Electronic Supplementary Material (ESI) for Journal of Materials Chemistry B.

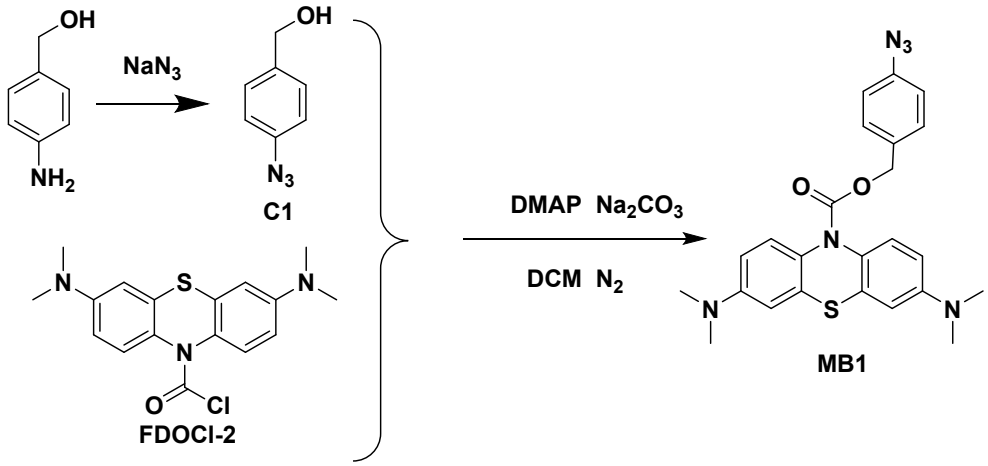
Supporting Information

Molecular isomerization triggered by H_2S to NIR accessible first direct visualization of Ca^{2+}-dependent production in living HeLa cells

Ying Wen, [a] Fangjun Huo, [b] Junping Wang, [a] and Caixia Yin [a]*

[a] Key Laboratory of Chemical Biology and Molecular Engineering of Ministry of Education, Key Laboratory of Materials for Energy Conversion and Storage of Shanxi Province, Institute of Molecular Science, Shanxi University, Taiyuan 030006, China.
[b] Research Institute of Applied Chemistry, Shanxi University, Taiyuan 030006, China.

