

## Structural reorganisation in polytopic receptors revealed by kinetic studies

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**Table S1** Logarithms of the stepwise protonation constants for the protonation of L determined in 0.15 mol dm<sup>-3</sup> NaCl at 298.1 ± 0.1 K.<sup>1</sup>

**Table S2.** Logarithms of the stability constants for the formation of mononuclear, binuclear and trinuclear complexes of Cu<sup>2+</sup>.L calculated in 0.15 mol·dm<sup>-3</sup> NaCl at 298.1 ± 0.1 K.

**Table S3.** 400 MHz <sup>1</sup>H NMR hyperfine-shifted resonances in D<sub>2</sub>O at 298 K and pH = 7.8 for the Cu<sup>2+</sup>-L in 2:1 molar ratio.

**Fig S1.** Distribution diagrams of the species for the L/Cu<sup>2+</sup> systems as a function of pH in aqueous solution in 0.15 mol dm<sup>-3</sup> at 298.1 K; [L] = 1 × 10<sup>-3</sup> mol dm<sup>-3</sup>. (a) [Cu<sup>2+</sup>] = 1 × 10<sup>-3</sup> mol dm<sup>-3</sup>, (b) [Cu<sup>2+</sup>] = 2 × 10<sup>-3</sup> mol dm<sup>-3</sup> and (c) [Cu<sup>2+</sup>] = 3 × 10<sup>-3</sup> mol dm<sup>-3</sup>.

**Fig. S2.** 400 MHz proton NMR spectrum in D<sub>2</sub>O at 298 K and pH=7.7 of Cu<sub>2</sub>-L. In the downfield and upfield region, the intensity of the spectrum is multiplied by five. The asterisks mark the residual solvent and impurity signals (\*, H<sub>2</sub>O; \*\*, HOD).

**Fig S3.** UV-Vis spectra of ligand: Cu<sup>2+</sup> solution in 1:1, 1:2, 1:3 molar ratio at pH=9 ([L] = 1 × 10<sup>-4</sup> mol dm<sup>-3</sup>).

**Scheme S1.** Tentative coordination geometry of Cu<sub>2</sub>L.

**S1.-Synthesis of L.**

The synthesis of L has been performed as described in ref. 1.

5-(2-Aminoethyl)-2,5,8,-triaz[9]-(2,6)-pyridinophane (1.04 g, 4.16 mmol) and 1,10-phenanthroline-2,9-dicarboxaldehyde<sup>2</sup> (0.49 g, 2.08 mmol) were dissolved in 40 mL anhydrous ethanol and the mixture was stirred for 2 h at room temperature. NaBH<sub>4</sub> (0.78 g, 20.70 mmol) was then added and the resulting solution stirred for 2 h at room temperature. The ethanol was removed under reduced pressure. The resulting residue was treated with H<sub>2</sub>O (10 mL) and extracted with CH<sub>2</sub>Cl<sub>2</sub> (3 x 20 mL). The organic phase was removed at reduced pressure and the resulting residue was dissolved in ethanol and precipitated as hydrochloride salt of L in 50 % yield. mp: decomp. 345 °C <sup>1</sup>H NMR (300 MHz, D<sub>2</sub>O): δ (ppm) = 8.73 (d, *J* = 8 Hz, 2H), 8.12 (s, 2H), 8.03 (d, *J* = 8 Hz, 2H), 7.95 (t, *J* = 7 Hz, 2H), 7.44 (d, *J* = 7 Hz, 4H), 4.87 (s, 4H), 4.62 (s, 8H), 3.60-3.53 (m, 4H), 3.31-3.20 (m, 12H), 3.00-2.90 (m, 8H). <sup>13</sup>C NMR (75.43 MHz, D<sub>2</sub>O): δ (ppm) = 150.6, 149.0, 141.7, 141.3, 139.9, 129.8, 127.8, 124.6, 122.3, 51.4, 51.2, 50.8, 49.6, 46.1, 43.8. Calc for C<sub>40</sub>H<sub>54</sub> N<sub>12</sub>·6HCl: C, 52.5; H, 5.9; N, 18.4. Found: C, 52.8; H, 8.0; N, 18.2. MS (FAB) *m/z* 704 [M+H]<sup>+</sup>.

