Supporting Information to
Near-infrared probes based on fluorinated Si-rhodamine for live cell image

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1. General Information

Materials

Pd_2dba_3, XPhOS and Cs_2CO_3 were purchased from BeiJing Greenchem Technology Co., Ltd; Azetidine and tert-BuLi were purchased from Beijing Zhongsheng Huateng Technology Co., Ltd. Trifluoromethanesulfonic anhydride were purchased from Energy Chemical Co., Ltd; Bromobenzene, 2-Bromotoluene and 2-Bromobenzotrifluoride were purchased from J&K Scientific Co., Ltd. Phosphate buffer saline (PBS) was purchased from Life Technologies Co., Ltd. Solvents were either employed as purchased from Tianjin Kemiou Chemical Reagent Co., Ltd or dried according to procedures described in the literature. Deionized water was obtained from a Milli-Q water purification system (Millipore). Dichloromethane were dried by standard procedures, and freshly distilled before each experiment Tetrahydrofuran and 2-Methyltetrahydrofuran was dried with sodium, stored over sodium benzophenone ketyl, and freshly distilled before each experiment. All glassware was oven-dried prior to use when water- and/or air-sensitive reagents were used. The synthetic steps were performed under ambient atmosphere unless stated otherwise.

All reactions were monitored by thin-layer chromatography (TLC) on gel F254 plates. Flash chromatography was carried out on silica gel (200-300 mesh; Yantai

City Chemical Industry Research Institute, Yantai, China). Chemical shifts of ^{1}H-NMR and ^{13}C-NMR spectra are reported in ppm at room temperature using $CDCl_3$ as solvent, tetramethylsilane as internal standard unless indicated otherwise. Abbreviations used for splitting patterns are s = singlet, d = dublett, t = triplet, qui = quintet, m = multiplet. Mass spectra were carried out using Agilent LC/MSD XCT Trap. UV/Vis spectra were recorded on a Shimadzu WV-2550 spectrophotometer. Fluorescence spectra were recorded on a Shimadzu RF-5301 fluorescence spectrophotometer. The fluorescence lifetimes were recorded on an Edinburgh Analytical Instruments FLS980 spectrometer, equipped with a supercontinuum ultrafast fiber lasers (Fianium), using the time correlated single photon-counting (TCSPC) method. The fluorescence quantum yields were determined by using Cyanine 5 monoacid ($\Phi = 1$ in DMSO) for **AZSiR** as the reference.

MTT assays were performed to assess the metabolic activity of Hela cells. Hela cells were incubated with 10 μM **AZSiR** probes for 24h. After the designated time intervals, the wells were washed twice with PBS buffer and freshly prepared MTT (10μL, 5mg/mL) solution in culture medium was added into each well. The MTT medium solution was carefully removed after 3h incubation in the incubator. DMSO (100μL) was then added into each well and gently shaken for 10 min at room temperature to dissolve all the precipitate formed. The absorbance of MTT at 490nm was monitored by the ELX-800 microplate reader (ELISA Reader). Cell viability was expressed by the ratio of the absorbance of the cells incubated with **AZSiR** probes solution. Each result is an average of data from 3 wells; 100% viability was determined using untreated cells.

2. UV-Vis absorption and fluorescent spectroscopy of AZSiR

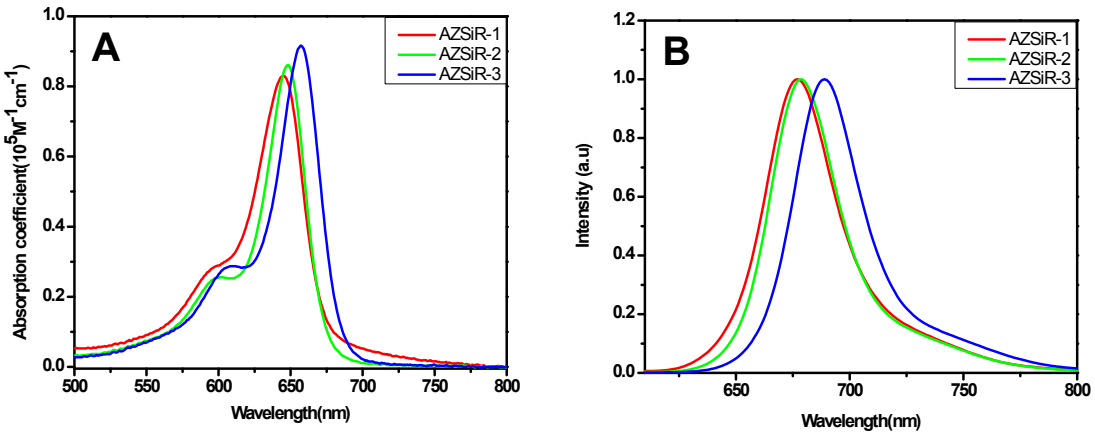

Figure S1 UV-Vis absorption and fluorescent spectroscopy of **AZSiR** in PBS+1% DMSO solution.

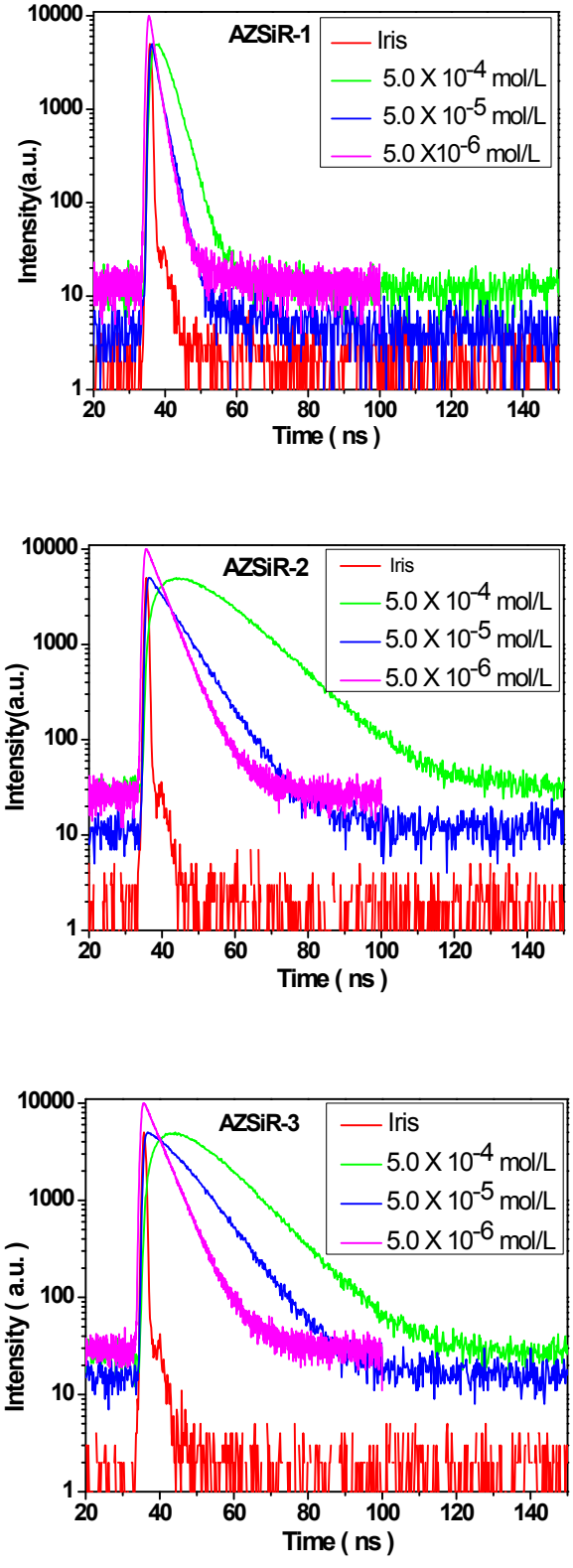

Figure S2 Time resolved fluorescence spectroscopy of **AZSiR** probes in different concentration dichloromethane solution

