-Hg1-I2	102.73(6)
I1-Hg1-I2	138.41(5)	S1-Hg1-S1A	87.28(11)

Symmetry operation: (A) $-x-1, y, z$