1. Synthesis routs of MB1



Scheme S1. Synthesis route of **MB1.**

2. The details of characterization of MB1

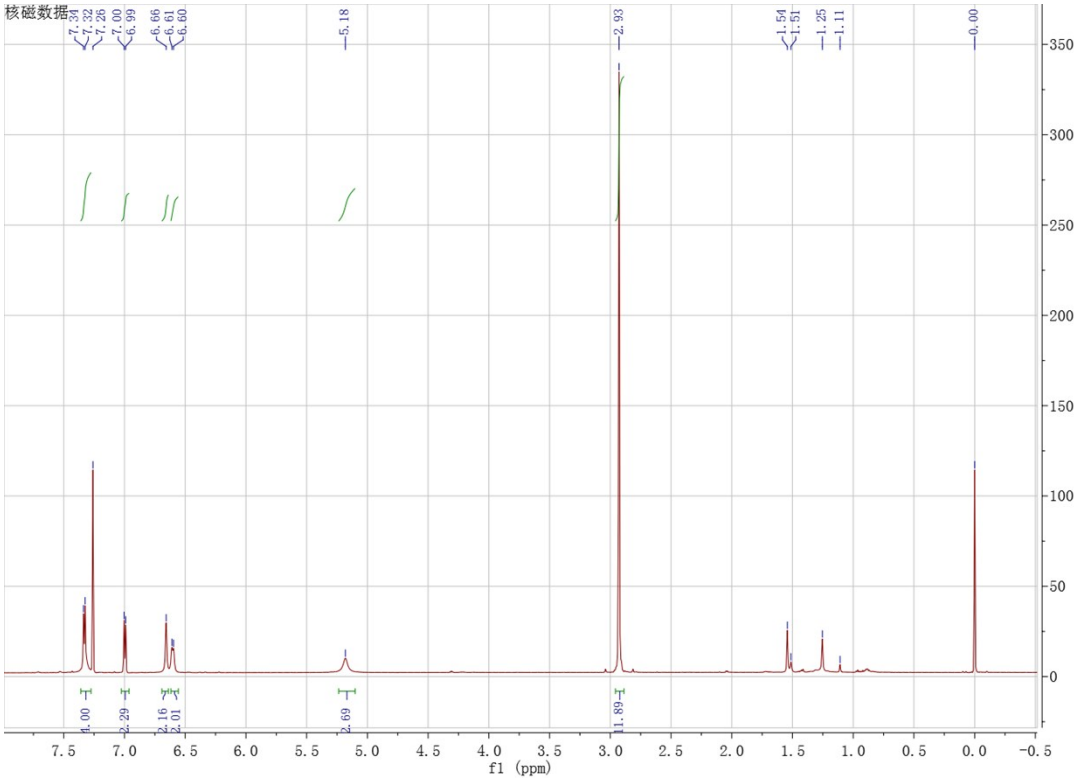


Fig. S1 ^{1}H NMR spectrum of **MB1** in $CDCl_3$. ^{1}H NMR (600 MHz, $CDCl_3$) δ 7.33 (d, J = 7.7 Hz, 4H), 7.00 (d, J = 7.8 Hz, 2H), 6.66 (s, 2H), 6.60 (d, J = 7.8 Hz, 2H), 5.18 (s, 3H), 2.93 (s, 12H).

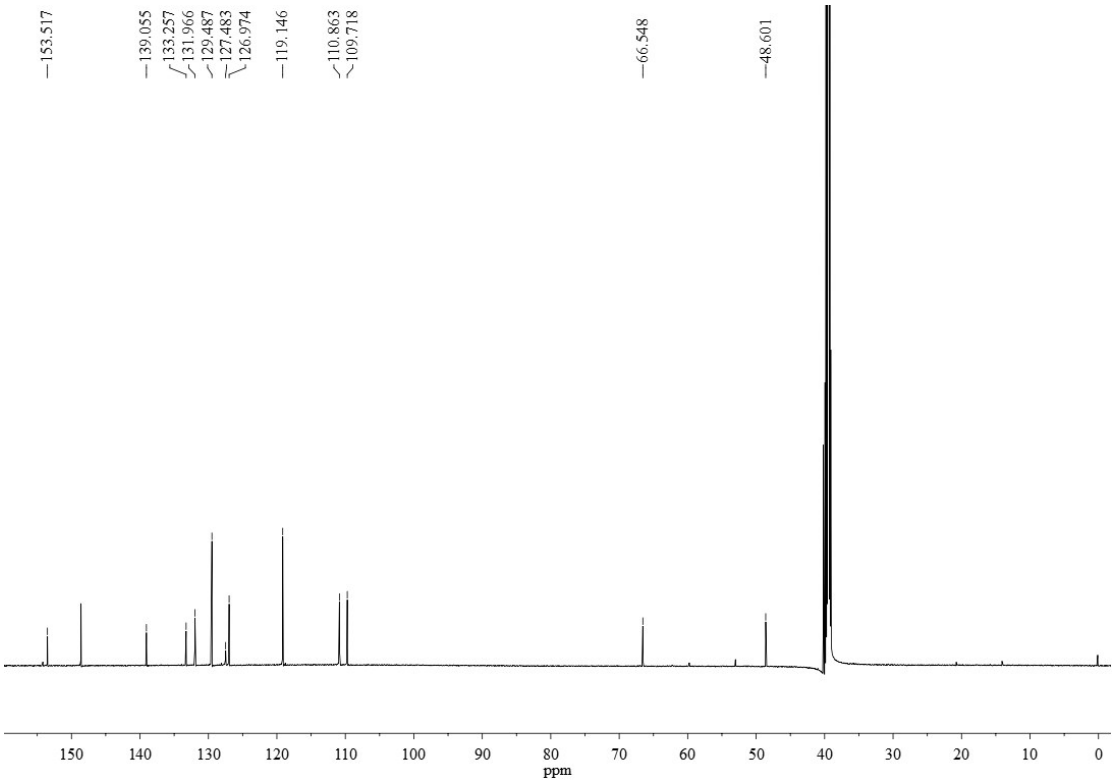


Fig. S2 ^{13}C NMR spectrum of **MB1** in DMSO. ^{13}C NMR (151 MHz, DMSO) δ 153.52 (s), 139.06 (s), 133.26 (s), 131.97 (s), 129.49 (s), 127.48 (s), 126.97 (s), 119.15 (s), 110.86 (s), 109.72 (s), 66.55 (s), 48.60 (s).

