

*Supplementary Information***Coumarin-Based Fluorescent Probes for Dual Recognition of Copper(II) and Iron(III) Ions and Their Application in Bio-Imaging. *Sensors* 2014, 14, 1385-1371**

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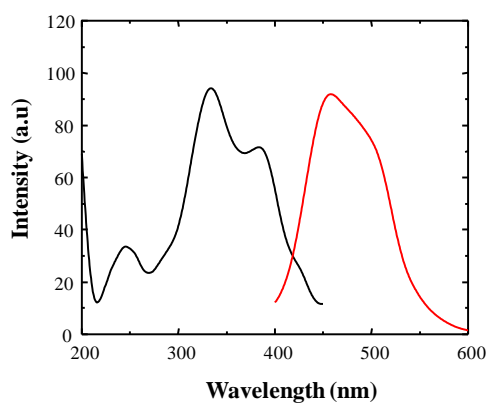
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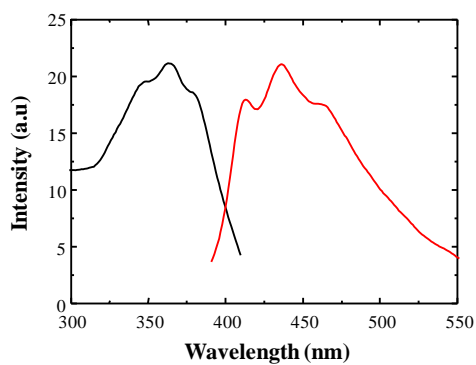
### Supplementary Information

<b>Figure S1.</b> Fluorescence spectra of excitation (black color line) and emission (red color line) of BS1 (2 $\mu$ M), $\lambda_{\text{Exc}} = 340$ nm, $\lambda_{\text{Em}} = 458$ nm.	<b>S3</b>
<b>Figure S2.</b> Fluorescence spectra excitation (black color line) and emission (red color line) of BS2 (2 $\mu$ M), $\lambda_{\text{Exc}} = 364$ nm, $\lambda_{\text{Em}} = 437$ nm.	<b>S3</b>
<b>Figure S3.</b> Fluorescence spectra excitation (black color line) and emission (red color line) of 3 (2 $\mu$ M), $\lambda_{\text{Exc}} = 336$ nm, $\lambda_{\text{Em}} = 454$ nm.	<b>S3</b>
<b>Figure S4.</b> $^1\text{H}$ -NMR spectra of 3 in DMSO- $(d_6)$ at 300 K. Red color represents t=0 and light blue color represents spectrum after water addition.	<b>S4</b>
<b>Figure S5.</b> $^1\text{H}$ -NMR spectra of BS2 at different incubation times. (A) Spectrum of a freshly prepared solution of BS2, (B) Spectrum of a solution containing BS2 after 120 min. of incubation and (C) Spectrum of a solution containing BS2 plus water.	<b>S4</b>
<b>Figure S6.</b> (A) Fluorescence spectra (2 $\mu$ M) of BS1 recorded upon the addition of copper ion (0–300 equiv.) in aqueous solution (30 mM HEPES buffer, pH 7.4, 1% DMSO). Excitation at 340 nm (slit = 5.0/5.0). (B) Fluorescence spectra (2 $\mu$ M) of BS1 recorded upon the addition of iron ion (0–300 equiv.) in aqueous solution (30 mM HEPES buffer, pH 7.4, 1% DMSO). Excitation at 340 nm.	<b>S5</b>
<b>Figure S7.</b> Reaction profile for the <i>auto-decomposition</i> reaction of BS1.	<b>S5</b>
<b>Figure S8.</b> Calculated structure for the binding modes for the complex formed between BS2 and copper ion.	<b>S5</b>
<b>Figure S9.</b> Changes in fluorescence intensity (expressed as Relative Fluorescence Units “RFU”) of solution 3 (2 $\mu$ M) induced by different metal ions.	<b>S6</b>