**S2.-EMF Measurements****-Experimental**

The potentiometric titrations were carried out at 298.1±0.1 K using NaCl 0.15 M as supporting electrolyte. The experimental procedure (burette, potentiometer, cell, stirrer, microcomputer, etc.) has been fully described elsewhere.<sup>3</sup> The acquisition of the emf data was performed with the computer program PASAT.<sup>4</sup> The reference electrode was an Ag/AgCl electrode in saturated KCl solution. The glass electrode was calibrated as a hydrogen-ion concentration probe by titration of previously standardized amounts of HCl with CO<sub>2</sub>-free NaOH solutions and the equivalent point determined by the Gran's method,<sup>5</sup> which gives the standard potential, E<sup>o</sup>, and the ionic product of water (pK<sub>w</sub>=13.73(1)).

The computer program HYPERQUAD was used to calculate the protonation and stability constants.<sup>6</sup> The HYSS<sup>7</sup> program was used to obtain the distribution diagrams. The pH range investigated was 2.5-11.0. In the binary Cu<sup>2+</sup>-L and Cu<sup>2+</sup>-L systems, the concentration of Cu<sup>2+</sup> and of the ligands ranged from 1x10<sup>-3</sup> to 5x10<sup>-3</sup> mol dm<sup>-3</sup> with Cu<sup>2+</sup>:L molar ratios varying from 2:1 to 1:2. The titration curves for each system (at least two titrations, ca. 200 experimental) were treated either as a single set or as separated curves without significant variations in the values of the stability constants.

When more than one model could fit the experimental data, the most reliable chemical model was chosen by performing F tests at the 0.05 confidence level.<sup>8,9</sup>

**-Acid-Base studies**

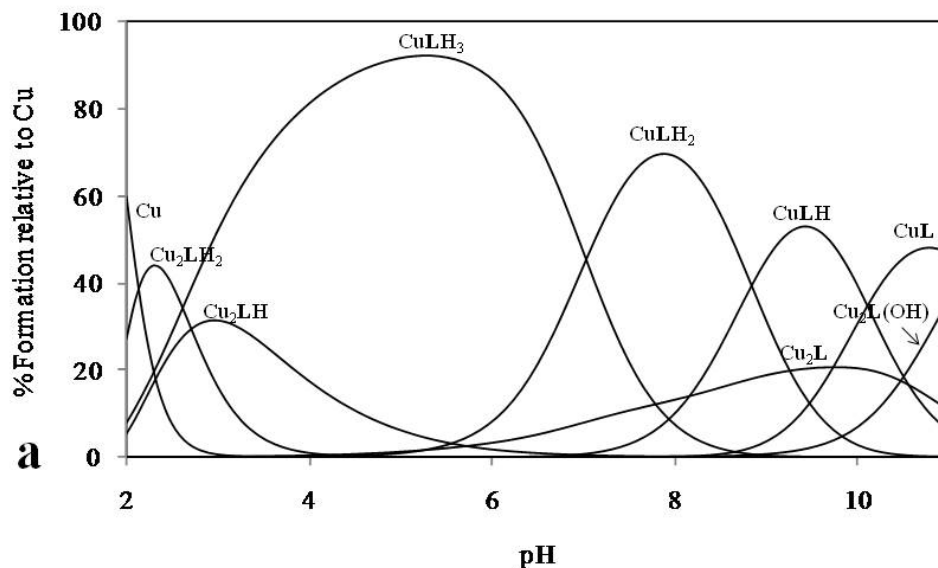
**Table S1** Logarithms of the stepwise protonation constants for the protonation of L determined in 0.15 mol dm<sup>-3</sup> NaCl at 298.1 ± 0.1 K.<sup>1</sup>