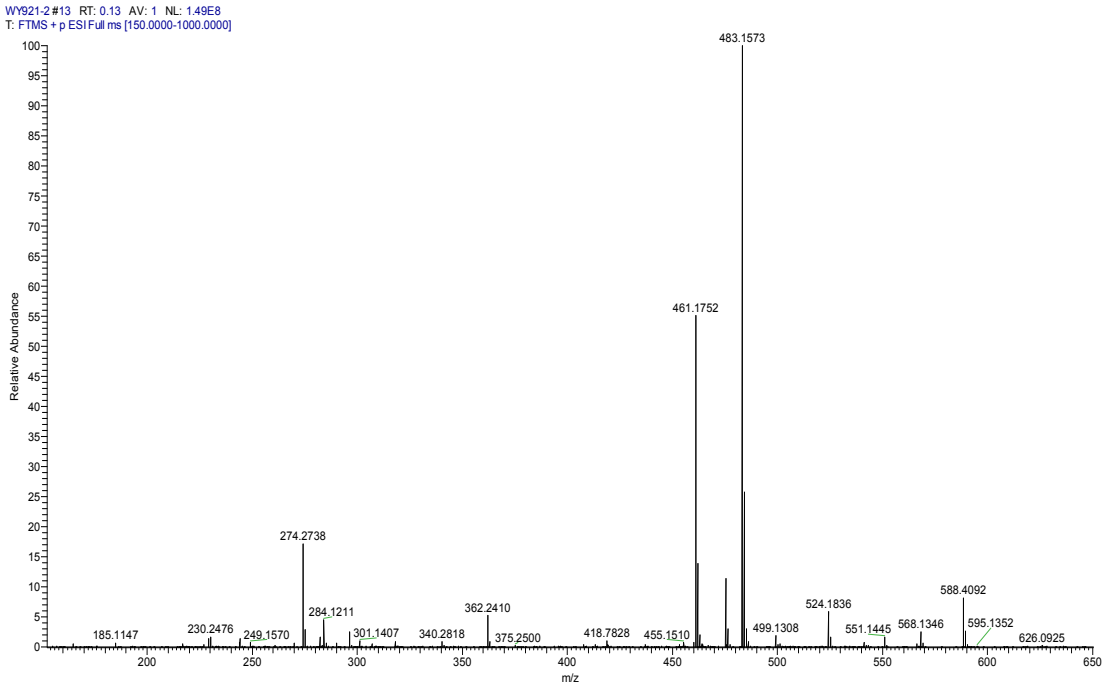


Fig. S3 HR-MS spectrum of **MB1**: calc. for $C_{24}H_{24}N_6O_2SNa^+$ $[M+Na]^+$ 483.1579, found 483.1573.

