Electronic Supplementary Information

A highly selective fluorescent probe for *in vitro* and *in vivo* detection of Hg^{2+}

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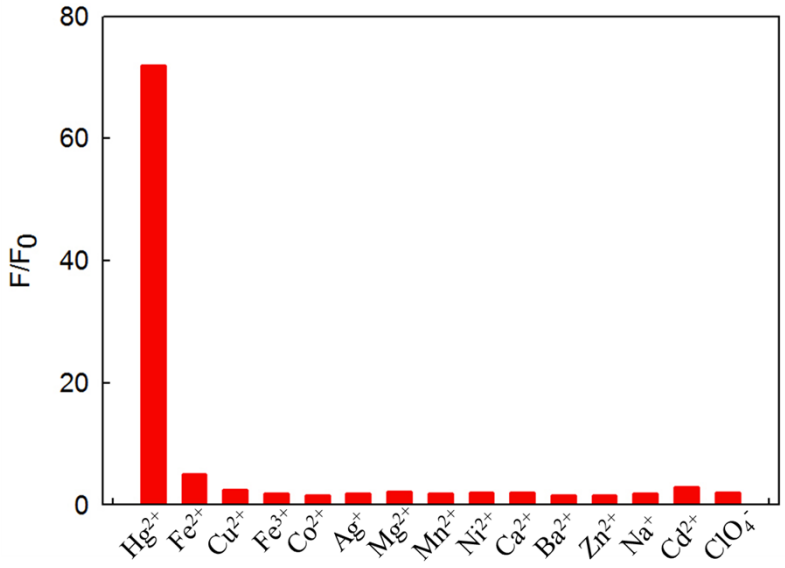

Figure S1. Fluorescence intensity changes of **RS** (10 μM) in response to various metal species and ClO_4^- (100 μM) in CH_3CN/H_2O. Excitation at 500 nm; emission 587 nm.

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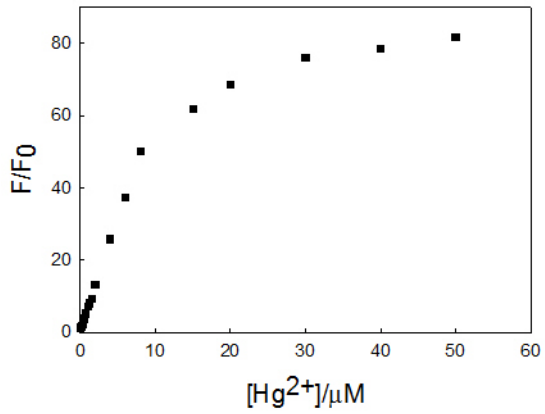


Figure S2. Plot of fluorescence intensity of RS (10 μM) as a function of added Hg^{2+} (0.1-50 μM) (λ_{ex} = 500 nm, λ_{em} = 587 nm).

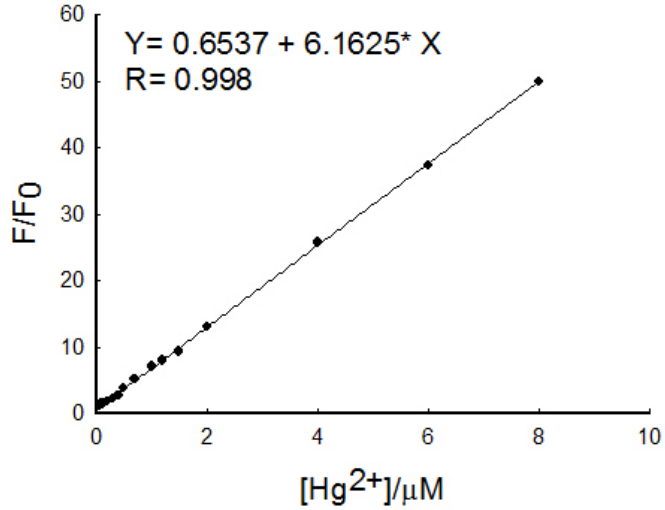


Figure S3. The liner equation of ratio of fluorescence and Hg^{2+} concentration.