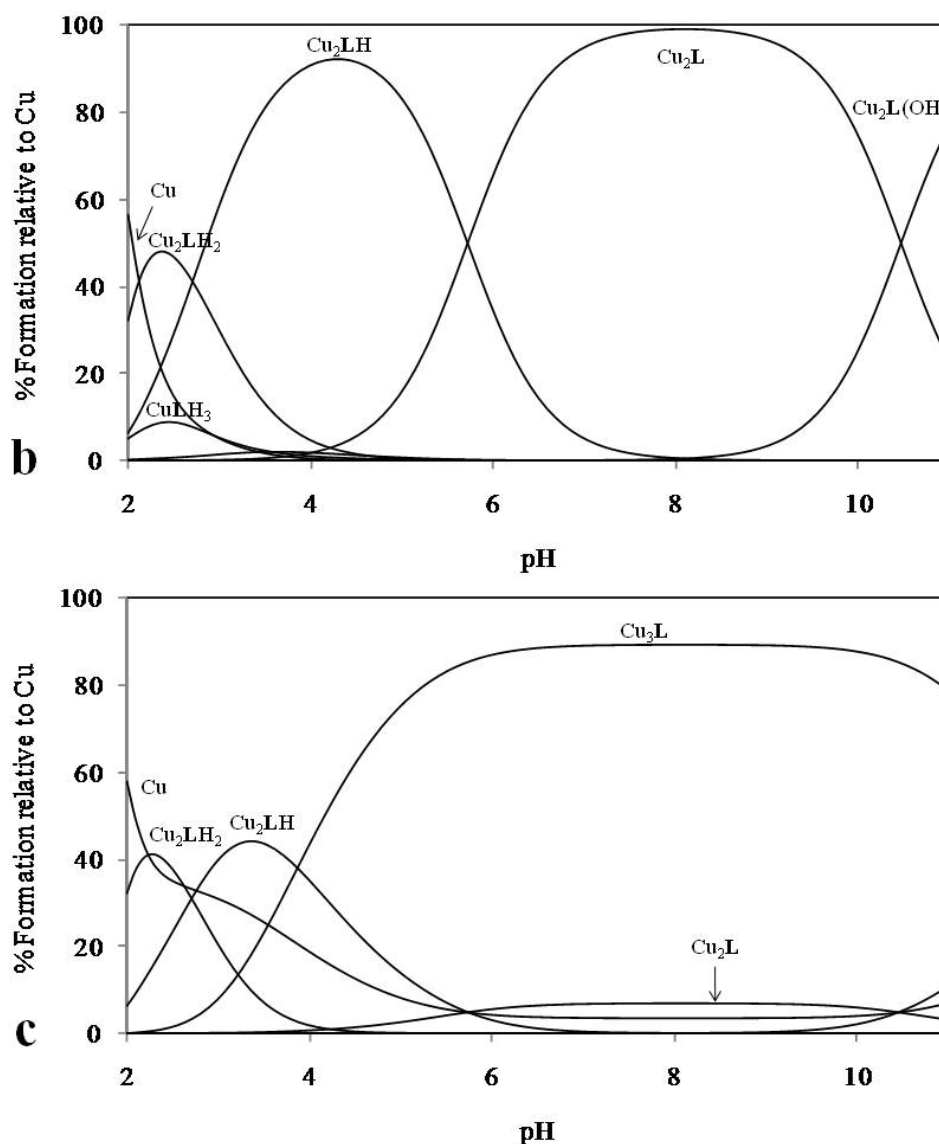
| Reaction <sup>a</sup>                   | L                     |
|---|-----------------------|
| H + L ⇌ HL                              | 10.04(1) <sup>b</sup> |
| H + HL ⇌ H <sub>2</sub> L               | 9.80(1)               |
| H + H <sub>2</sub> L ⇌ H <sub>3</sub> L | 8.88(1)               |
| H + H <sub>3</sub> L ⇌ H <sub>4</sub> L | 8.15(1)               |
| H + H <sub>4</sub> L ⇌ H <sub>5</sub> L | 6.78(1)               |
| H + H <sub>5</sub> L ⇌ H <sub>6</sub> L | 5.95(1)               |
| H + H <sub>6</sub> L ⇌ H <sub>7</sub> L | 2.25(2)               |
| Log β <sup>c</sup>                      | 51.84                 |