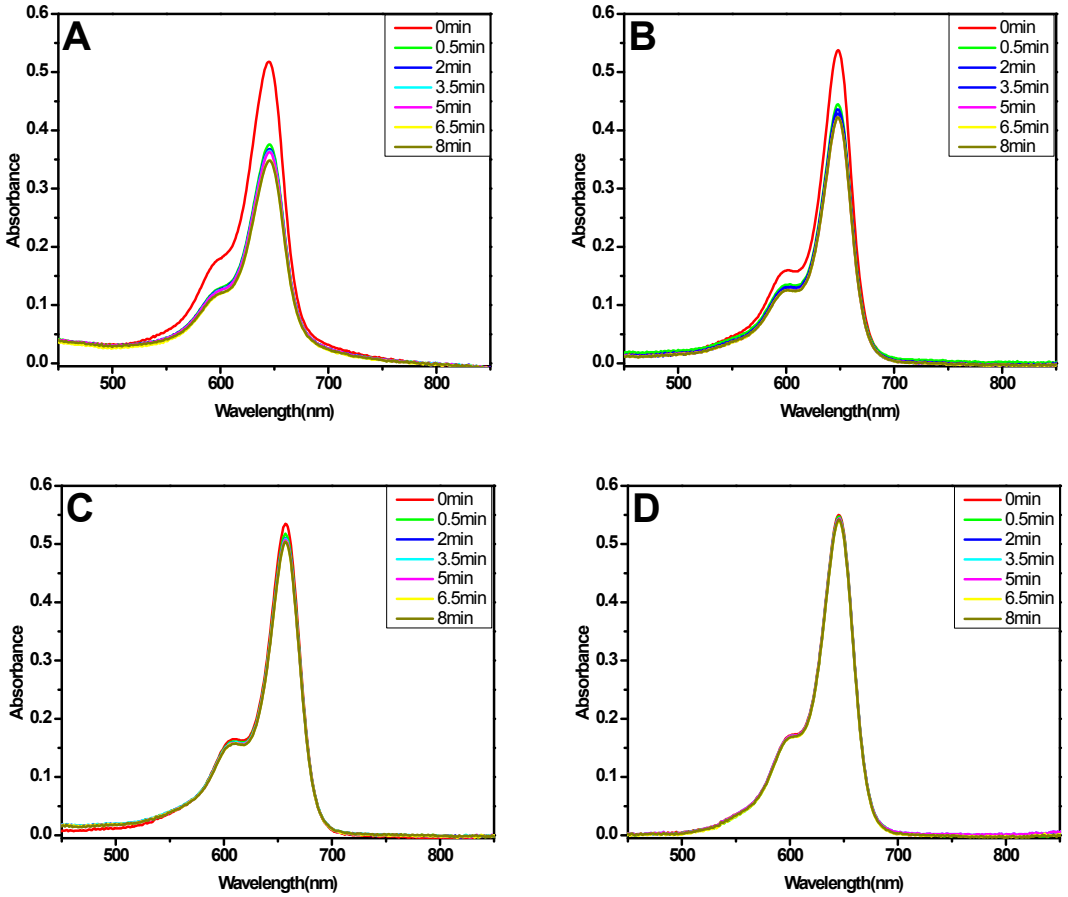

Figure S3 Time-dependent UV-Vis absorption spectroscopy of AZSiR probes with 0.5 uM of cysteine in PBS+1% DMSO solution. (A) AZSiR-1. (B) AZSiR-2. (C) AZSiR-3. (D) Cy5

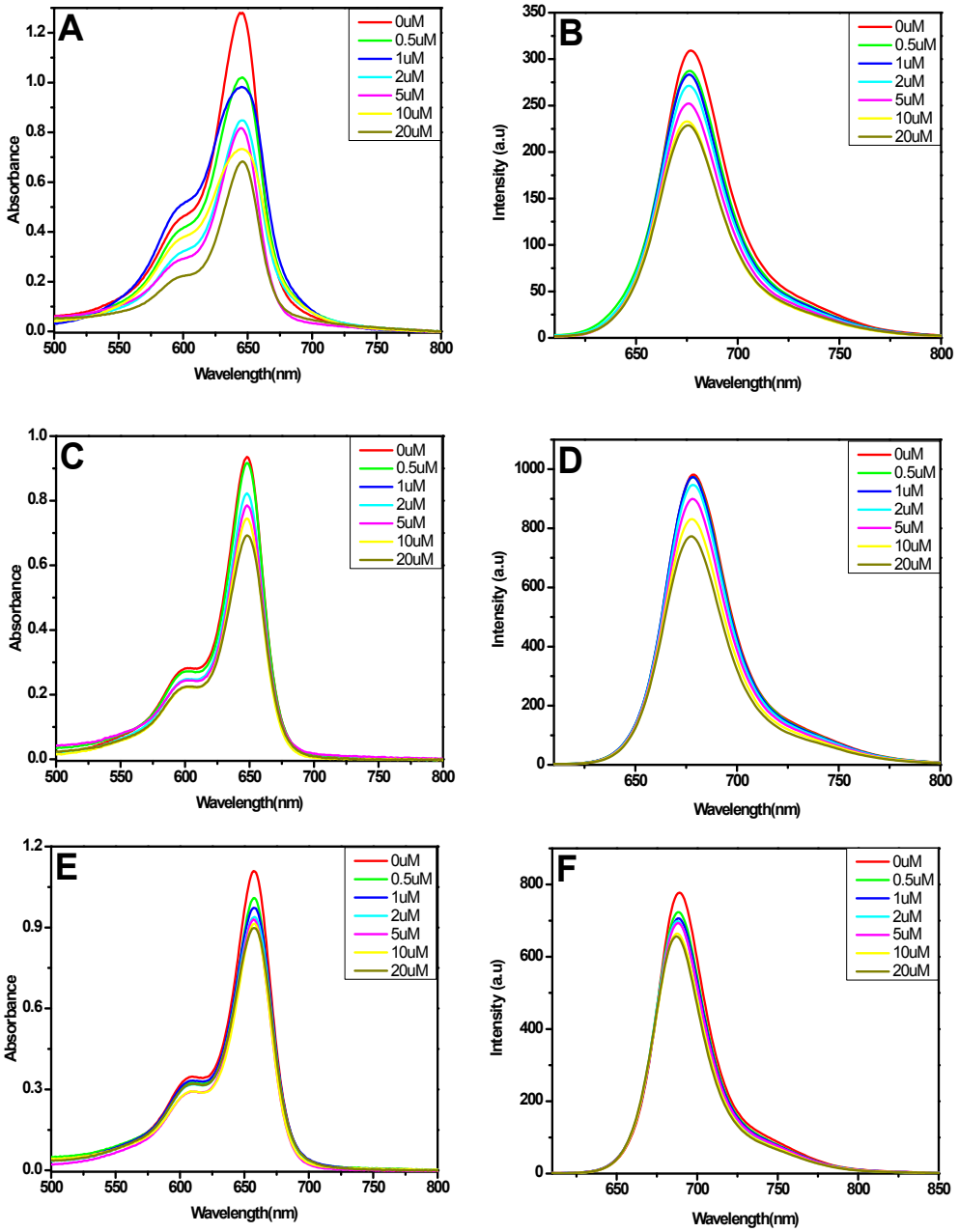

Figure S4 UV-Vis absorption and fluorescent spectroscopy of AZSiR probes with different concentration cysteine. UV-Vis absorption spectroscopy: (A) AZSiR-1. (C) AZSiR-2. (E) AZSiR-3. Fluorescent spectroscopy: (B) AZSiR-1. (D) AZSiR-2. (F) AZSiR-3.