2. Additional data of MB1

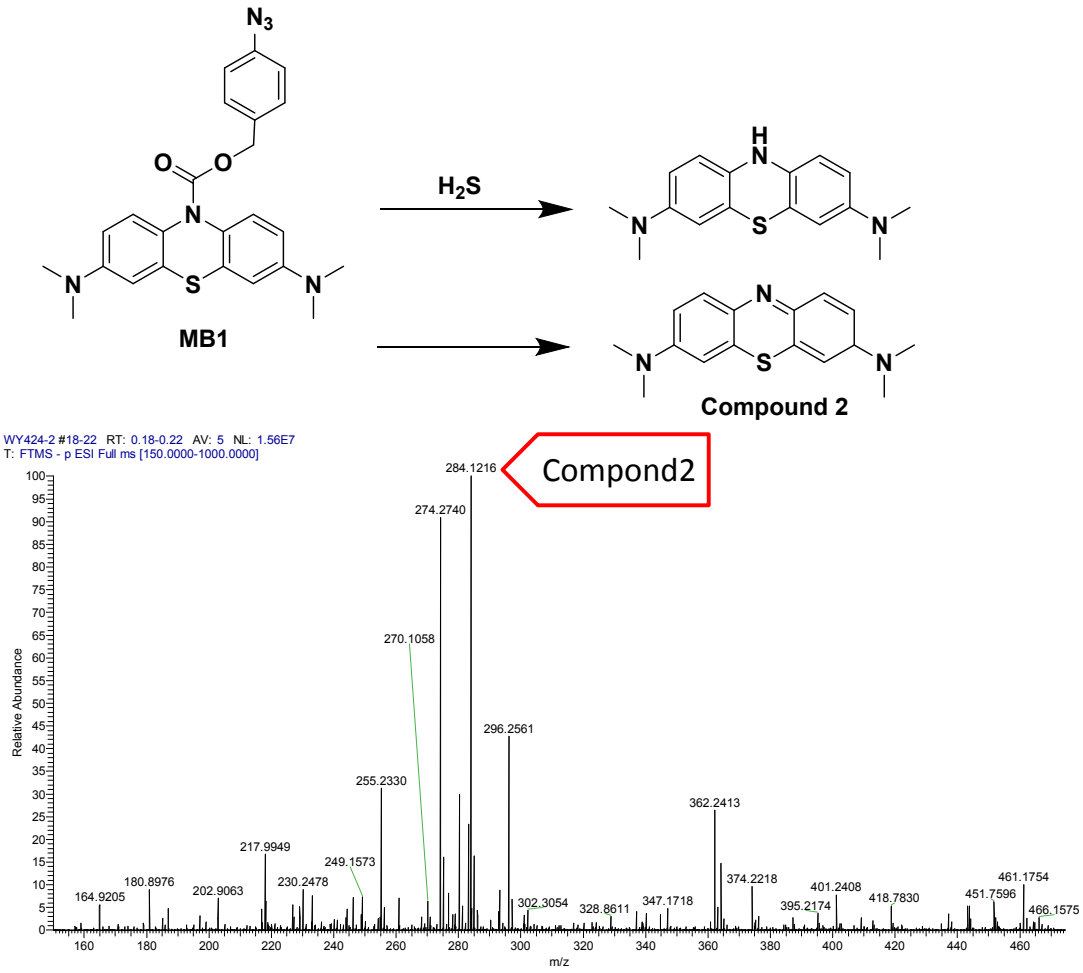


Fig. S4 HR-MS spectrum of the product after **MB1** reacted with H_2S: calcd. for $C_{16}H_{18}N_3S^-$ [M-H]$^-$, 284.1212; found, 284.1216.

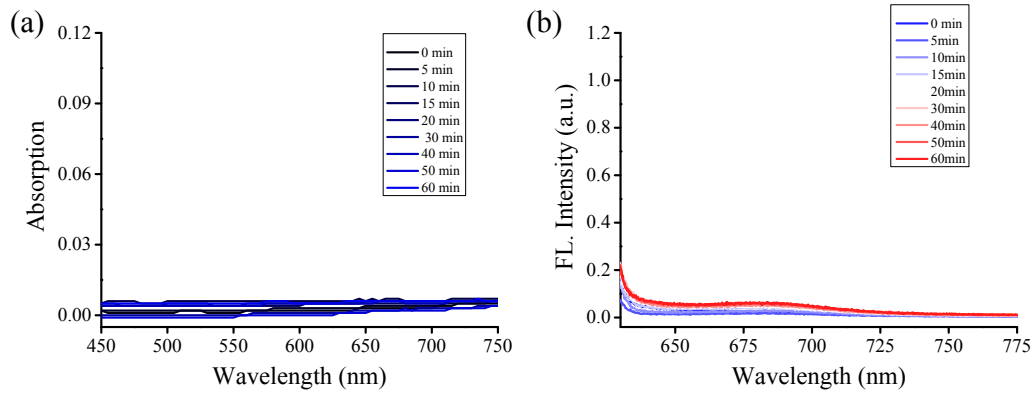


Fig. S5 Absorption (a) and fluorescence (b) spectral changes of **MB1** (5 μM) over time in a mixed solvent of DMSO and PBS (1:1, v/v). λ_{ex} = 600 nm.