<sup>a</sup>Charges omitted for clarity. <sup>b</sup>Values in parentheses are standard deviation in the last significant figure. <sup>c</sup>Log β = Σ log K

Interaction with Cu<sup>2+</sup>**Table S2.** Logarithms of the stability constants for the formation of mononuclear, binuclear and trinuclear complexes of Cu<sup>2+</sup>:L calculated in 0.15 mol·dm<sup>-3</sup> NaCl at 298.1 ± 0.1 K.

| Reaction <sup>a</sup>  | L <sup>b</sup> |
|--|----------------|
| Cu + H <sub>3</sub> L ⇌ CuH <sub>3</sub> L                       | 17.55(6)       |
| Cu + H <sub>2</sub> L ⇌ CuH <sub>2</sub> L                       | 19.42(7)       |
| Cu + HL ⇌ CuHL   | 20.3(1)        |
| Cu + L ⇌ CuL   | 20.3(1)        |
| CuH <sub>2</sub> L + H ⇌ CuH <sub>3</sub> L                      | 7.02(6)        |
| CuHL + H ⇌ CuH <sub>2</sub> L                                    | 8.85(3)        |
| CuL + H ⇌ CuHL   | 10.1(1)        |
| 2Cu + H <sub>2</sub> L ⇌ Cu <sub>2</sub> H <sub>2</sub> L        | 27.9(1)        |
| 2Cu + HL ⇌ Cu <sub>2</sub> HL                                    | 35.00(7)       |
| 2Cu + L ⇌ Cu <sub>2</sub> L                                      | 39.31(9)       |
| Cu <sub>2</sub> HL + H ⇌ Cu <sub>2</sub> H <sub>2</sub> L        | 2.71(5)        |
| Cu <sub>2</sub> L + H ⇌ Cu <sub>2</sub> HL                       | 5.72(4)        |
| CuL + Cu ⇌ Cu <sub>2</sub> L                                     | 19.0(1)        |
| Cu <sub>2</sub> L + H <sub>2</sub> O ⇌ Cu <sub>2</sub> L(OH) + H | -10.46(6)      |
| Cu <sub>2</sub> L + OH ⇌ Cu <sub>2</sub> L(OH)                   | 3.26(6)        |
| Cu <sub>2</sub> L + Cu ⇌ Cu <sub>3</sub> L                       | 4.9(1)         |

<sup>a</sup>Charges omitted. <sup>b</sup>Values in parenthesis show standard deviation in the last significant figure.



**Fig S1.** Distribution diagrams of the species for the  $L/Cu^{2+}$  systems as a function of pH in aqueous solution in  $0.15 \text{ mol dm}^{-3}$  at  $298.1 \text{ K}$ ;  $[L] = 1 \times 10^{-3} \text{ mol dm}^{-3}$ . (a)  $[Cu^{+2}] = 1 \times 10^{-3} \text{ mol dm}^{-3}$ , (b)  $[Cu^{+2}] = 2 \times 10^{-3} \text{ mol dm}^{-3}$  and (c)  $[Cu^{+2}] = 3 \times 10^{-3} \text{ mol dm}^{-3}$ .