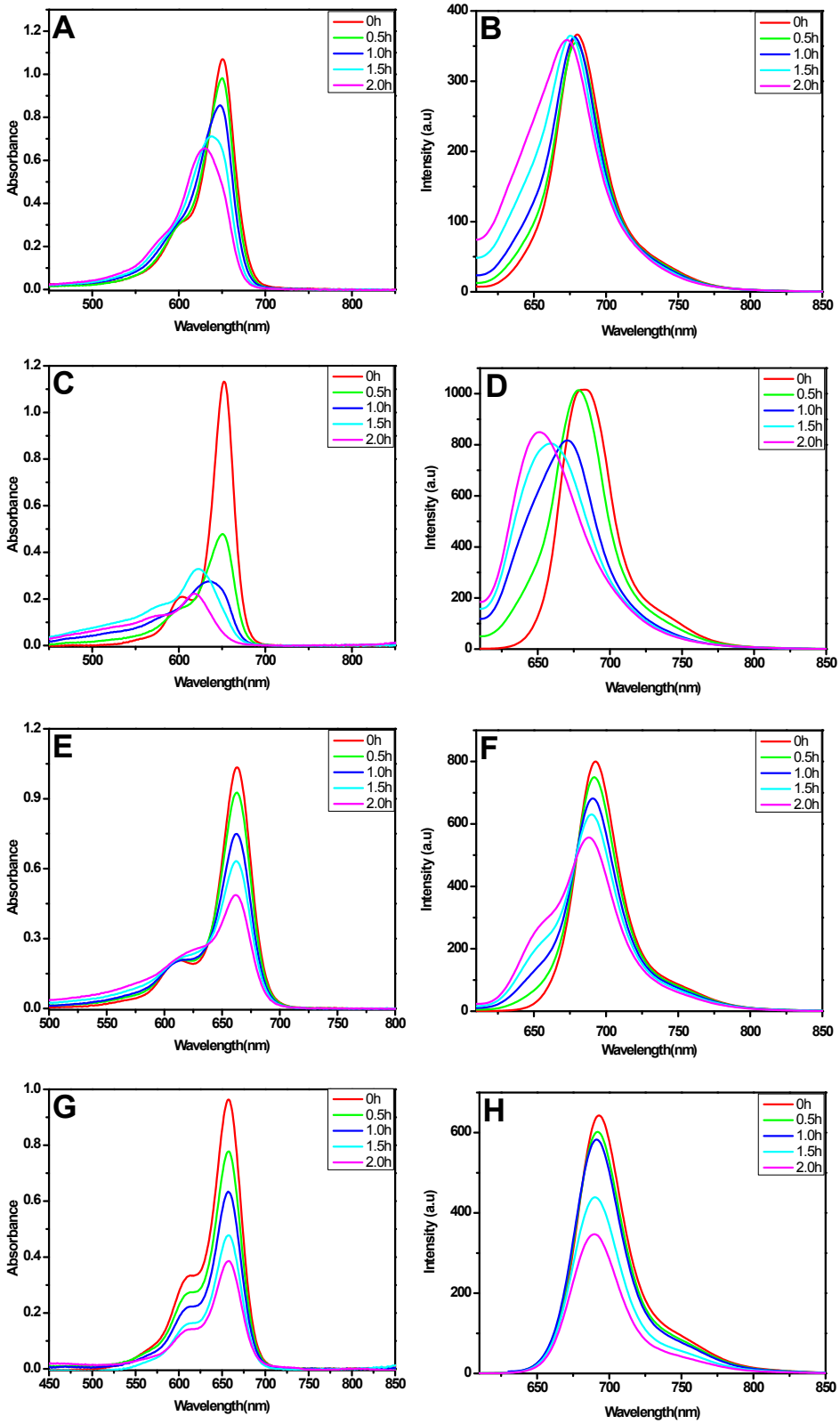

Figure S5 Time-dependent absorption of **AZSiR** probes in dichloromethane with a concentration of 10^{-5} mol/L under continuous irradiation with 660 nm LED laser. UV-Vis absorption spectroscopy: (A) AZSiR-1. (C) AZSiR-2. (E) AZSiR-3. (G) Cy5. Fluorescent spectroscopy: (B) AZSiR-1. (D) AZSiR-2. (F) AZSiR-3. (H) Cy5.

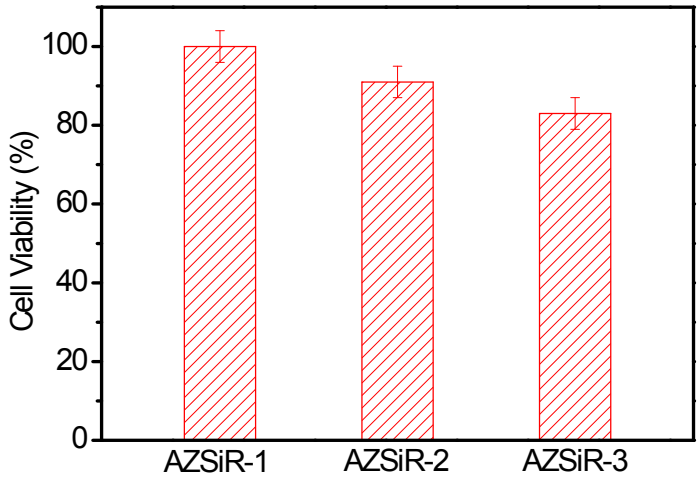

Figure S6 In vitro viability of Hela cells treated with **AZSiR** probes solution at 10 μM for 24 h