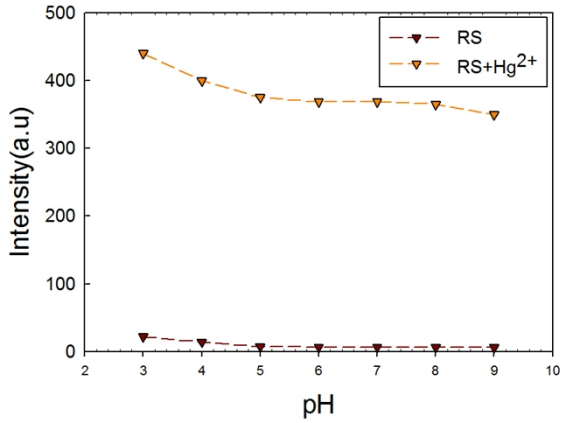


Figure S4 . Effect of pH on the emission intensity of free RS (10 μM) and [Hg^{2+}-RS] system (10:1, mole ratio, λ_{ex}= 500 nm, λ_{em} = 587 nm)

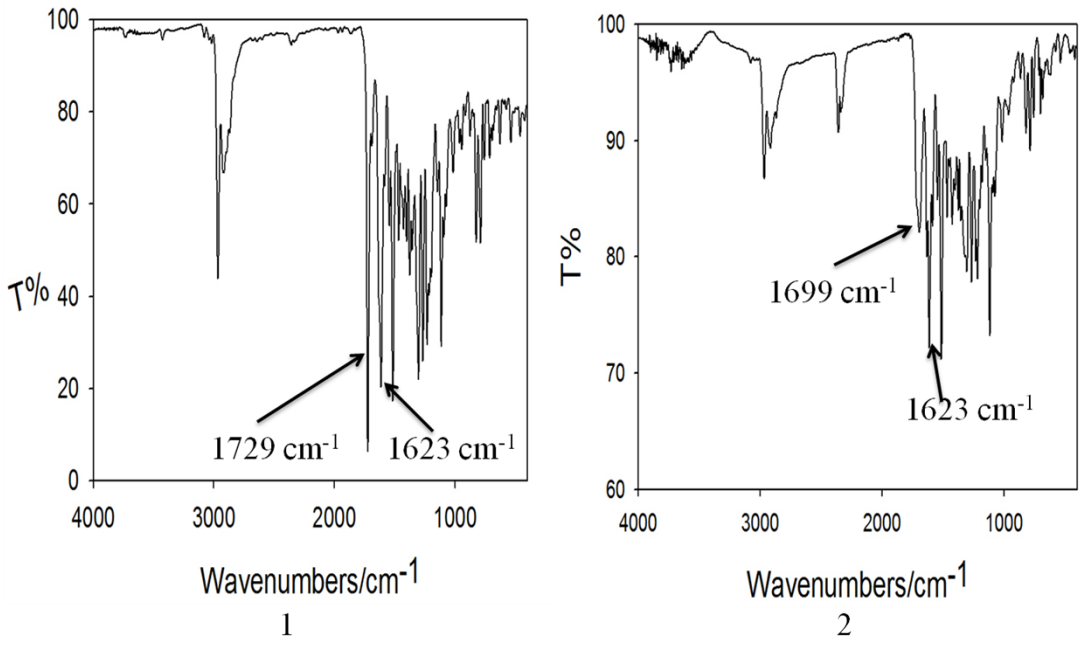


Figure S5. Infrared spectra of sensor RS (1), and sensor RS with 2.0 equiv. of Hg^{2+} (2).