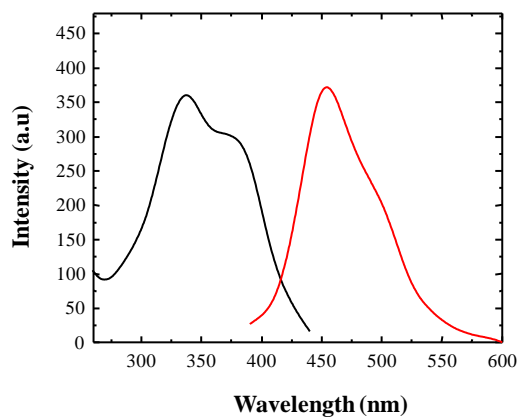
**Figure S1.** Fluorescence spectra of excitation (black color line) and emission (red color line) of **BS1** ( $2\text{ }\mu\text{M}$ ),  $\lambda_{\text{Exc}} = 340\text{ nm}$ ,  $\lambda_{\text{Em}} = 458\text{ nm}$ . Slit 5.0/5.0.



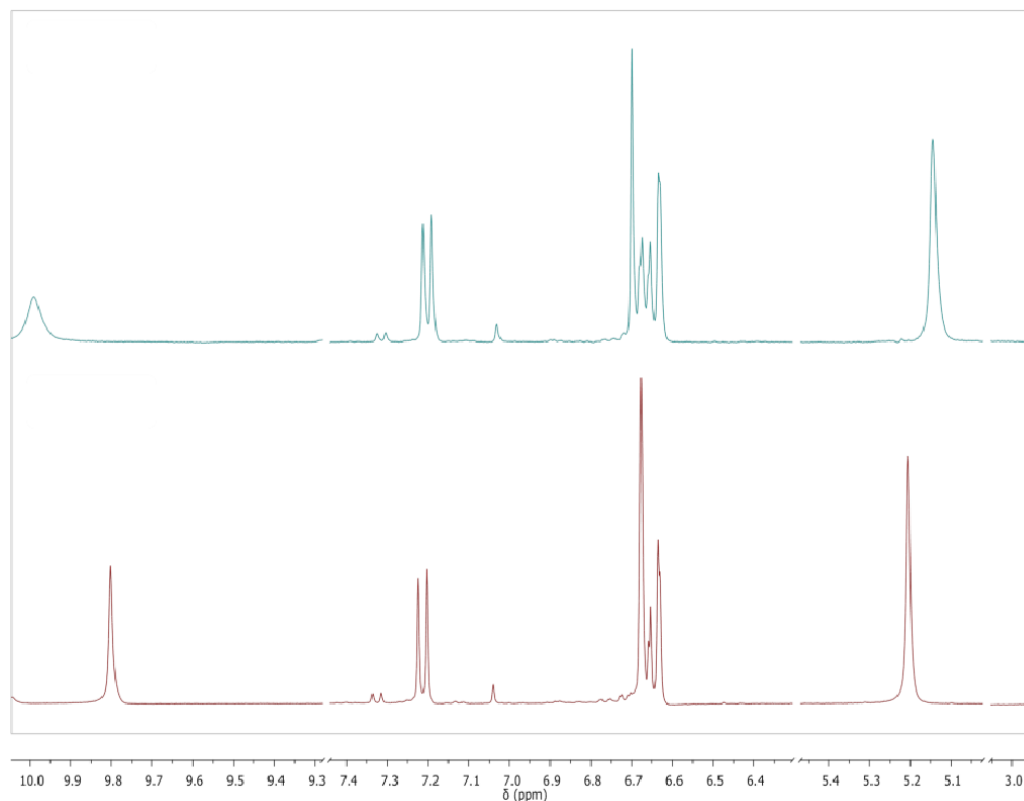
**Figure S2.** Fluorescence spectra excitation (black color line) and emission (red color line) of **BS2** ( $2\text{ }\mu\text{M}$ ),  $\lambda_{\text{Exc}} = 364\text{ nm}$ ,  $\lambda_{\text{Em}} = 437\text{ nm}$ . Slit 5.0/5.0.