Part B: ^{1}H-NMR spectrum, ^{13}C NMR spectrum and MALDI-TOF spectrum

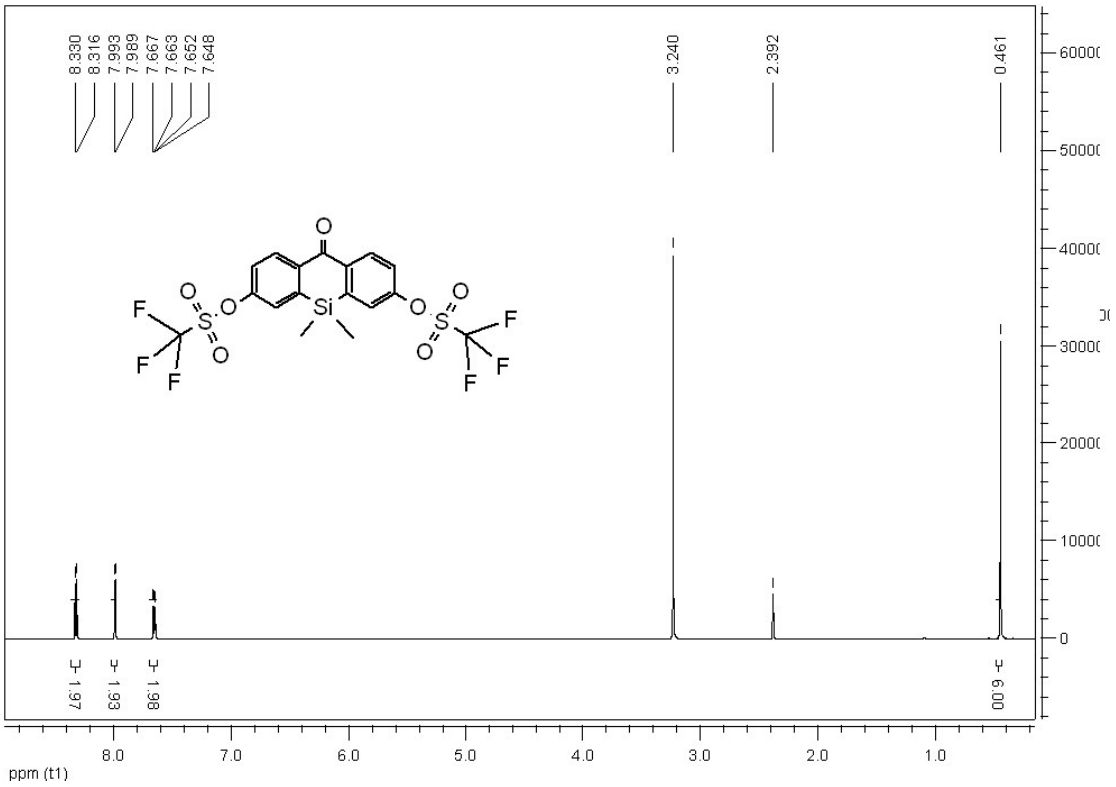

Figure S7. ^{1}H NMR spectrum ($CDCl_3$, 600 MHz, 20 ℃) of compound 2

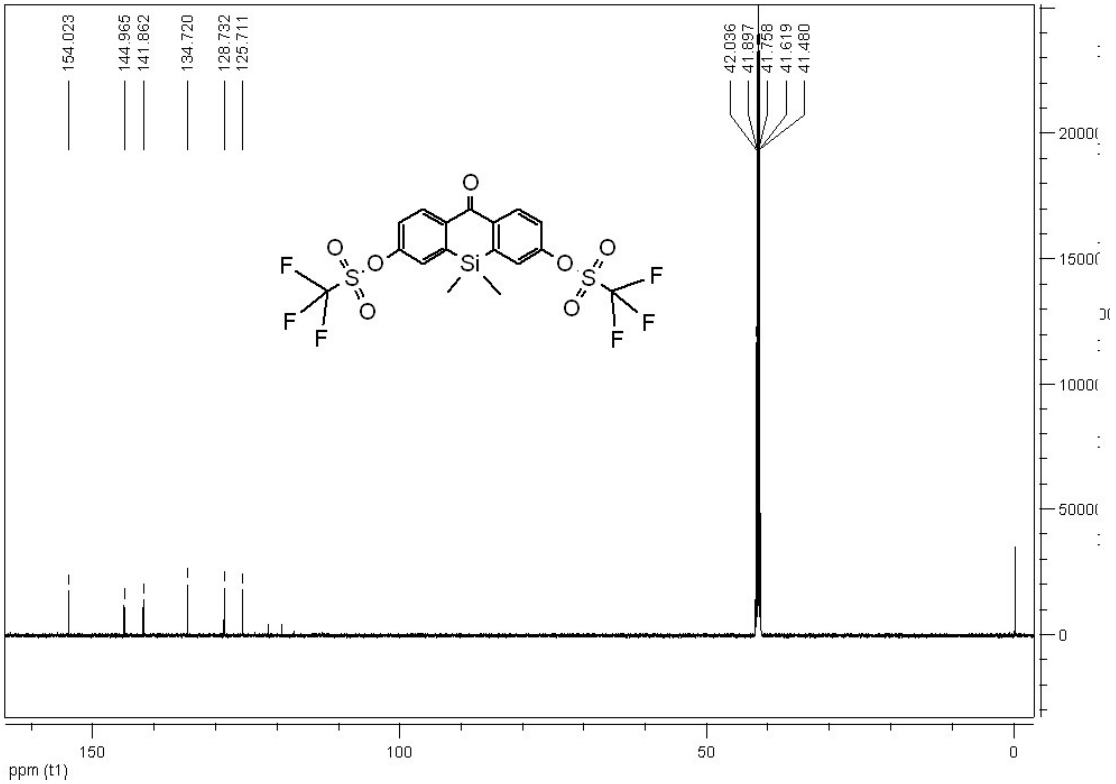

Figure S8. ^{13}C NMR spectrum ($CDCl_3$, 150 MHz, 20 ℃) of compound 2

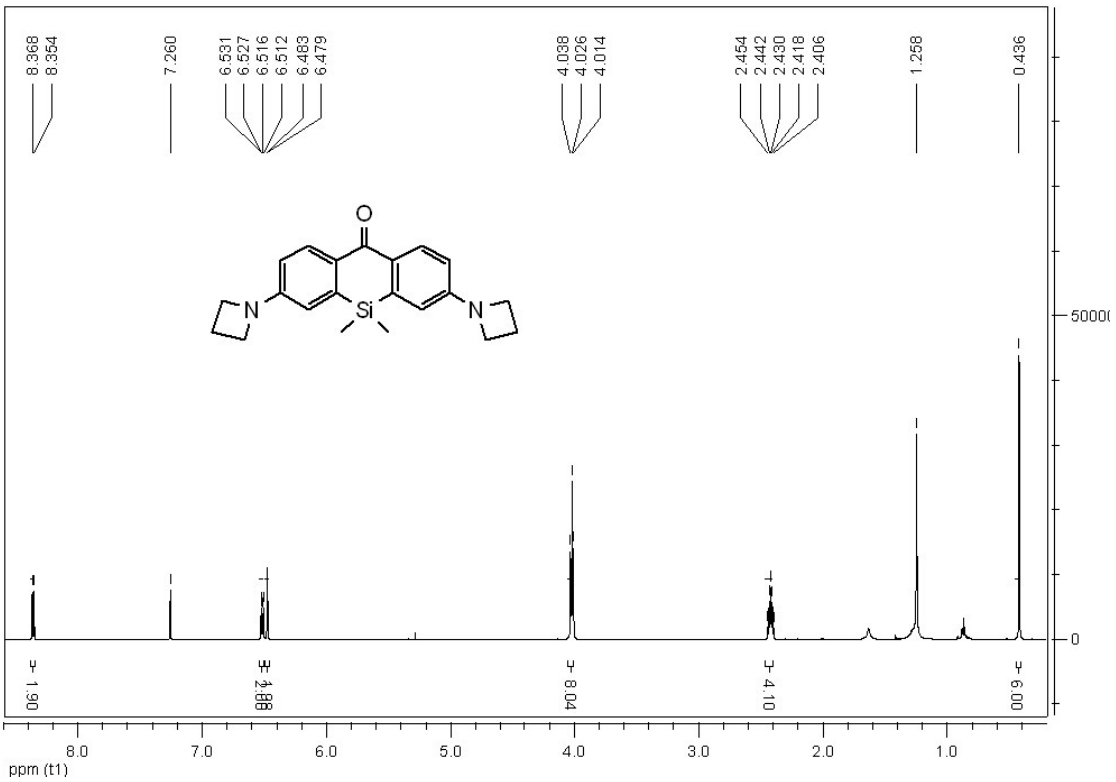