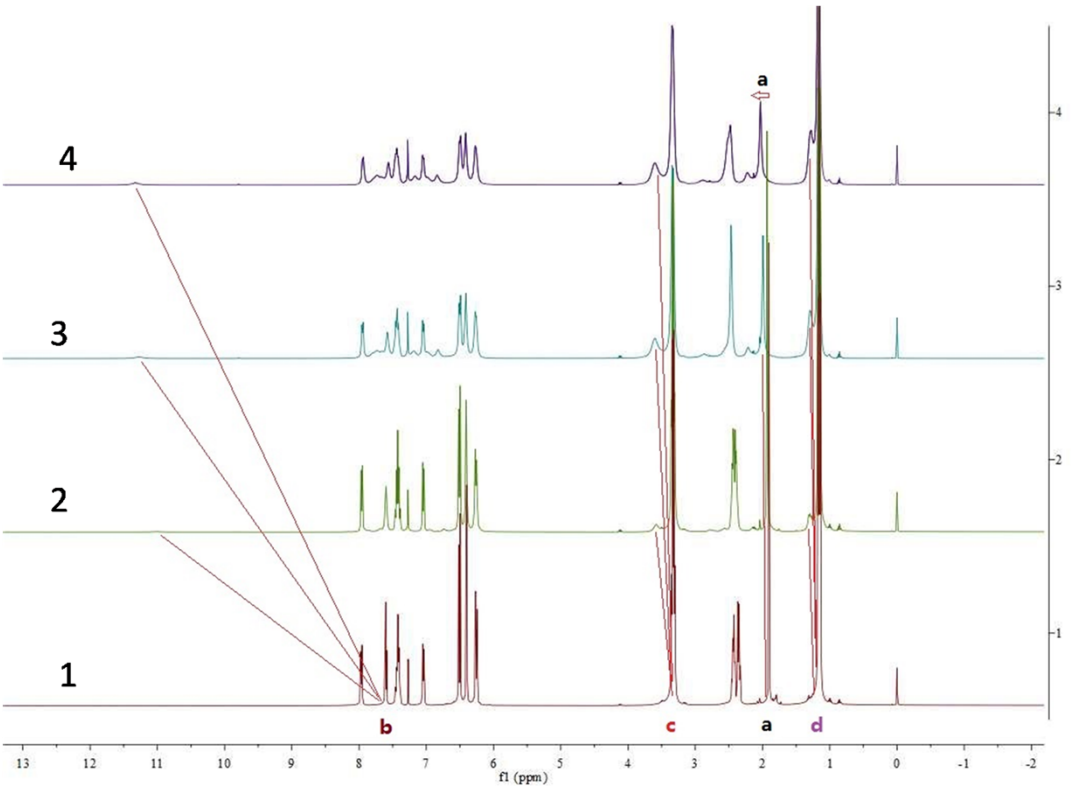


Figure S6. 1H NMR spectra of RS (1), and RS after addition of 2.0 equiv. $Hg(ClO_4)_2$ in $CDCl_3$ for 5 mins (2), 20 mins (3), and 30 mins (4).

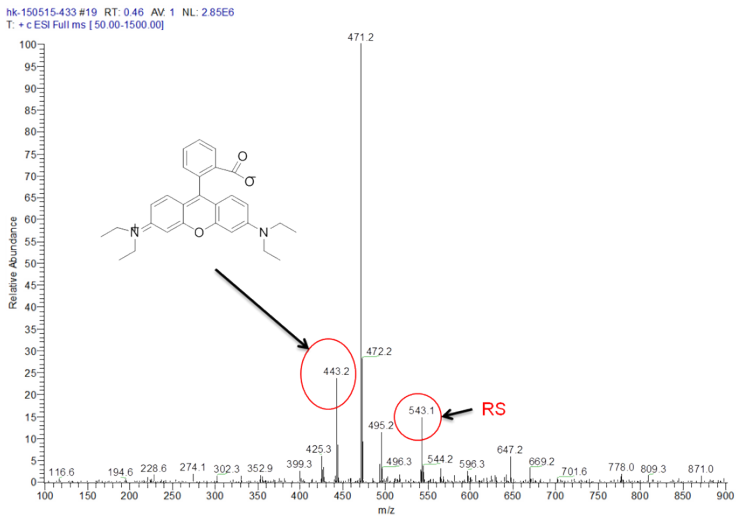


Figure S7. ESI-MS spectrum of the product formed after Hg^{2+} assisted hydrolysis of RS.

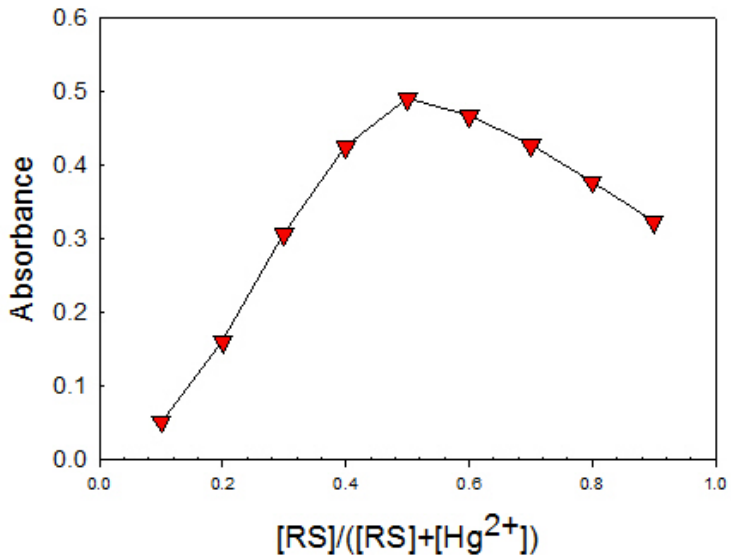


Figure S8 Job's plot for RS and Hg^{2+} complexation (the total concentration of RS and Hg^{2+} was 50 μM. λ = 558nm)