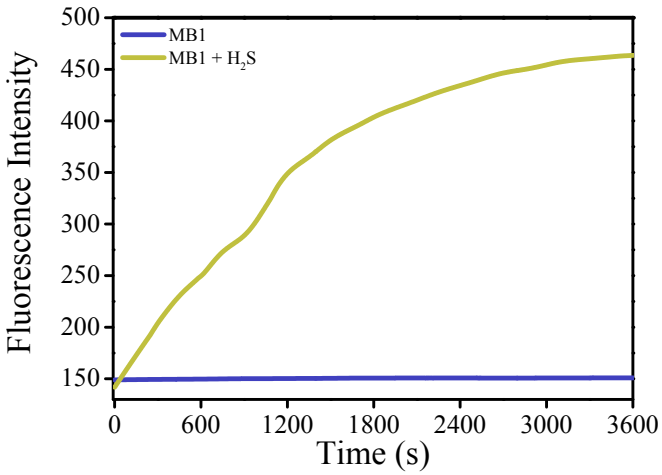


Fig. S6 Time-dependent fluorescence intensity at 687 nm of **MB1** (5 μM, in PBS, 0.25 % DMSO, pH 7.2) without or with 50 μM H_2S, λ_{ex}=600 nm.

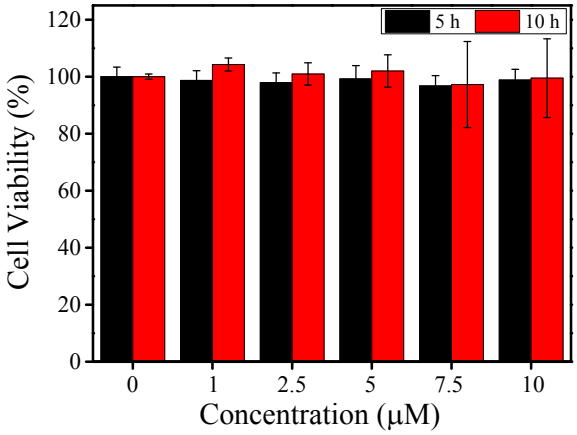


Fig. S7. Cell viability values (%) estimated by CCK-8 assay in HeLa cells, which were cultured in the presence of 0-10 μM **MB1** for 5 and 10 h.

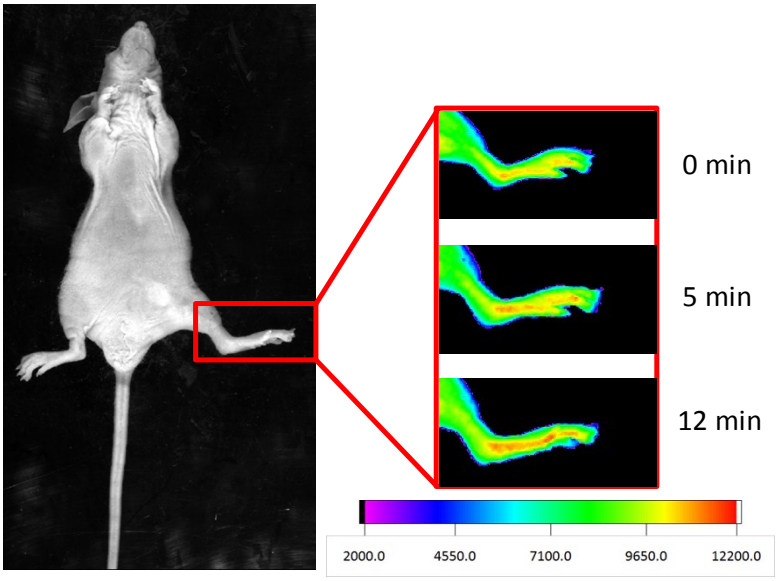


Fig. S8 *In vivo* fluorescence images of BALB/c Nude Mice after injection **MB1** (1 mM, 50 μL) and Na_2S (1 mM, 50 μL) into the right tibiotarsal joint (right ankle). The fluorescence signal was collected at λ_{em} = 720 ± 60 nm under an excitation filter 630 nm.