Figure S9. ^{1}H NMR spectrum ($CDCl_3$, 600 MHz, 20℃) of compound 3

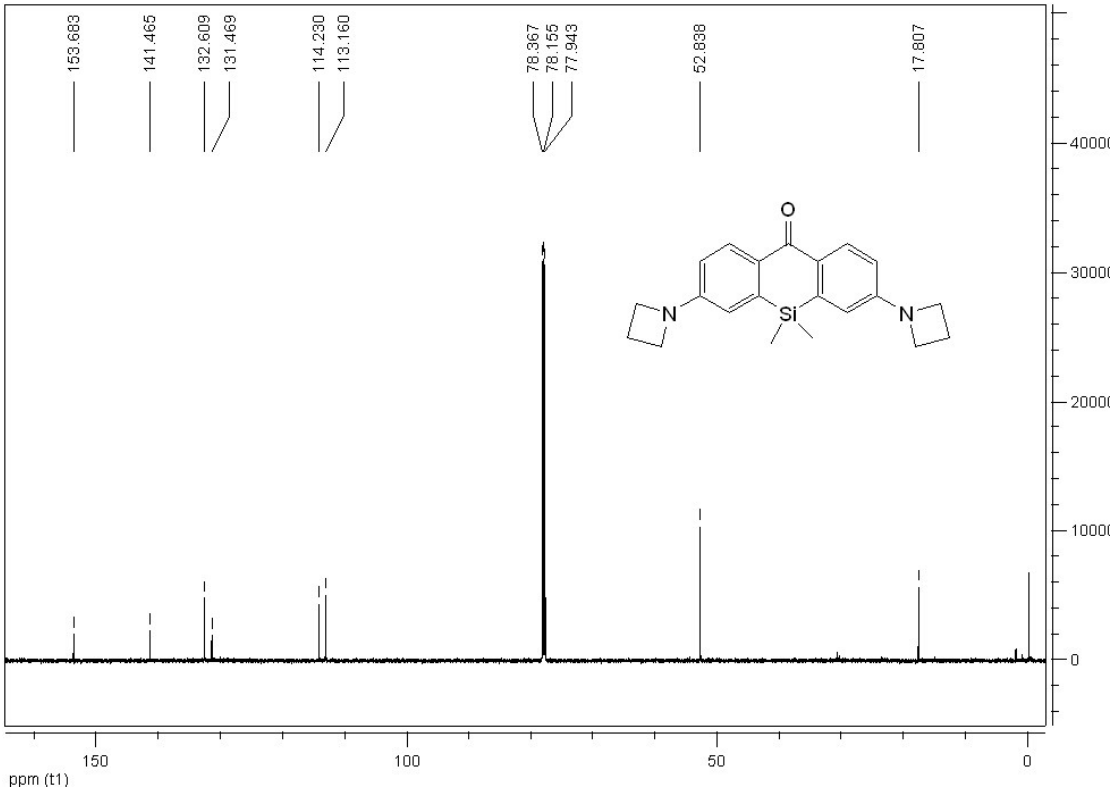

Figure S10. ^{13}C NMR spectrum ($CDCl_3$, 150 MHz, 20 ℃) of compound 3

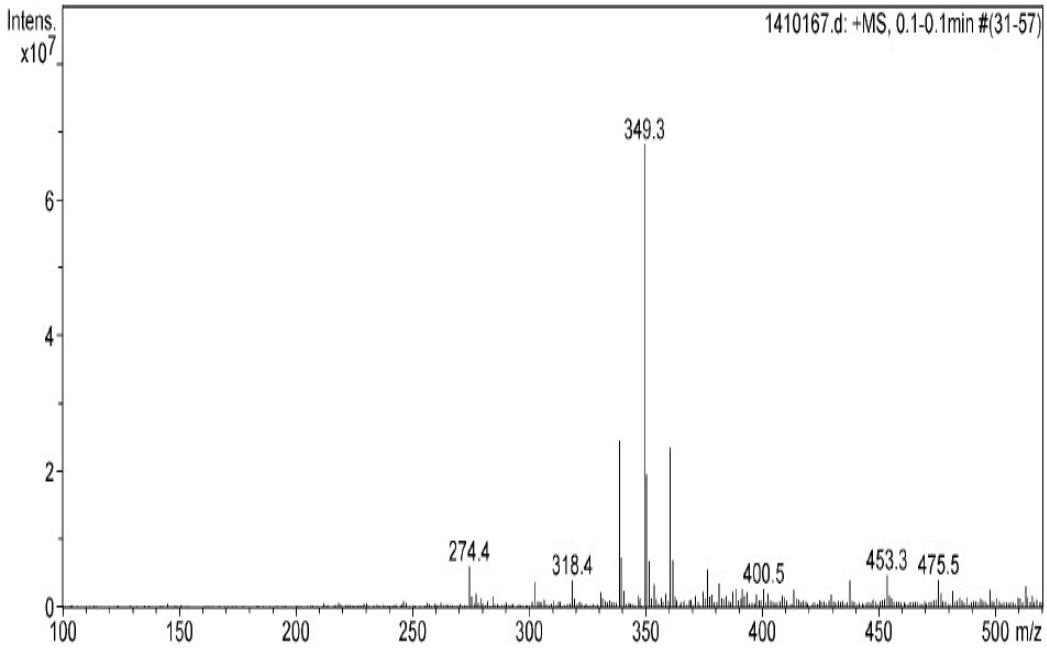

Figure S11. MALDI-TOF spectrum of compound 3

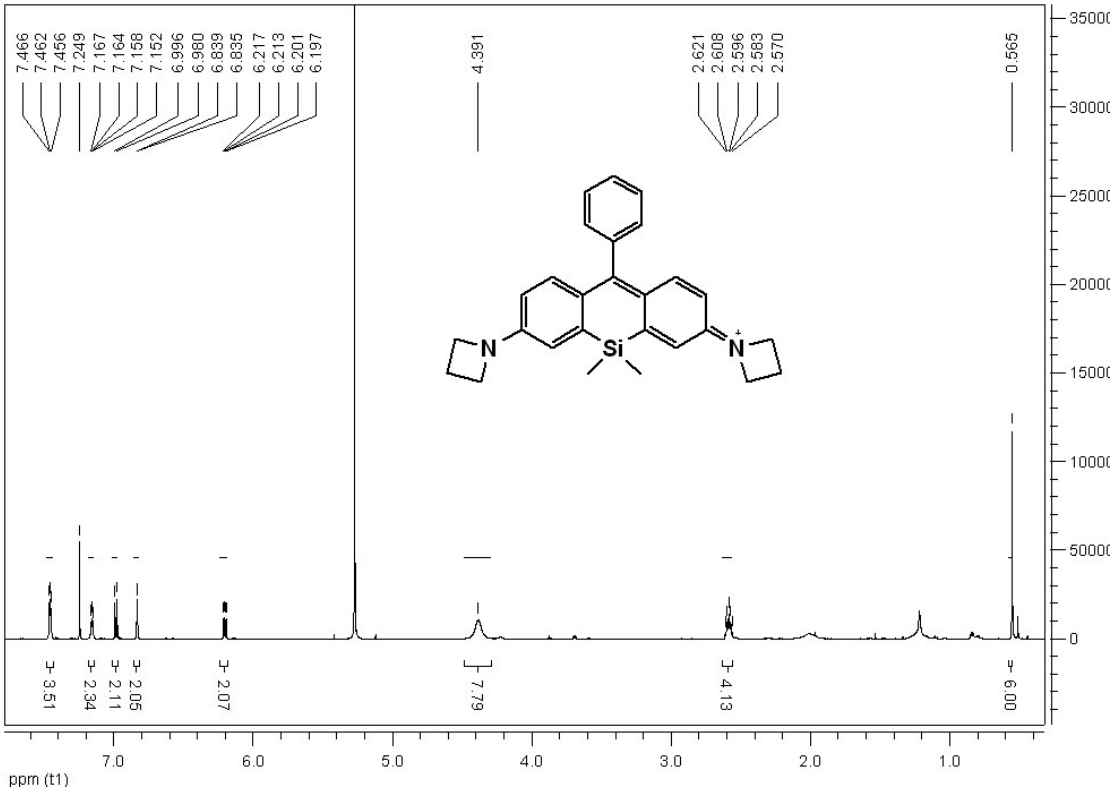

Figure S12. ^{1}H NMR spectrum ($CDCl_3$, 600 MHz, 20 ℃) of AZSiR-1