Calculation of Association Constant

The association constant was determined from the fluorescence titration data according to a reported method [1] for a 1:1 metal-ligand binding mode. If a 1:1 metal-ligand complex is formed between a metal ion and a ligand, one can describe the equilibrium as follows:

$$M + Ligand \overset{K}{\rightleftharpoons} M\,(Ligand) \quad \ldots\ldots\ldots\ldots\ldots\ldots (1)$$

Where M and M(Ligand) denote a metal ion and its complex, respectively. The corresponding association constant, *K*, can be expressed as follows:

$$K = \frac{[M(Ligand)]}{[Ligand][M]} \quad \text{.....(2)}$$

A response function for M is given below following the mass law:

$$\frac{\alpha}{1-\alpha} = \frac{1}{KC_T[M]} \quad \text{.....(3)}$$

where C_T denotes the total concentration of the ligand in the system, α defined as the ratio between the free ligand concentration ([C]) and the total concentration of ligand C_T:

$$\alpha = \frac{[C]}{C_T} \quad \text{.....(4)}$$

α can be determined from the emission changes in the presence of different concentrations of M:

$$\alpha = \frac{F - F_{max}}{F_{min} - F_{max}} \quad \text{.....(5)}$$

where Fmax and Fmin are the limiting emission values for α = 1 (in the absence of M) and α = 0 (the ligand is completely complexed with M), respectively.

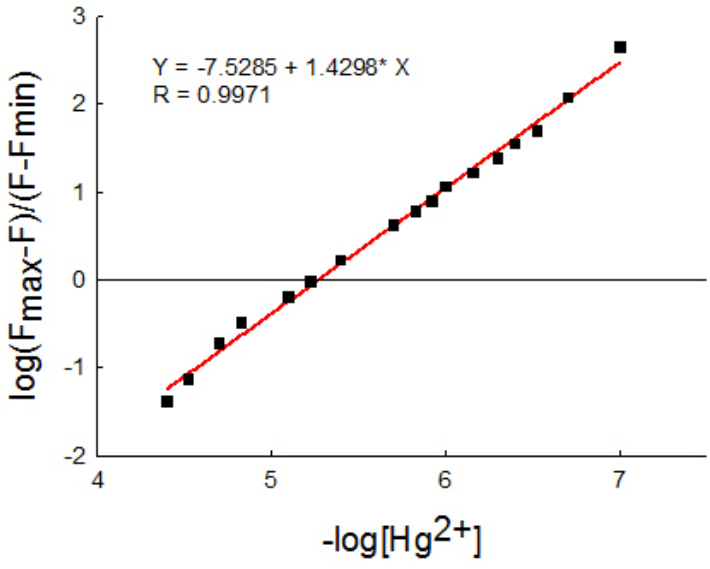


Figure S9. The bind constant of sensor RS and Hg2+ in CH3CN/H2O, 5:5, V/V. K=3.2 × 10^7. LogK=7.5.

References

[1] R. Yang, K. Li, K. Wang, F. Zhao, N. Li, F. Liu, *Anal. Chem.* 2003, *75*, 612-621.

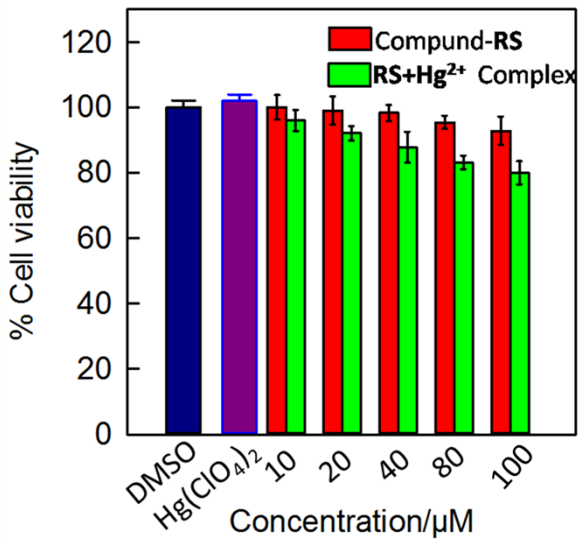
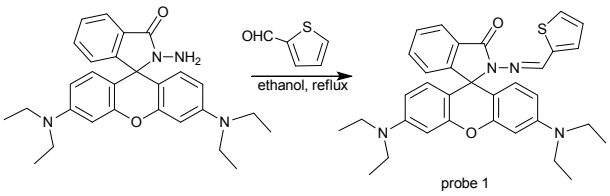


Figure S10 MTT assay to determine the cytotoxic effect of compound RS and RS-Hg^{2+} complex in HeLa cells.