### S3.-Paramagnetic NMR spectroscopy

#### Experimental

The paramagnetic NMR measurements were acquired on a Bruker Avance400 spectrometer operating at  $399.91 \text{ MHz}$ . One-dimensional spectra were recorded in  $D_2O$  solvent with presaturation of the  $H_2O$  signal during part of the relaxation delay to eliminate the  $H_2O$  signal. Relaxation delay times of  $50\text{--}400 \text{ ms}$ ,  $30\text{--}75 \text{ KHz}$  spectral widths ranging and acquisition times of  $60\text{--}300 \text{ ms}$  were used. 1D spectra were processed using exponential line-broadening weighting functions as apodization with values of  $5\text{--}30 \text{ Hz}$ . Chemical shifts were referenced to residual solvent protons of  $D_2O$  resonating at  $4.76 \text{ ppm}$  ( $298 \text{ K}$ ) relative to TMS. Sample concentrations for paramagnetic  $^1H$  NMR were  $2.5\text{--}3 \text{ mmol dm}^{-3}$  in complexes. The longitudinal relaxation times of the hyperfine shifted resonances were determined using the inversion recovery pulse sequence ( $d_1\text{--}180^\circ\text{--}\tau\text{--}90^\circ\text{--acq}$ , where  $d_1$  is the relaxation delay and  $acq$  the acquisition time), 15 values of  $\tau$  were selected between  $0.4 \text{ ms}$  and  $500 \text{ ms}$ <sup>10</sup>, ( $d_1+acq$ ) values were at least five times the longest expected  $T_1$  ranging from  $100$  to  $800 \text{ ms}$ , and the number of scans was  $15000$ . The  $T_1$  values were calculated from the inversion-recovery equation. Transversal relaxation times were obtained measuring the line broadening of the isotropically shifted signals at half-height through the equation  $T_2^{-1} = \pi \Delta\nu_{1/2}$ .

#### Discussion

Paramagnetic  $^1H$  NMR has revealed as an useful tool to get information about electronic structure and coordination geometries in magnetically coupled polynuclear metal sites.<sup>11</sup> To this purpose, paramagnetic  $^1H$  NMR

spectra were recorded in D<sub>2</sub>O for the Cu<sup>2+</sup>-L system in 2:1 molar ratio at pH = 7.8, where the Cu<sub>2</sub>L<sup>4+</sup> species predominates in solution (see distribution diagram in Fig. S1). The spectrum shows in the down-field region, three well resolved isotropically shifted signals (a), (b), (e) and three non-resolved signals (B), (c) and (d), Fig S2. In addition it displays three signals (f-h) shifted up-field. Chemical shift values, longitudinal relaxation times (T<sub>1</sub>), linewidths at half-height and assignments are reported in Table S3 and Scheme S1.

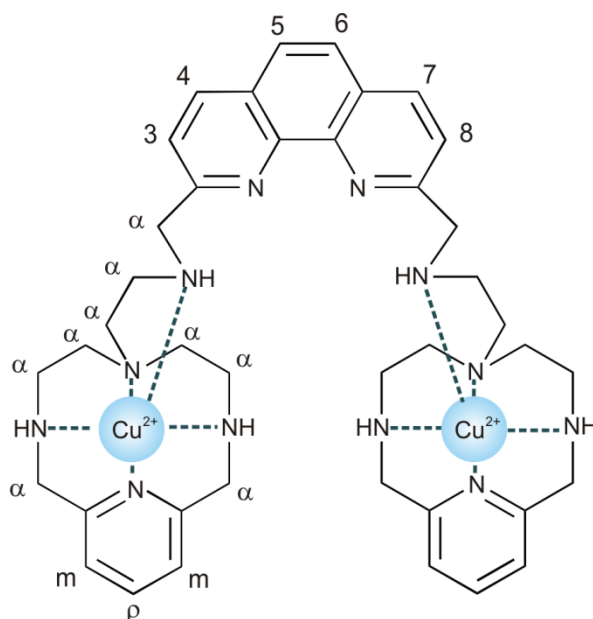
Signals a, b, c and h that present short T<sub>1</sub> values (from < 1 to 2.7 ms) and broad linewidths at half-height (280 to 840 Hz) can be attributed to methylene protons located in α-position adjacent to the paramagnetic nucleus. These signals that integrate for 36 protons, would account for a penta-coordination of both metal ions as depicted in Scheme S1. Moreover, the relatively large longitudinal relaxation times and narrow linewidths of the paramagnetically shifted signals support a significant magnetic exchange through the phenanthroline ring. This further supported by the isotropic shift undergone by the phenanthroline protons and narrow linewidths (Table S3). Variable temperature <sup>1</sup>H NMR spectra were registered for Cu<sub>2</sub>L<sup>4+</sup> from 283 to 323 K. Practically all observed isotropically shifted resonances follow an anti-Curie or independent behaviour with temperature, with the exception of resonances a, h, which correspond to α-CH<sub>2</sub> groups of the macrocycle. These results agree with the existence of an antiferromagnetic interaction between the Cu<sup>2+</sup> ions which will occur through the phenanthroline unit.