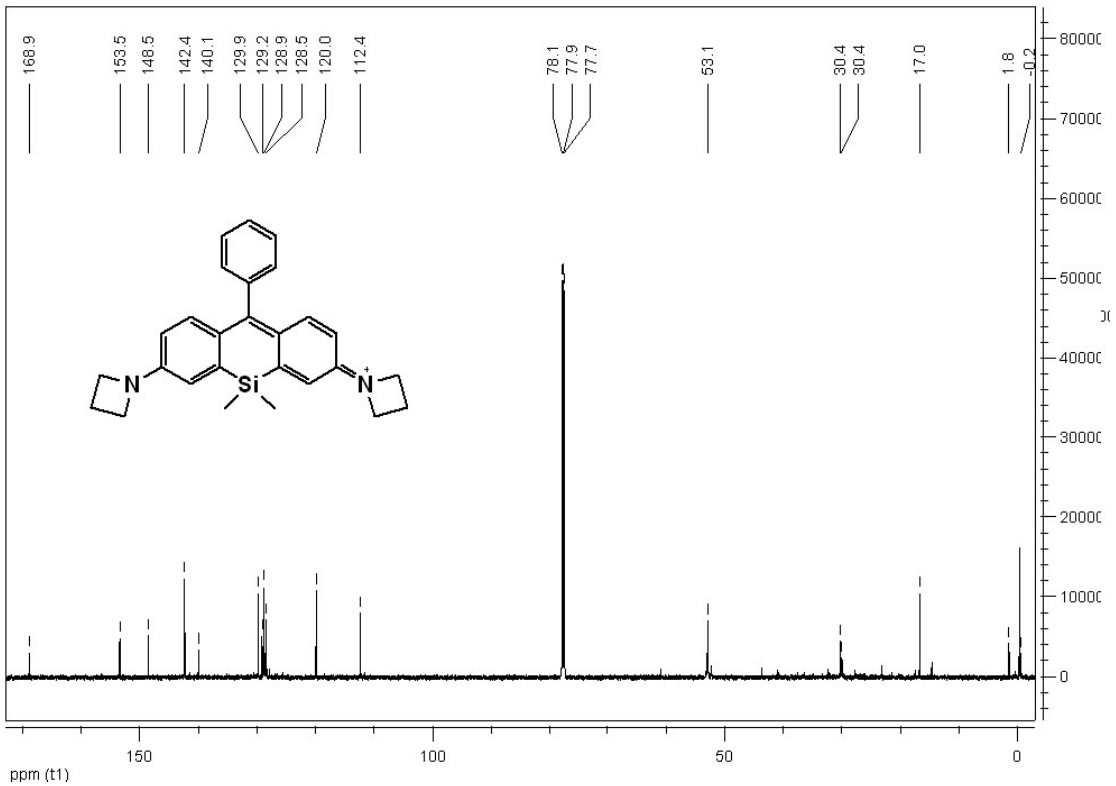

Figure S13. ^{13}C NMR spectrum ($CDCl_3$, 150 MHz, 20 ℃) of AZSiR-1

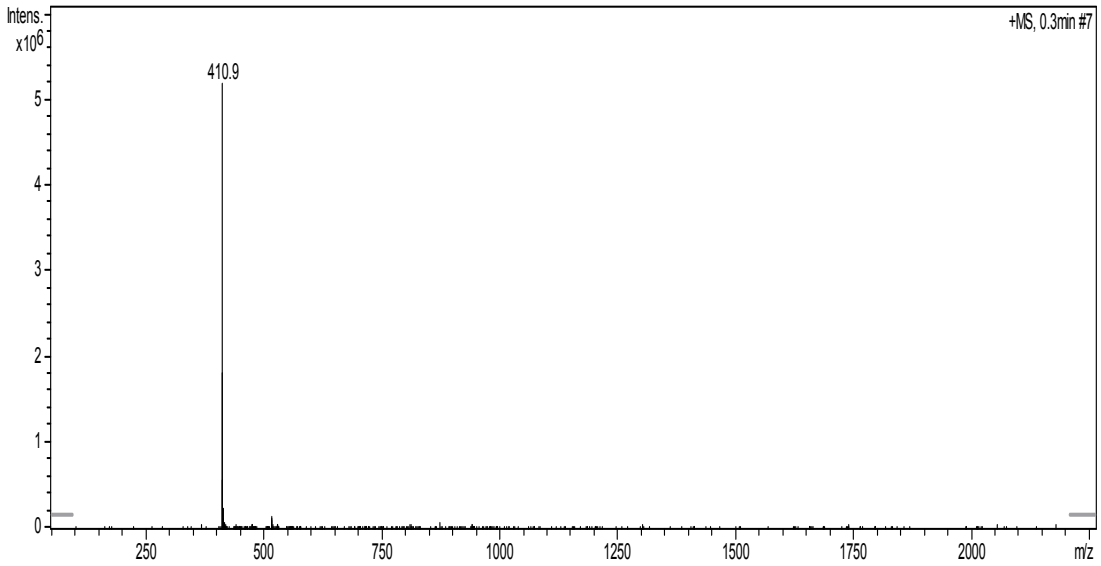

Figure S14. ESI mass spectrum of AZSiR-1

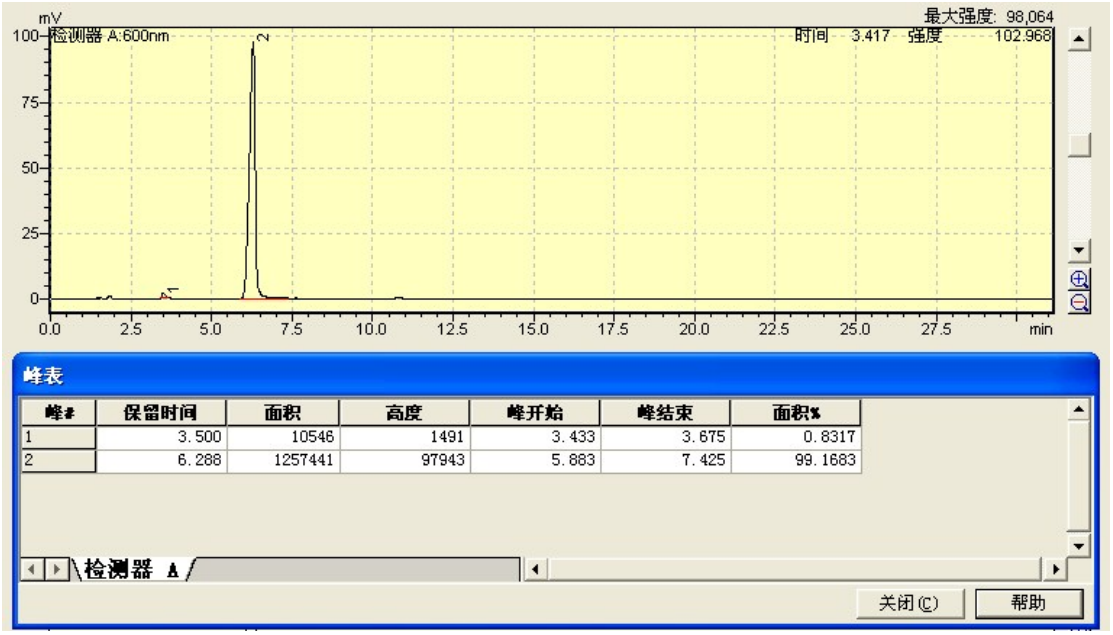

Figure S15. HLCP analysis of AZSiR-1

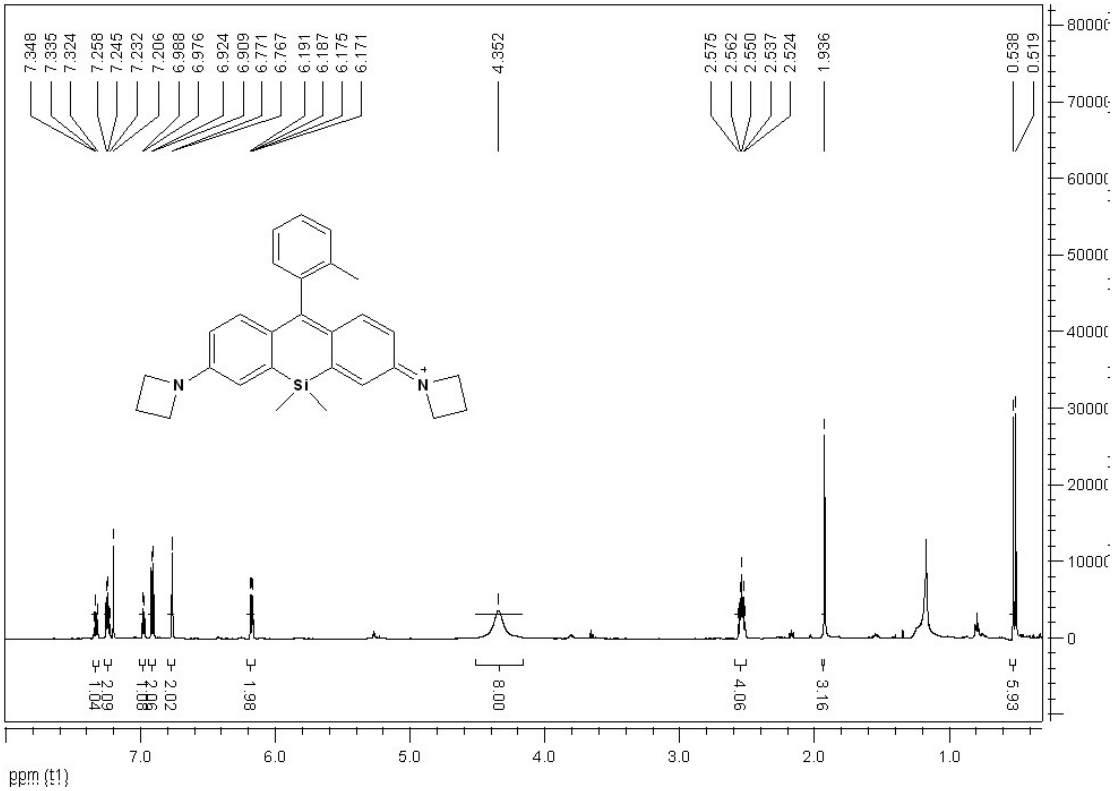