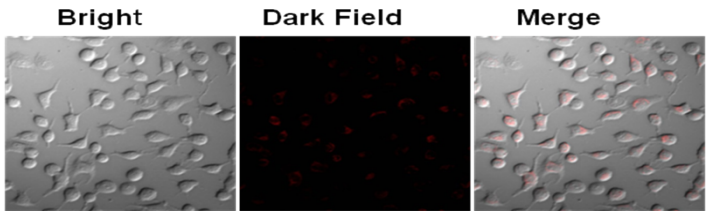


Figure 11. Fluorescence microscopic images of HeLa cells: treating with 100 μM probe 1 and after addition100 μM of Hg^{2+} (under green light) to the probe 1 treated cells.

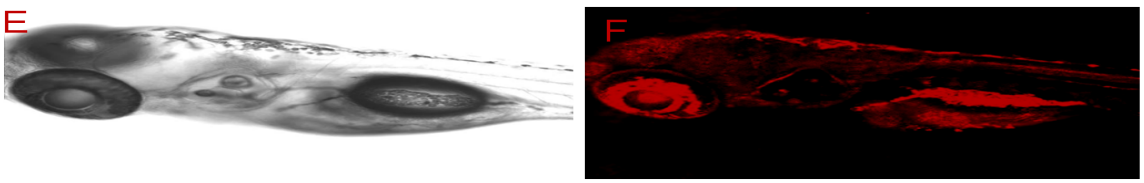
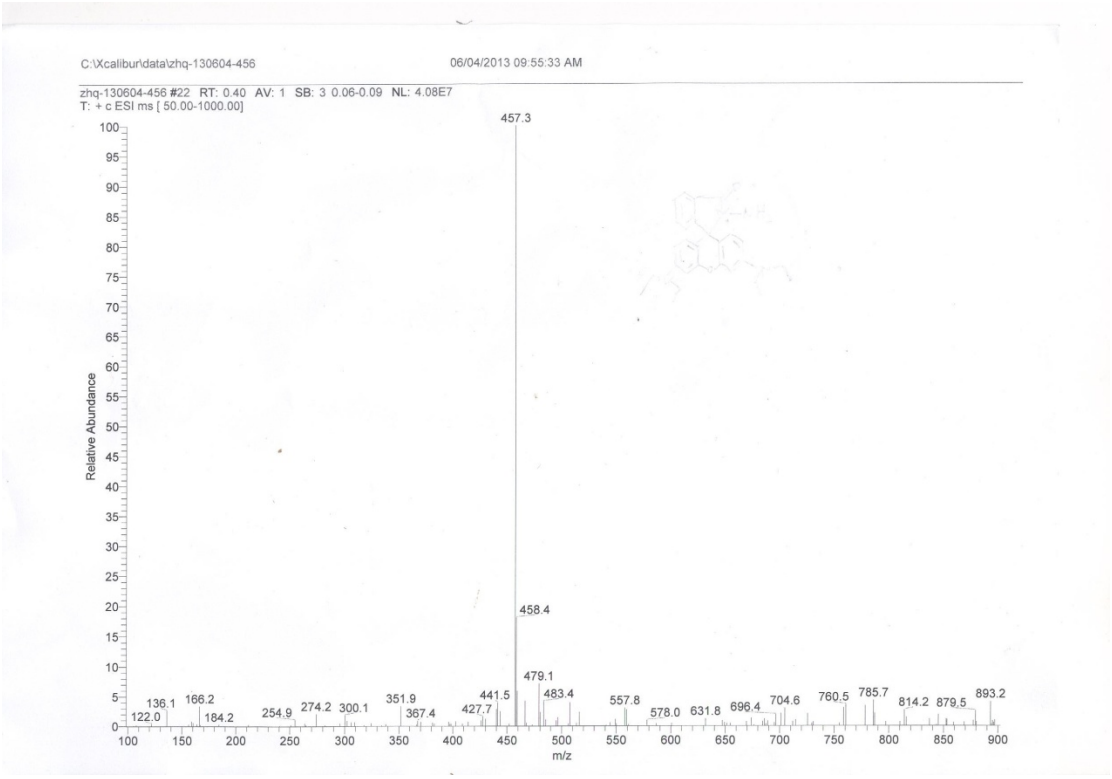
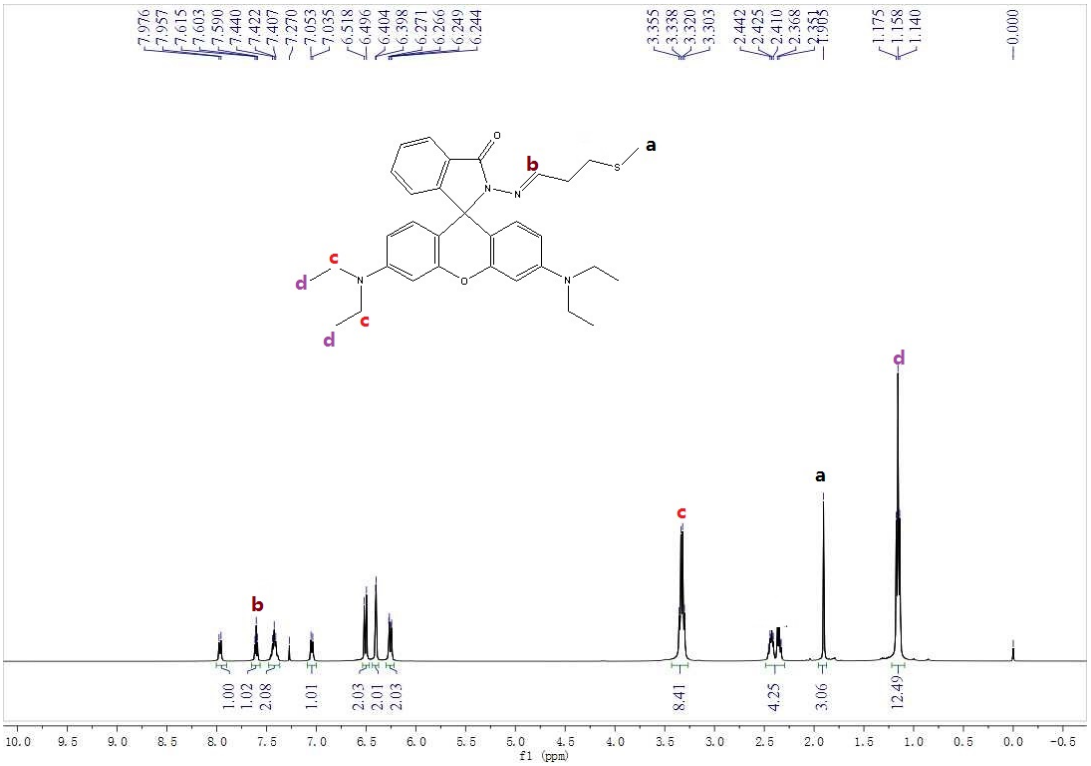