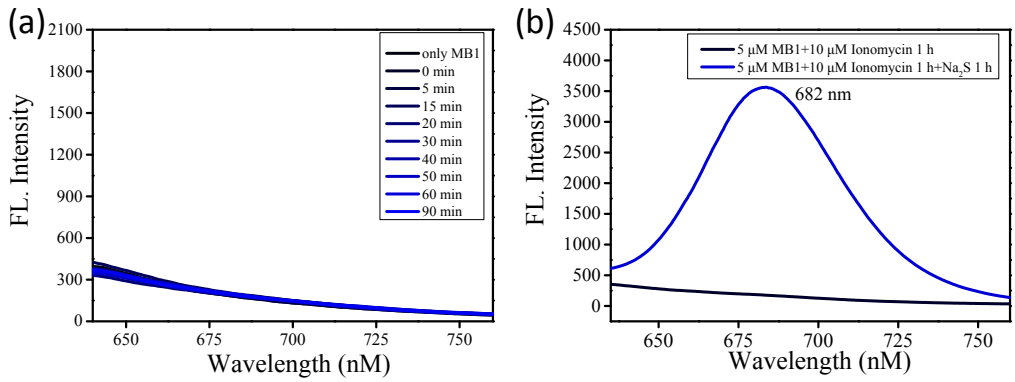


Fig. S9 (a) Fluorescence spectral changes of **MB1** (5 μM) with time after addition of Ionomycin (10 μM) in a mixed solvent of DMSO and PBS (1:1, v/v). (b) Fluorescence spectral changes of the mixture of **MB1** (5 μM) and Ionomycin (10 μM) after addition of Ionomycin (10 μM) in a mixed solvent of DMSO and PBS (1:1, v/v) for 0 min and 1 h. λ_{ex} = 600 nm.

