

## Supplemental Information

### **A Ratiometric Two-photon Fluorescent Probe for Fluoride Ions Imaging in Living HeLa Cells and Zebrafish**

*Wei Hu<sup>†</sup>, Lingyu Zeng<sup>‡</sup>, Zhihong Liu<sup>‡</sup>, Yanying Wang<sup>†</sup>, Xiaoxue Ye<sup>†</sup>, and Chunya Li<sup>†,\*</sup>*

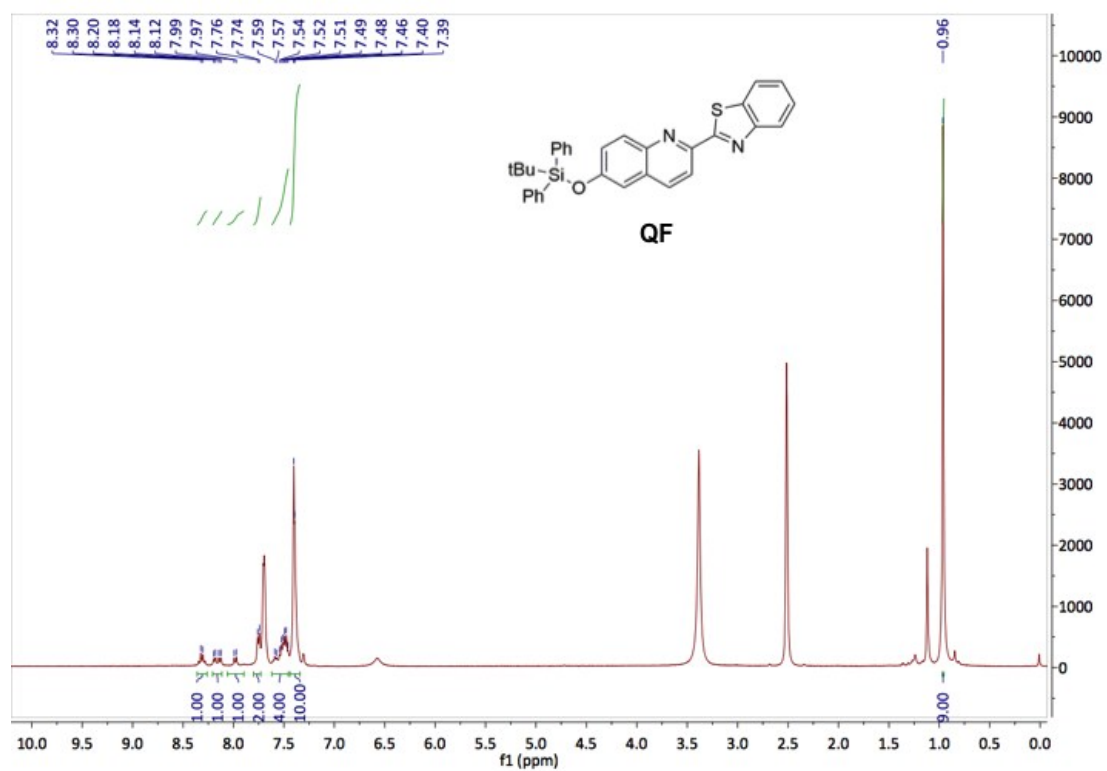
<sup>†</sup> Key Laboratory of Analytical Chemistry of the State Ethnic Affairs Commission, College of Chemistry and Material Science, South-central University of Nationalities, Wuhan 430074, China.

<sup>‡</sup> Key Laboratory of Analytical Chemistry for Biology and Medicine (Ministry of Education), College of Chemistry and Molecular Sciences, Wuhan University, Wuhan 430072, China.