Figure S16. ^{1}H NMR spectrum ($CDCl_3$, 600 MHz, 20 ℃) of AZSiR-2

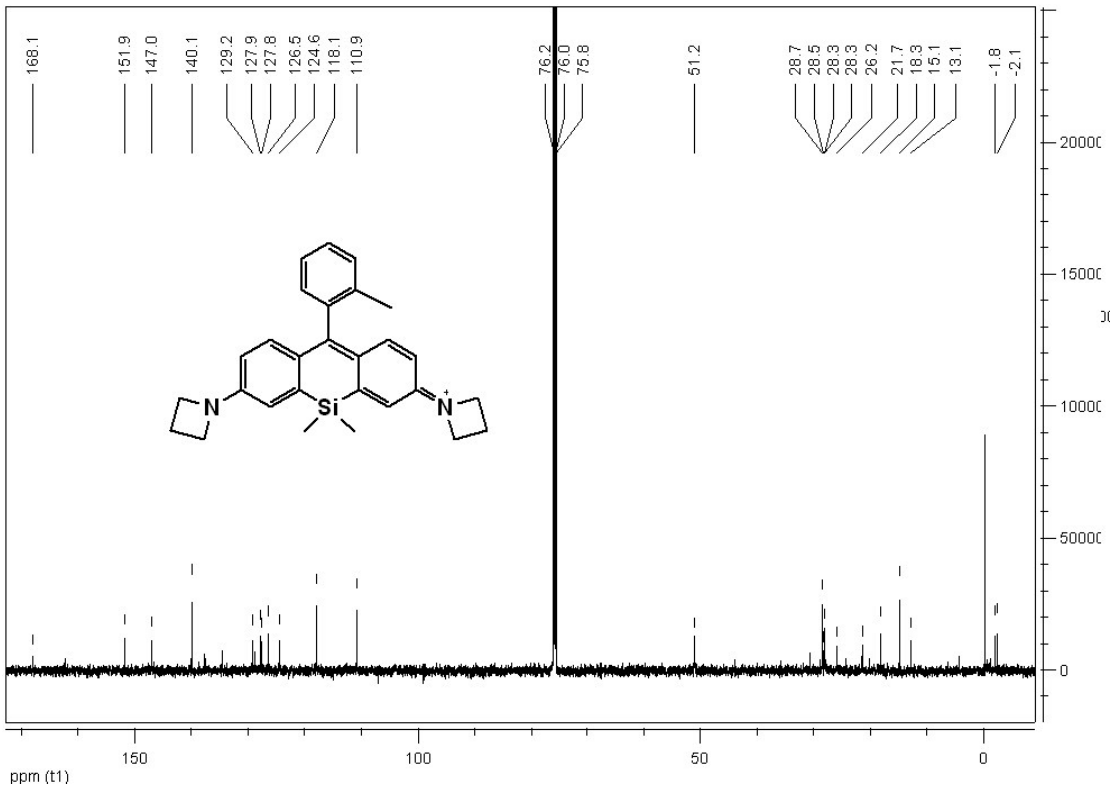

Figure S17. ^{13}C NMR spectrum ($CDCl_3$, 150 MHz, 20 ℃) of AZSiR-2

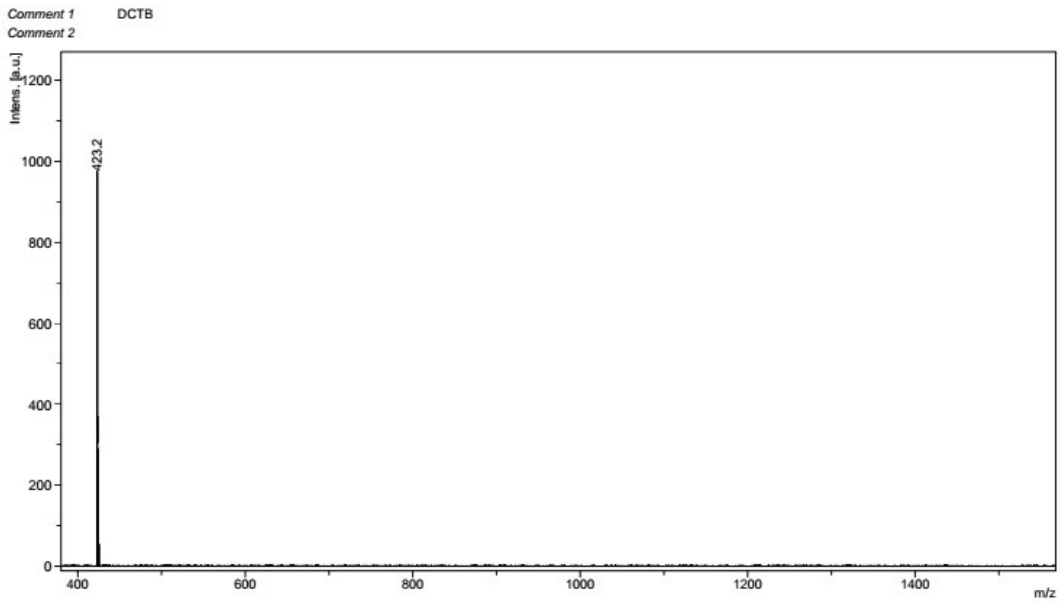

Figure S18. ESI mass spectrum of AZSiR-2

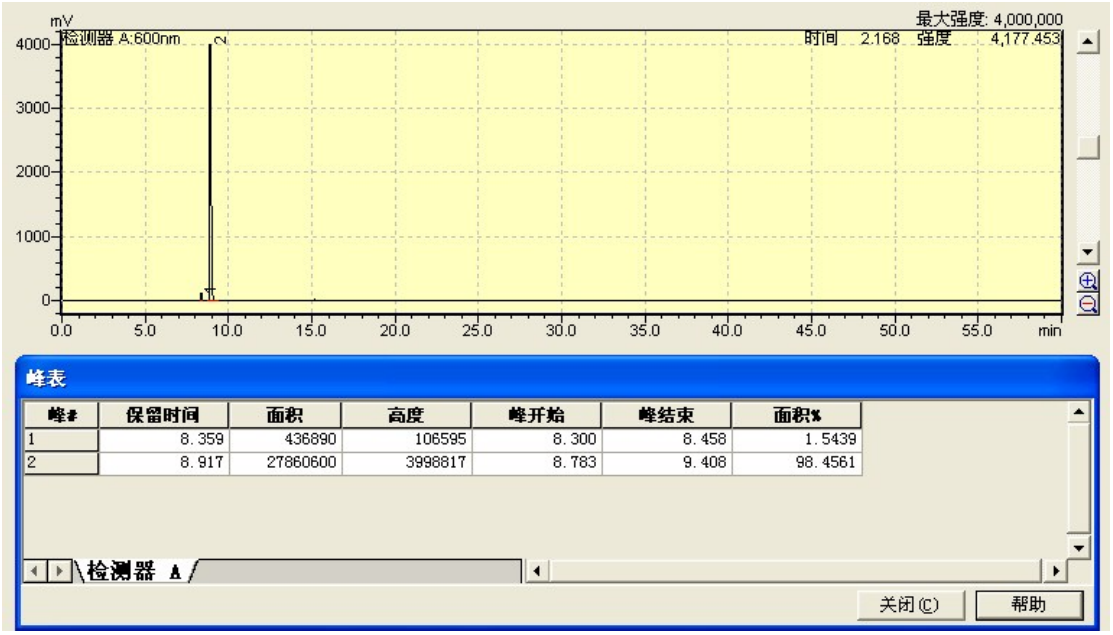

Figure S19. HLCP analysis of AZSiR-2

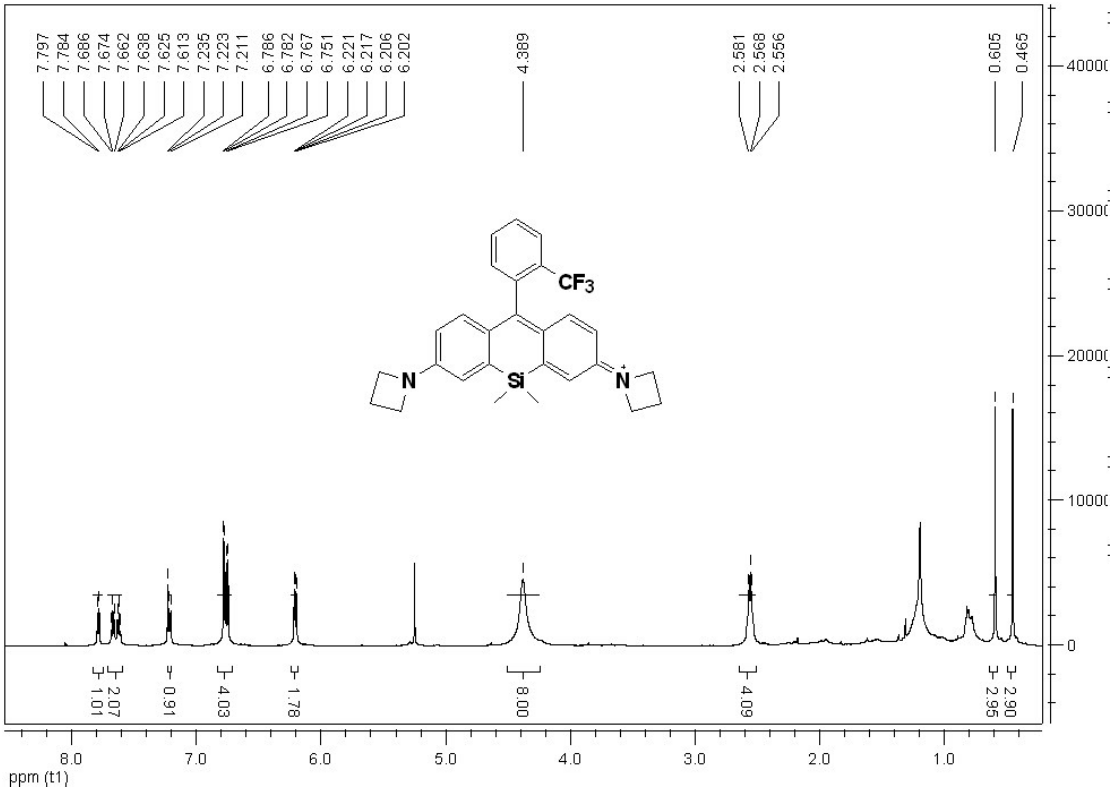