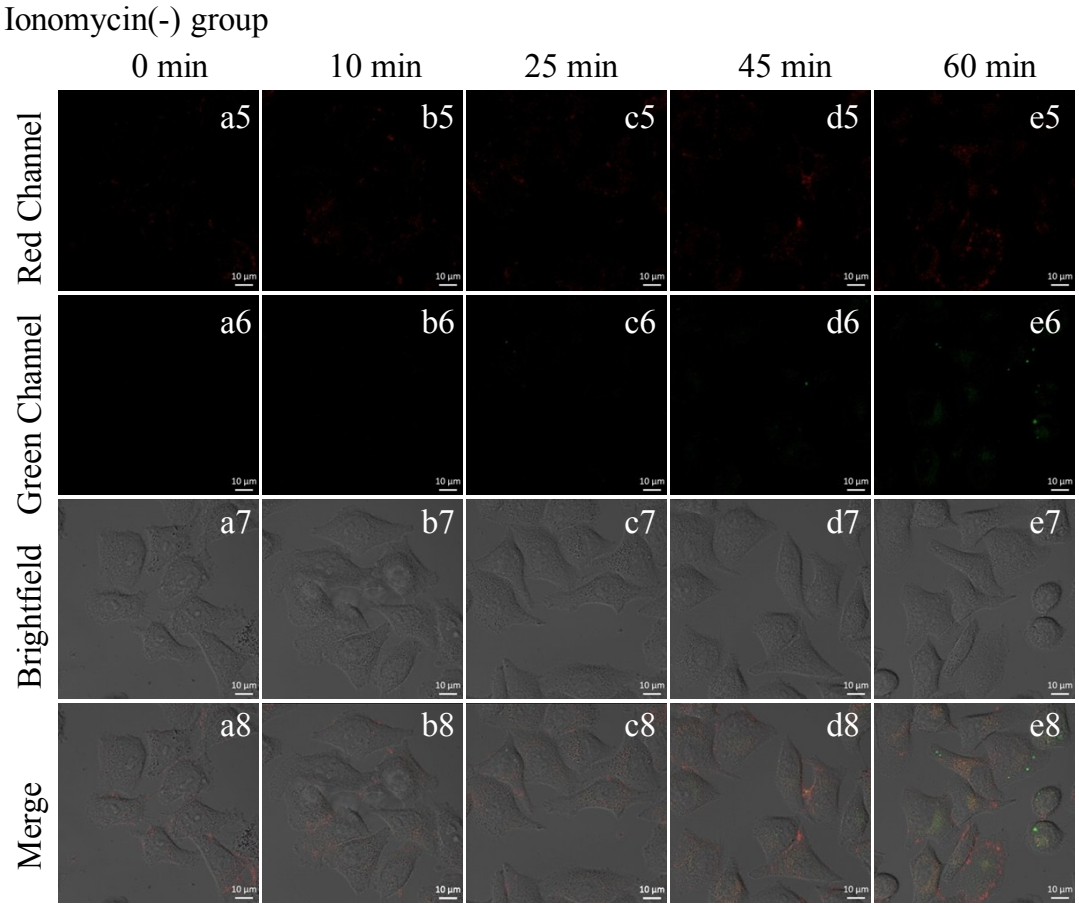


Fig. S10 CLSM images of **MB1** (5 μM) and **Fluo-3 AM** (1 μM)-loaded HeLa cells incubated without ionomycin (1 μM). The control group of ionomycin-treated cells for different time, shown in Figure 4A. Red channel, 690 ± 30 nm; λ_{ex} = 633 nm for **MB1**; Green channel, 525 ± 25 nm; λ_{ex} = 488 nm for Fluo-3 AM; Scale bar = 10 μm.

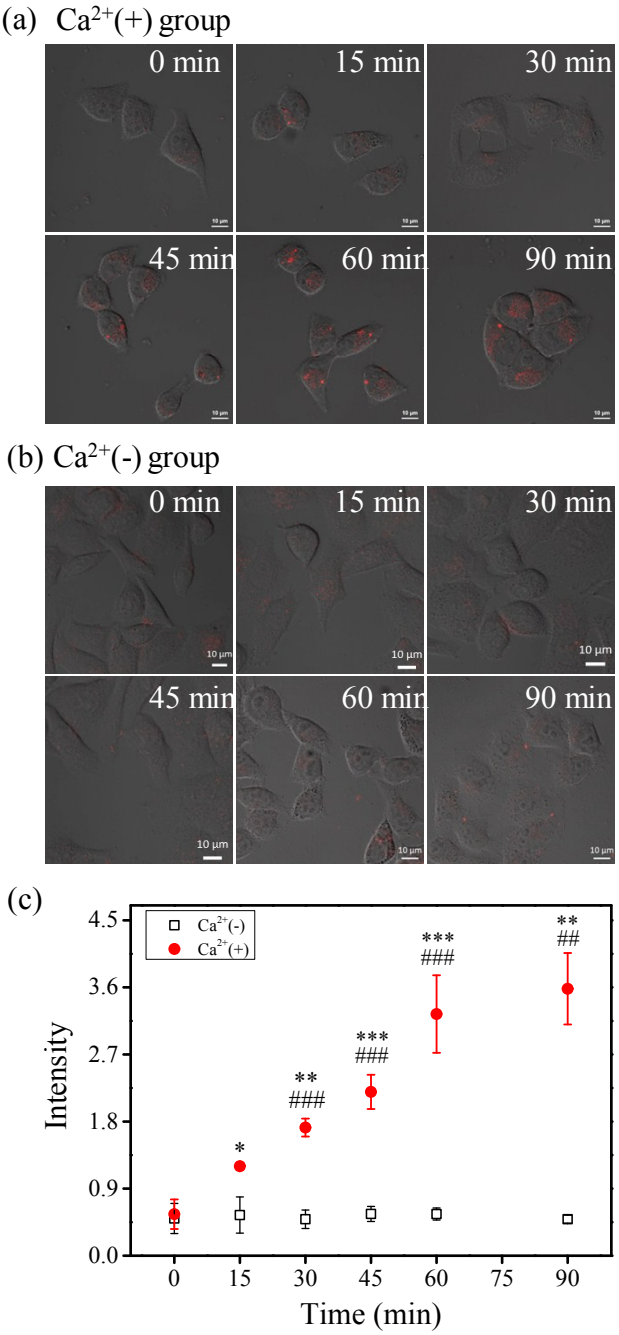


Fig. S11 CLSM images of **MB1** (5 μM)-loaded Hela cells. The cells were preincubated with (a) or without (b) $CaCl_2$ (200 μM) for 0 min, 15 min, 30 min, 45 min, 60 min and 90 min. Red channel, 690 ± 30 nm; λ_{ex} = 633 nm for **MB1**; Scale bar = 10 μm. (c) Corresponding average fluorescence intensities of cells in (a) and (b). Statistical analyses were employed with Student's *t*-test (n = 3). Compared with 0 min group: *$p < 0.05$, **$p < 0.01$ and ***$p < 0.001$. Compared with Ca^{2+} (-) group: $^{\#\#}p < 0.01$, $^{\#\#\#}p < 0.001$ and error bars are ± S.D.