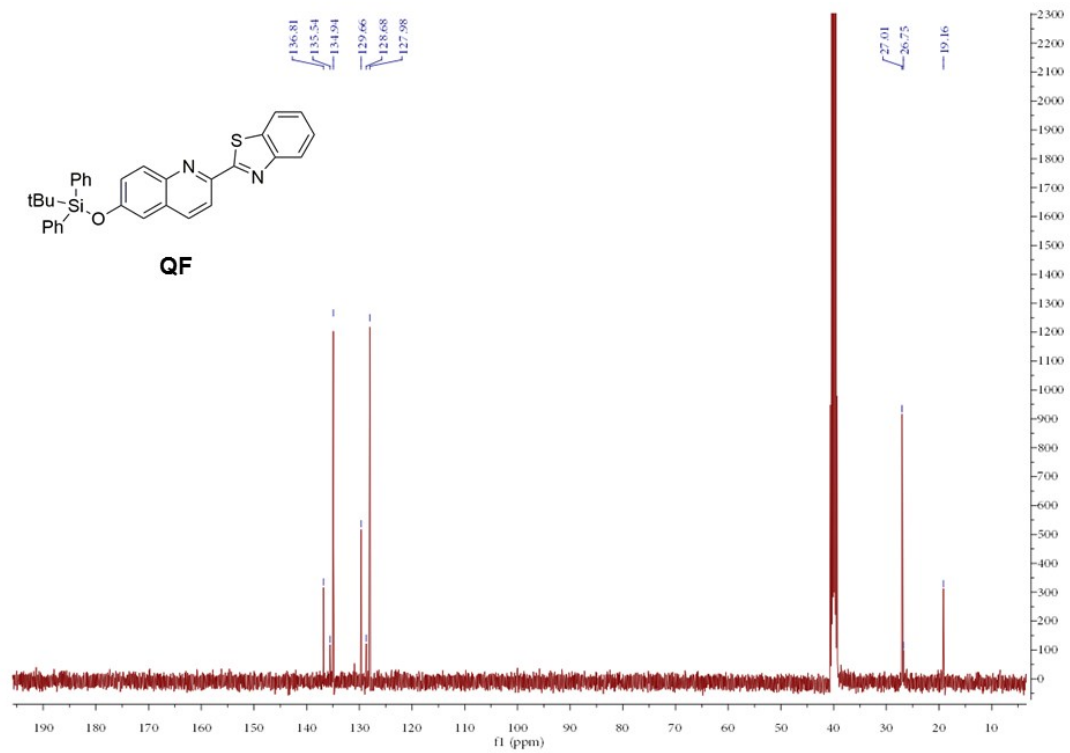
\* Corresponding author.

Tel: +86 27 67842752;