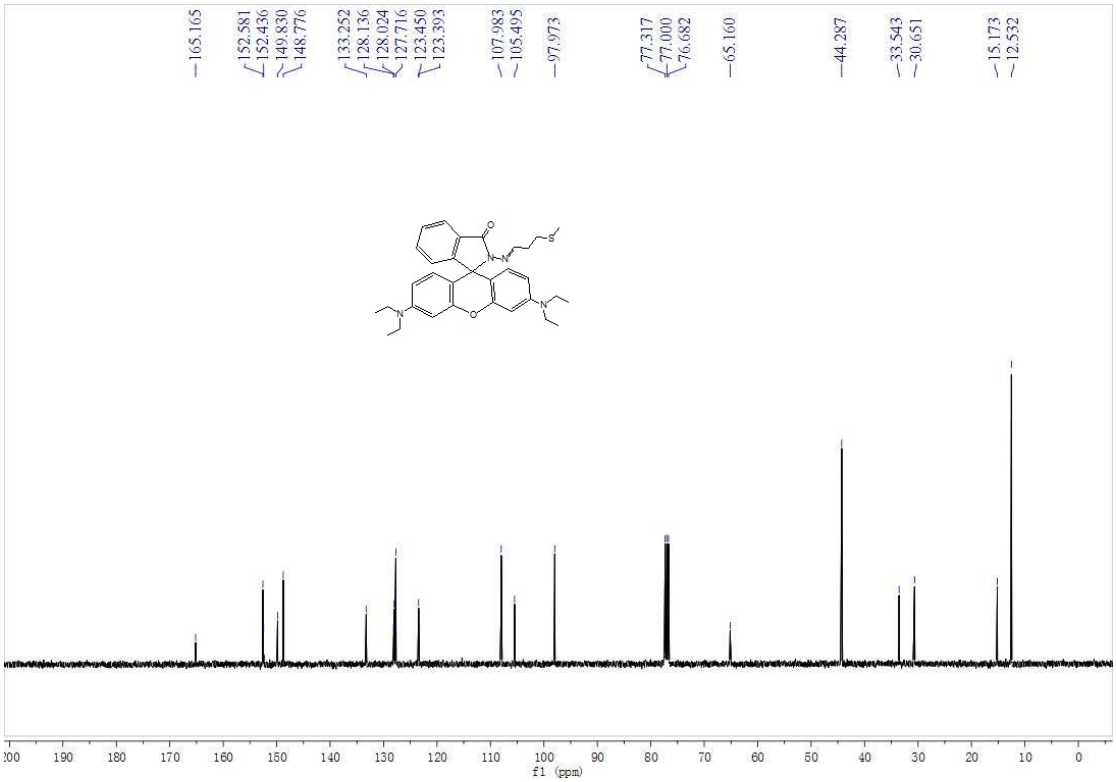
Figure 11. Bright-field (E) and Fluorescence microscopic (F) images of three-day-old zebrafish incubated with Hg^{2+} (20 μM)) and after addition of RS (20 μM).