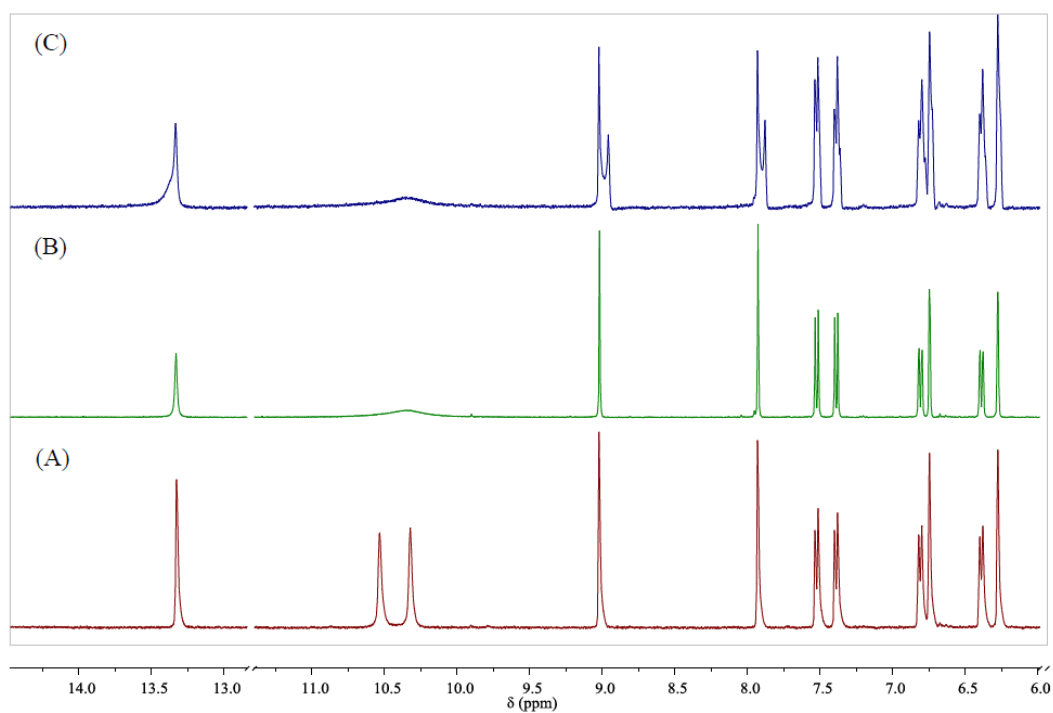
**Figure S3.** Fluorescence spectra excitation (black color line) and emission (red color line) of **3** ( $2\text{ }\mu\text{M}$ ),  $\lambda_{\text{Exc}} = 336\text{ nm}$ ,  $\lambda_{\text{Em}} = 454\text{ nm}$ . Slit 5.0/5.0.



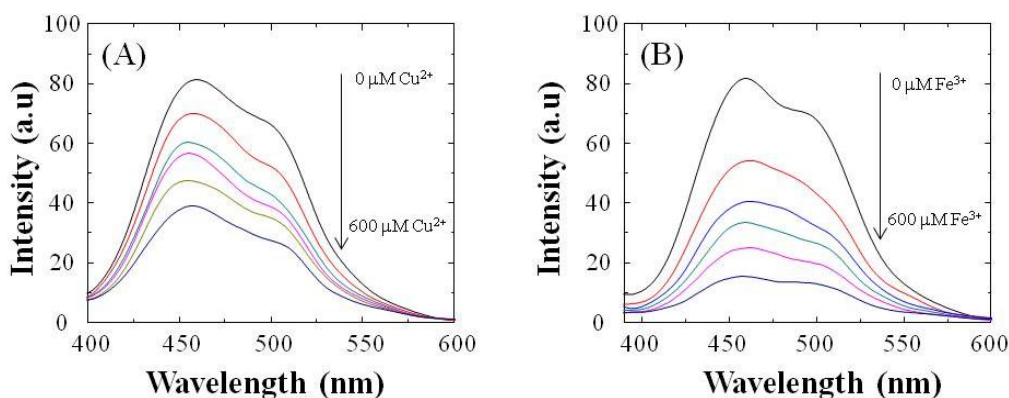
**Figure S4.**  $^1\text{H}$ -NMR spectra of **3** in  $\text{DMSO-}(d_6)$  at 300 K. Red color represents  $t = 0$  and light blue color represents spectrum after water addition (10 %).