E-mail address: lcychem@yahoo.com

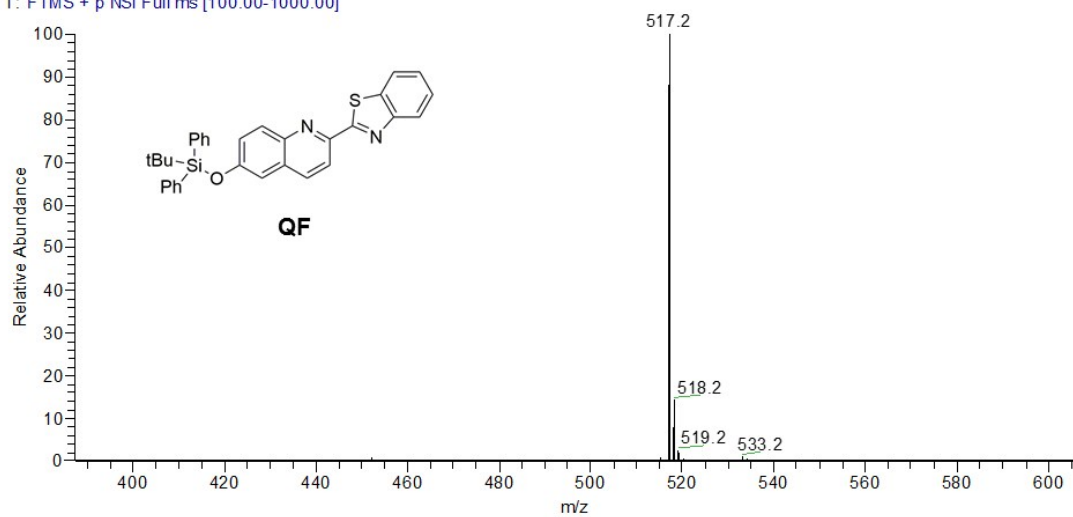


**Fig. S 1**  $^1\text{H-NMR}$  spectrum of **QF** (300 MHz,  $d_6$ -DMSO)

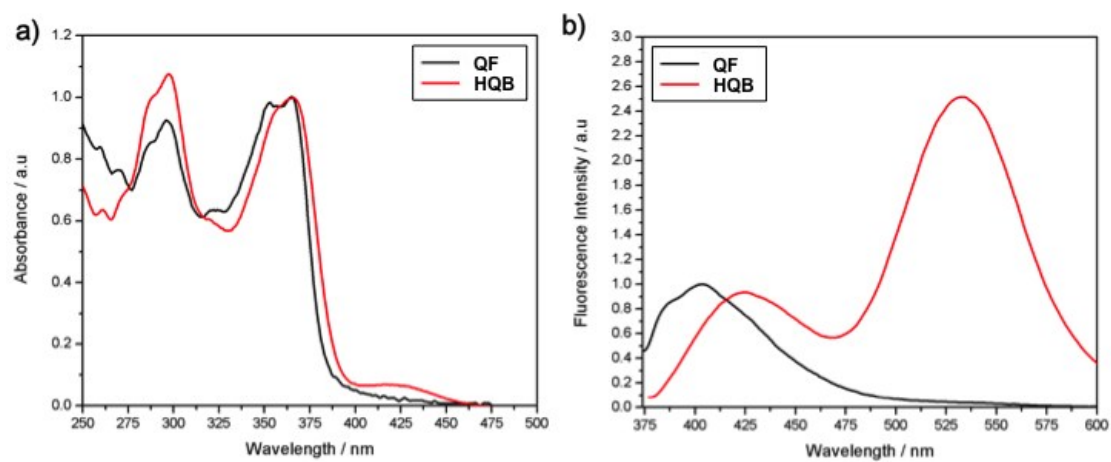


**Fig. S 5**  $^{13}\text{C}$ -NMR spectrum of **QF** (100 MHz,  $d_6$ -DMSO)

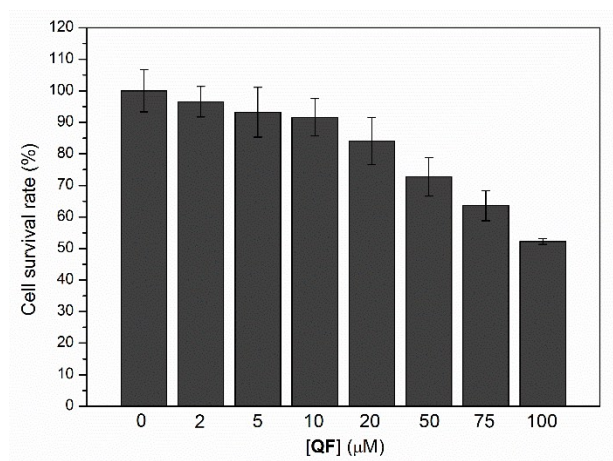
D160115 #24 RT: 0.36 AV: 1 NL: 1.34E8  
T: FTMS + p NSI Full ms [100.00-1000.00]



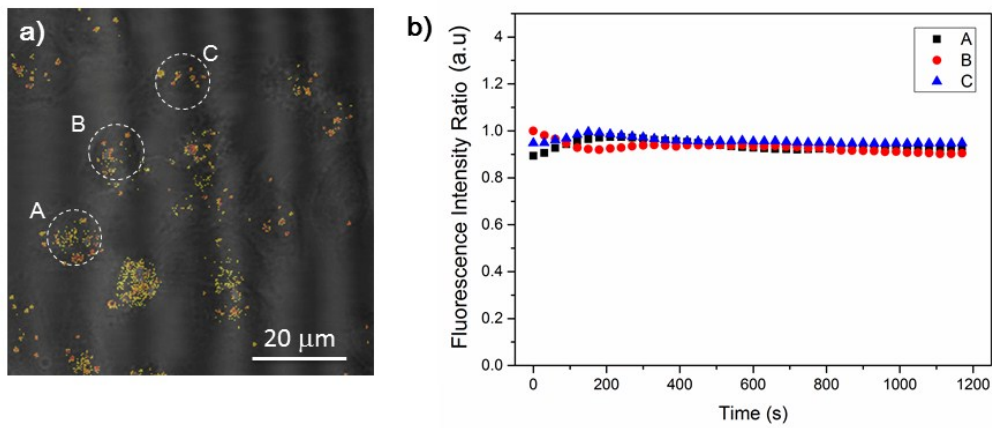
**Fig. S 3** HRMS (MALDI) spectrum of **QF**