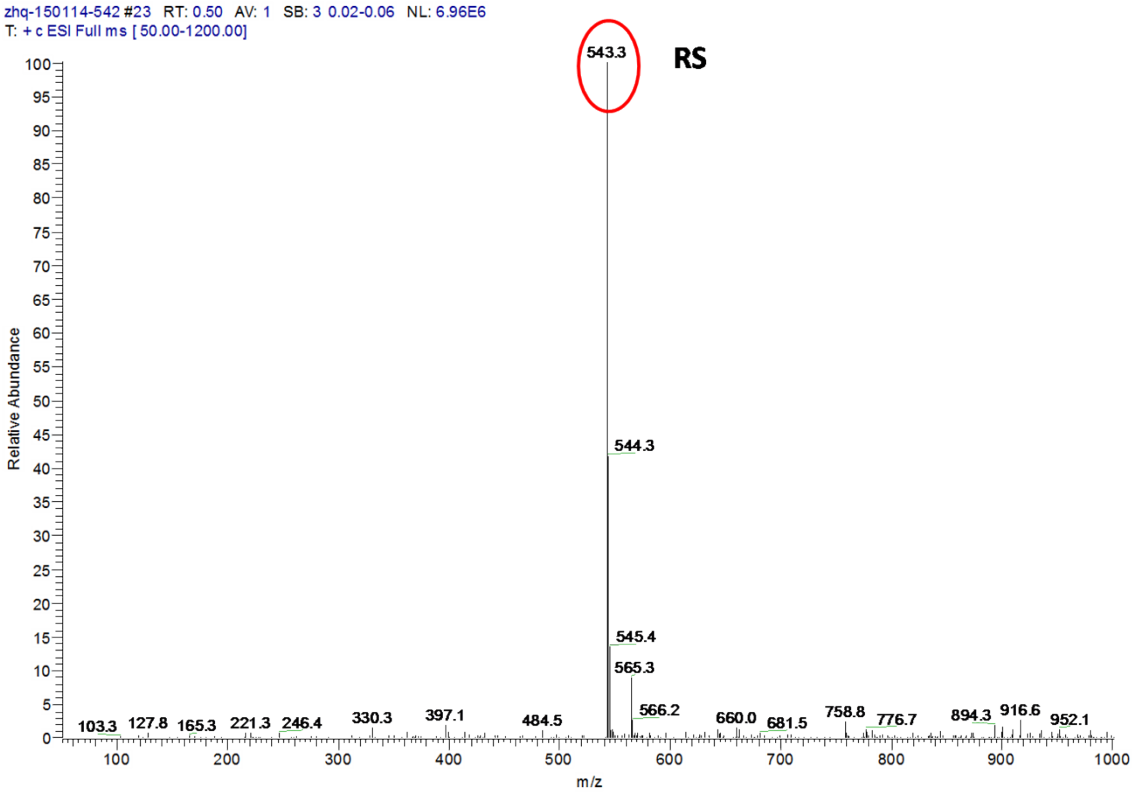
Ms of compound Rhodamine-$NHNH_2$



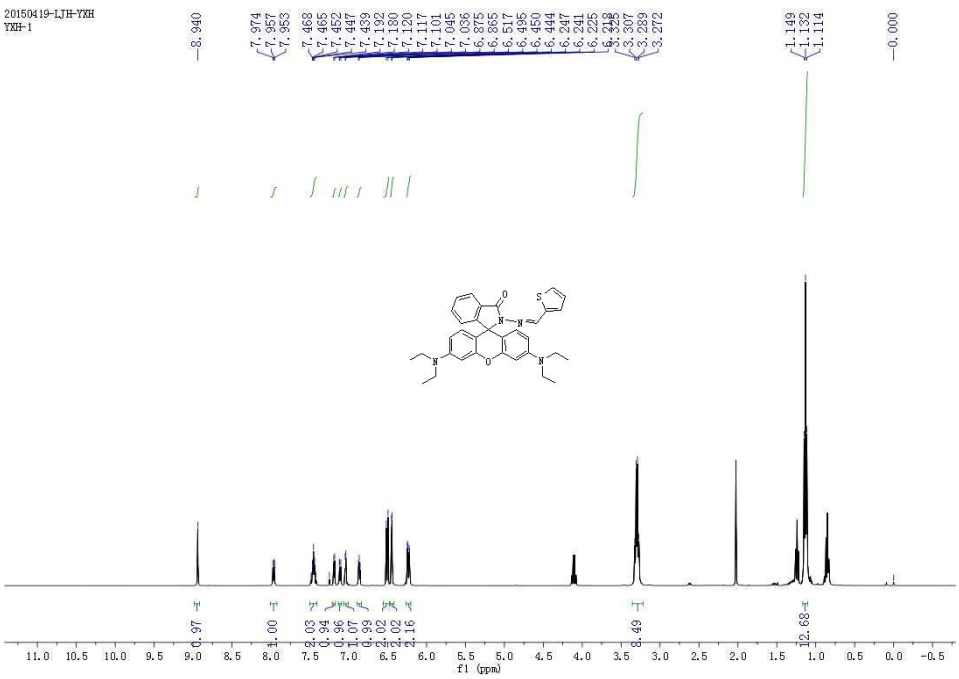
^{1}H NMR of RS in $CDCl_3$