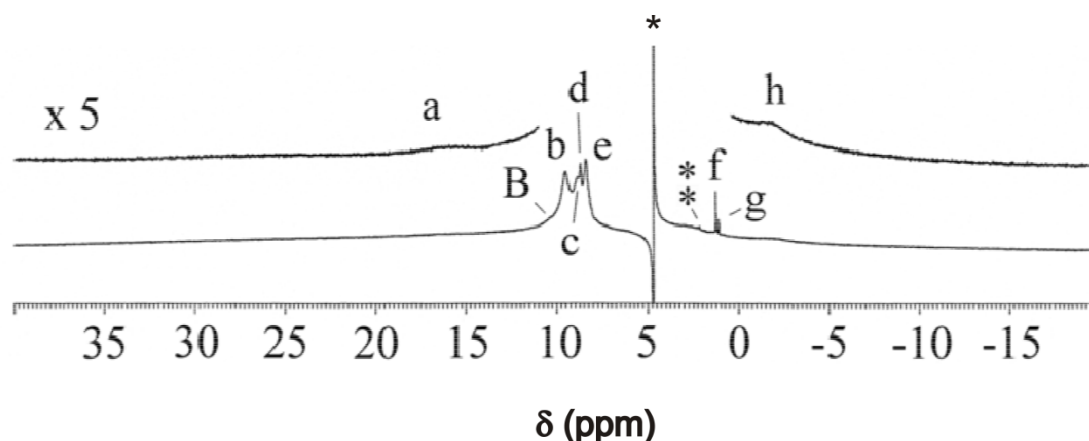
**Table S3.** 400 MHz <sup>1</sup>H NMR hyperfine-shifted resonances in D<sub>2</sub>O at 298 K and pH = 7.8 for the Cu<sup>2+</sup>-L in 2:1 molar ratio.

| Signal         | δ (ppm) | N <sup>o</sup> of protons | Assign.                    | Temp. dependence | T <sub>1</sub> (ms) | Δν <sub>1/2</sub> (Hz) | T <sub>2</sub> (ms) <sup>a</sup> |
|----------------|---------|---------------------------|----------------------------|------------------|---------------------|------------------------|----------------------------------|
| a              | 16.4    |                           |                            | Curie            | <1                  | 840                    | 0.38                             |
| b              | 9.5     |                           |                            | Indep. of T      | 2.7                 | ~280                   | ~1.1                             |
| B <sup>b</sup> | 10.3    | 36                        | αCH <sub>2</sub>           | -                | c                   | c                      | c                                |
| c              | 8.8     |                           |                            | anti-Curie       | 2.3                 | c                      | c                                |
| h              | -1.9    |                           |                            | Curie            | <1                  | 720                    | 0.44                             |
| e              | 8.4     | 6                         | H <sub>m,p</sub> -Py       | anti-Curie       | 4.2                 | ~128                   | ~2.5                             |
| d              | 8.7     | 4                         | H <sub>3,4,7,8</sub> -Phen | anti-Curie       | 5.2                 | c                      | c                                |
| f              | 1.3     |                           |                            | anti-Curie       | 167.0               | 16                     | 19.9                             |
| g              | 1.1     | 2                         | H <sub>5,6</sub> -Phen     | anti-Curie       | 62.9                | 14                     | 22.7                             |