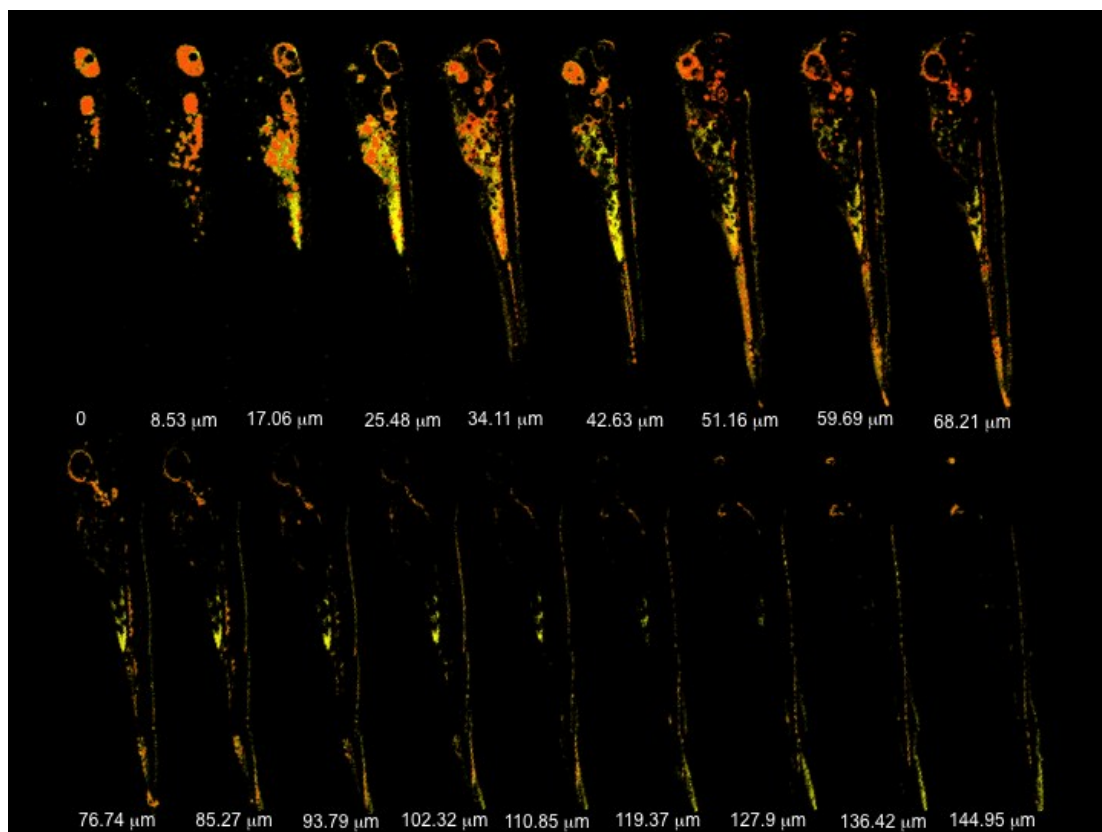
**Fig. S 4.** a) The normalized absorption spectra of **QF** and **HQB**; b) The fluorescence spectra of 10  $\mu\text{M}$  **QF** and **HQB** under one-photon excitation mode. All spectra were determined in a phosphate buffer (50 mM, pH 7.4, 0.9% NaCl, 25  $\mu\text{M}$  CTAB).



**Fig. S 5** Cell survival rate of control groups (without **QF**) and experimental group (with 2, 5, 10, 20, 50, 75, and 100 μM of **QF**). All groups contain 2% DMSO in 100 μL DMEM).



**Fig. S 6** a) TPM images of HeLa cells labeled with 20 μM QF for 30 min and further incubated with NaF for 1 h. The color reflects the ratio of fluorescence intensity ( $F_{\text{green}}/F_{\text{blue}}$ ). b) Two-photon fluorescence intensity ratio from circle A-C as a function of time. The images were collected with 30 sec intervals for the duration of 30 min under *xyt* mode. Scale bar: 20 μm.



**Fig. S 7** Separate images at different z-axis depth of the QF-labeled Zebrafish treated with NaF. The fluorescence intensity ratio ( $F_{\text{green}}/F_{\text{blue}}$ ) was reflected by color. Scale bar: 500  $\mu\text{m}$ .