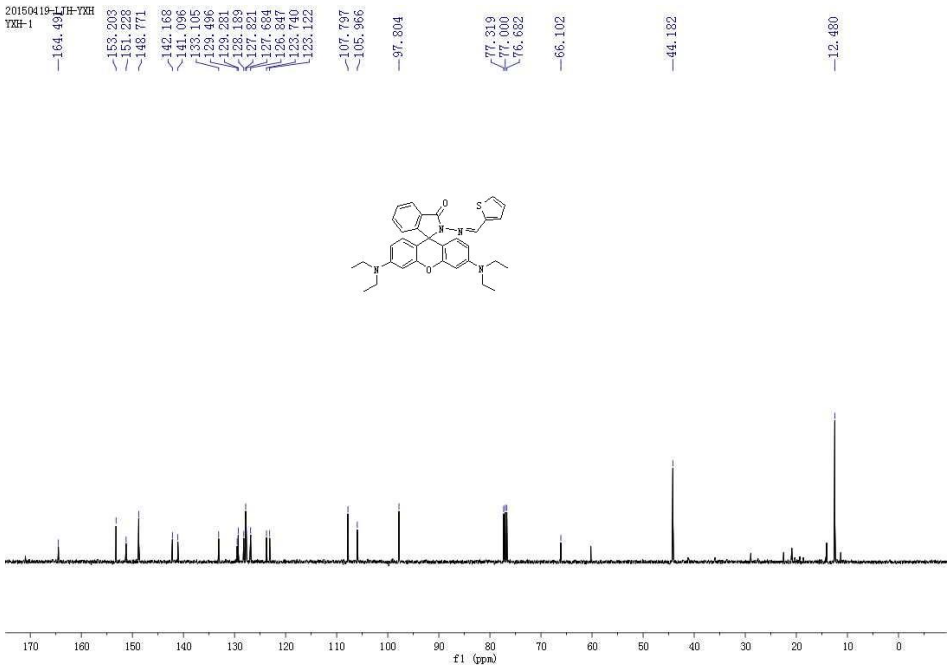
^{13}C NMR of RS in $CDCl_3$



Ms of compound RS



^{1}H NMR (400 MHz) of probe 1 in $CDCl_3$.



^{13}C NMR of probe 1 in $CDCl_3$.