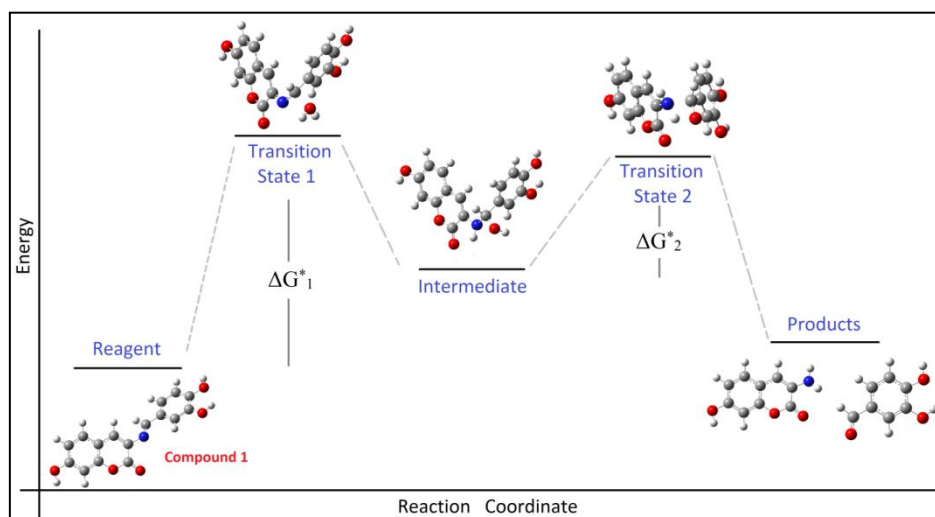
**Figure S5.**  $^1\text{H}$ -NMR spectra of **BS2** at different incubation times. (A) Spectrum of a freshly prepared solution of **BS2**; (B) Spectrum of a solution containing **BS2** after 120 min. of incubation and (C) Spectrum of a solution containing **BS2** plus water.



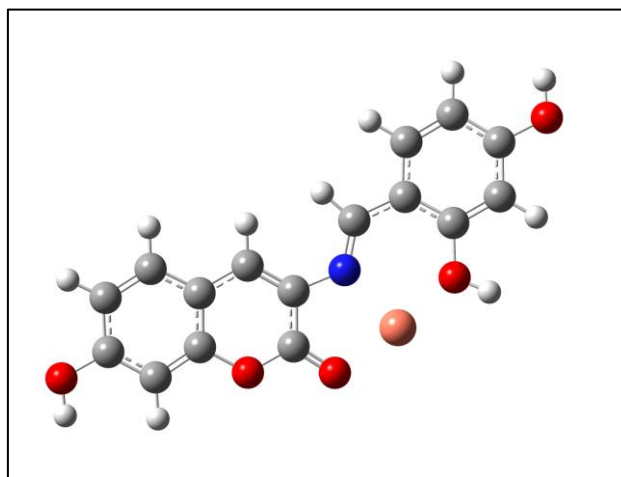
**Figure S6.** (A) Fluorescence spectra (2  $\mu\text{M}$ ) of **BS1** recorded upon the addition of copper ion (0–300 equiv.) in aqueous solution (30 mM HEPES buffer, pH 7.4, 1% DMSO). Excitation at 340 nm (slit = 5.0/5.0); (B) Fluorescence spectra (2  $\mu\text{M}$ ) of **BS1** recorded upon the addition of iron ion (0–300 equiv.) in aqueous solution (30 mM HEPES buffer, pH 7.4, 1% DMSO). Excitation at 340 nm (Slit = 5.0/5.0).



**Figure S7.** Reaction profile for the *auto-decomposition* reaction of **BS1**.



**Figure S8.** Calculated structure for the binding modes for the complex formed between **BS2** and copper ion.



**Figure S9.** Changes in fluorescence intensity (expressed as Relative Fluorescence Units “RFU”) of solution **3** (2  $\mu$ M) induced by different metal ions. In graph (A) the black bars represent the fluorescence intensity due to **3** alone, grey bars represent the fluorescence intensity due to **3** plus 300 equiv. of miscellaneous metal ions and the white bars represent the fluorescence intensity of the above solution upon further addition of 10 equiv of  $\text{Fe}^{3+}$  ( $\lambda_{\text{em}} = 454$  nm); In graph (B) the black bars represent the fluorescence intensity due to **BS2** alone, grey bars represent the fluorescence intensity due to **BS2** plus 300 equiv. of miscellaneous metal ions and the white bars represent the fluorescence intensity of the above solution upon further addition of 10 equiv of  $\text{Fe}^{3+}$  ( $\lambda_{\text{em}} = 437$  nm).