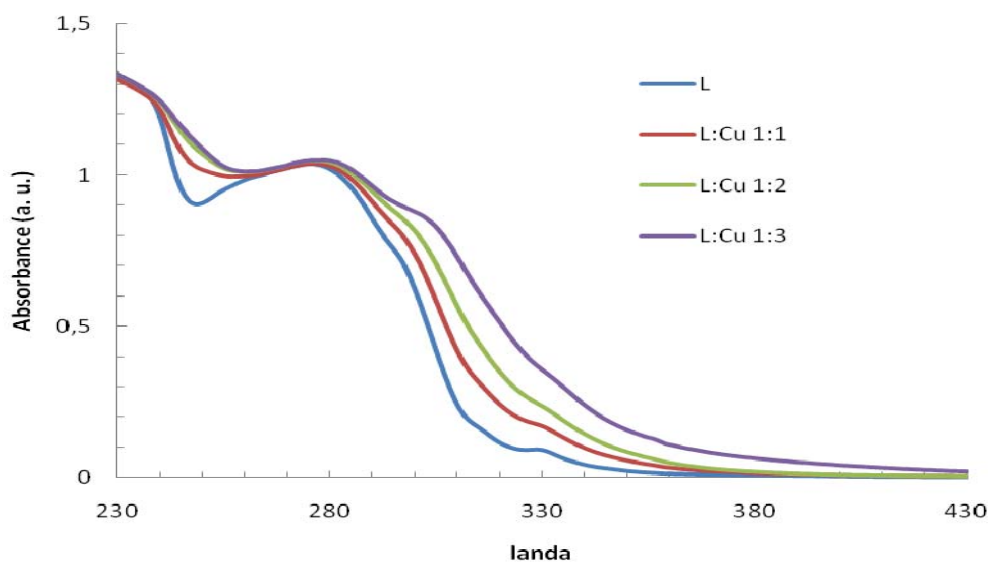
<sup>a</sup>Measured from the line width at half-height. <sup>b</sup>Measured at 313 K. <sup>c</sup>Overlap prevents measurement of this value.

**Scheme S1**





**Fig. S2.** 400 MHz proton NMR spectrum in D<sub>2</sub>O at 298 K and pH=7.7 of Cu<sub>2</sub>-L. In the downfield and upfield region, the intensity of the spectrum is multiplied by five. The asterisks mark the residual solvent and impurity signals (\*, H<sub>2</sub>O; \*\*, HOD).



**Fig S3.** UV-Vis spectra of ligand: Cu<sup>2+</sup> solution in 1:1, 1:2, 1:3 molar ratio at pH=9 ([L] = 1 × 10<sup>-4</sup> mol dm<sup>-3</sup>).

#### S4.- Kinetic experiments

The kinetic experiments were carried out at 298.1±0.1 K with either a Cary 50-BIO spectrophotometer or an Applied Photophysics SX17MV stopped-flow instrument provided with a PDA-1 diode array detector. The ionic strength was adjusted to 0.15 mol dm<sup>-3</sup> for the formation studies and 1.0 mol dm<sup>-3</sup> for the decomposition studies by adding the required amount of NaCl. The kinetic work on complex decomposition was carried out under pseudo-first-order conditions of acid excess and the solutions contained Cu<sup>2+</sup> and the corresponding ligand in 1:1, 2:1 and 3:1 molar ratio. The pH was adjusted with NaOH to values at which the [CuH<sub>3</sub>L]<sup>5+</sup>, [Cu<sub>2</sub>L]<sup>4+</sup> and [Cu<sub>3</sub>L]<sup>6+</sup> complexes were the major species in solution. For kinetic studies on complex formation, a solution of the ligand whose pH had been previously adjusted with HCl and NaOH was mixed in the stopped-flow instrument with a solution of Cu<sup>2+</sup> in 1:1, 2:1 and 3:1 molar ratio (M:L) and with the same pH than the ligand solution. The kinetic experiments covered a pH range of 3.0-5.5 and provided spectral changes with time that were analyzed with either the SPECFIT (R. A. Bistead, B. Jung et al., 2000. SPECFIT-32, Chappel Hill, Spectrum Software Associates) or Pro-KII software (M. Maeder, Y. M.

Neuhold, G. Putxy and P. B. King, *Phys. Chem. Chem. Phys.*, 2003, 13, 2836). The kinetic models used in each case are discussed in the Results and Discussion section.

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