Figure S20. ^{1}H NMR spectrum ($CDCl_3$, 600 MHz, 20 ℃) of AZSiR-3

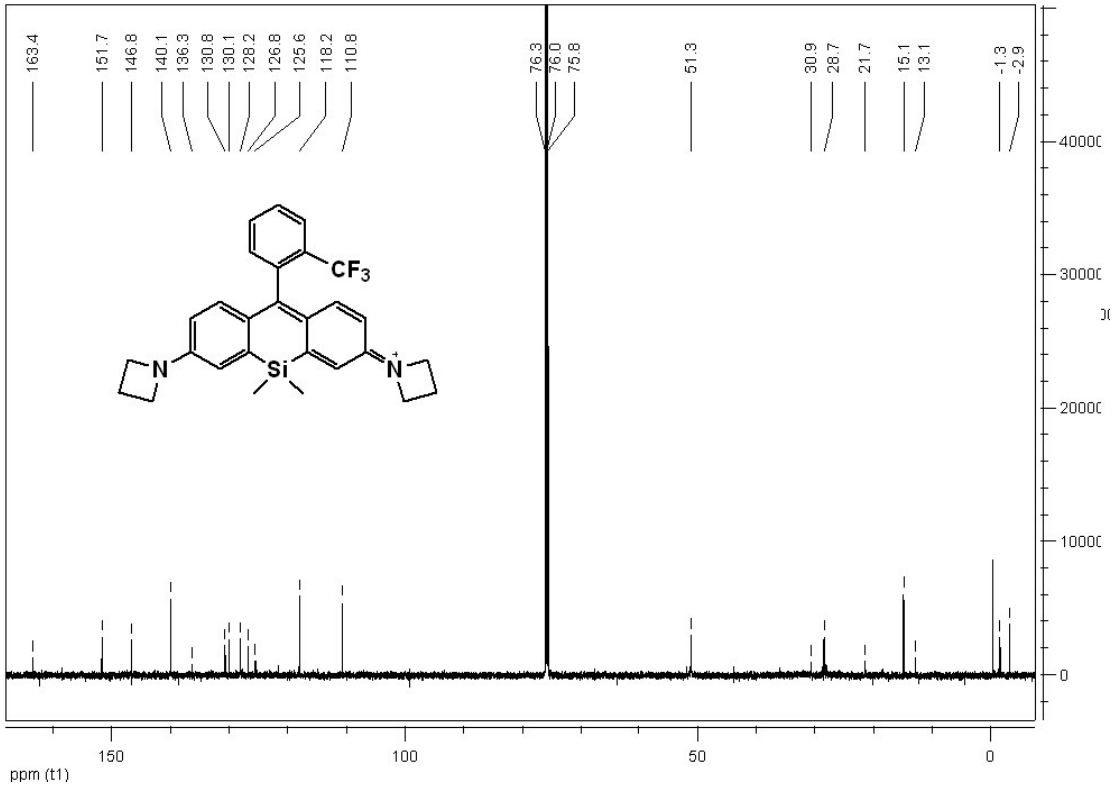

Figure S21 ^{13}C NMR spectrum ($CDCl_3$,150 MHz, 20 ℃) of AZSiR-3

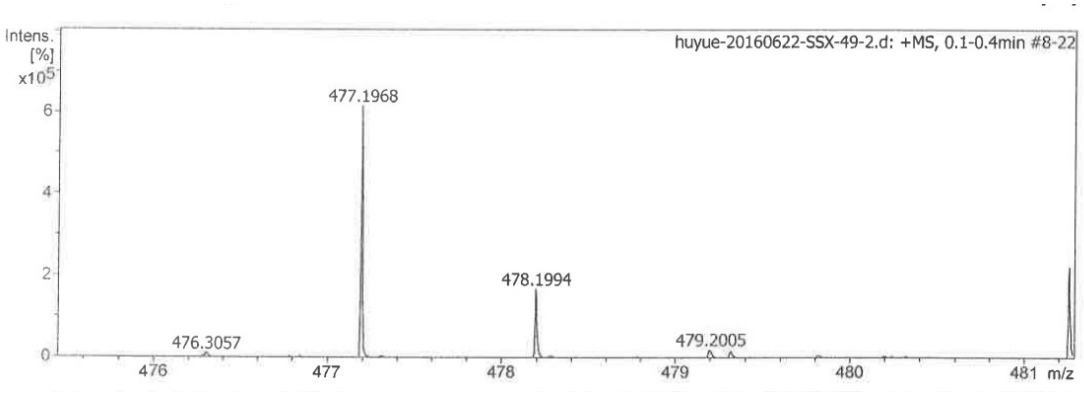

Figure S22 ESI mass spectrum of AZSiR-3

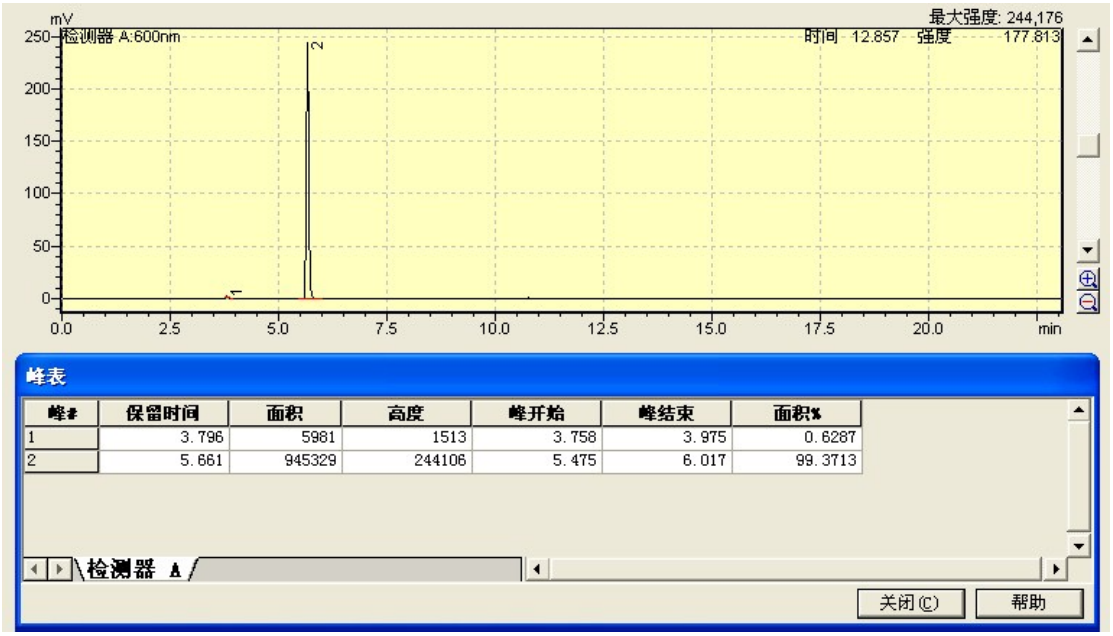

Figure S23 HLCP analysis of AZSiR-3