A lysosome-targetable versatile fluorescent probe for imaging viscosity and peroxynitrite with different fluorescence signals in living cells

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The synthetic route of Lyso-NA

1. UV-vis absorption spectra of Lyso-NA

Fig S1. UV-vis absorption spectra of Lyso-NA (5 μM) in different ratio of glycerol-water.
2. The effect of pH

![Graph showing the effect of pH on Lyso-NA](image)

**Fig S2.** The effect of pH on Lyso-NA in different viscosity

3. The kinetic profile of the recognition of Lyso-NA for ONOO⁻

![Graph showing the kinetic profile](image)

**Fig S3.** The fluorescence spectra of Lyso-NA (5 μM) after the addition of ONOO⁻ (150 μM) in H₂O: Ethanol=5:5 (v/v), pH=5.0, 10 mM CPBS at room temperature. Excitation wavelength = 440 nm, excitation and emission slit widths = 5 nm and 5 nm.
4. The affect of DMSO to HClO

**Fig S4.** (a) The fluorescence spectra of Lyso-NA (5 μM) in the presence of HClO (150 μM) or ONOO⁻ (150 μM) in H₂O: Ethanol=5:5 (v/v) contained 0.5% DMSO, pH=5.0, 10 mM CPBS at room temperature. (b) The fluorescence spectra of Lyso-NA (5 μM) in the presence of HClO (150 μM) or ONOO⁻ (150 μM) in H₂O: Ethanol=5:5 (v/v) contained 0.5% Methanol, pH=5.0, 10 mM CPBS at room temperature.

5. The two channels of Lyso-NA in the cells

**Fig. S5** Confocal fluorescence images of RAW.264.7 cells stained by Lyso-NA (5 μM). (a) overlay of green and red channels. (b) green channel of Lyso-NA (460-540 nm), excited at 404 nm. (c) red channel of Lyso-NA (580-670 nm), excited at 543 nm. (d) bright field image.
6. Spectral data

**Fig S6.** H-NMR of N-(Morpholinoethylamino)-4-Bromo-1,8-Naphthalimide in CDCl$_3$

**Fig S7.** $^{13}$C-NMR of N-(Morpholinoethylamino)-4-Bromo-1,8-Naphthalimide in CDCl$_3$
**Fig S8.** The mass spectrum of N-(Morpholinoethylamino)-4-Bromo-1,8-Naphthalimide

**Fig S9.** H-NMR of N-(Morpholinoethylamino)-4-hydroxy-1,8-naphthalimide in DMSO
**Fig S10.** $^{13}$C-NMR of N-(Morpholinoethylamino)-4-hydroxy-1,8-naphthalimide in DMSO

**Fig S11.** H-NMR of N-(Morpholinoethylamino)-4-hydroxy-1,8-naphthalimide
Fig S12. H-NMR of N-(Morpholinoethylamino)-3-formyl-4-hydroxy-1,8-naphthalimide in DMSO

Fig S13. $^{13}$C-NMR of N-(Morpholinoethylamino)-3-formyl-4-hydroxy-1,8-naphthalimide in CDCl$_3$
Fig S14. H-NMR of N-(Morpholinoethylamino)-3-formyl -4-hydroxy-1,8-naphthalimide

Fig S15. H-NMR of Lyso-NA in DMSO
Fig S16. $^{13}$C-NMR of Lyso-NA in DMSO

Fig S17. The mass spectrum of Lyso-NA
Fig S18. The mass spectrum of the reaction mixture of Lyso-NA with ONOO⁻