

Supplementary Information

A Fast Response Fluorescence Probe Specific for Hypochlorous Acid Detection and Its Applications in Bioimaging

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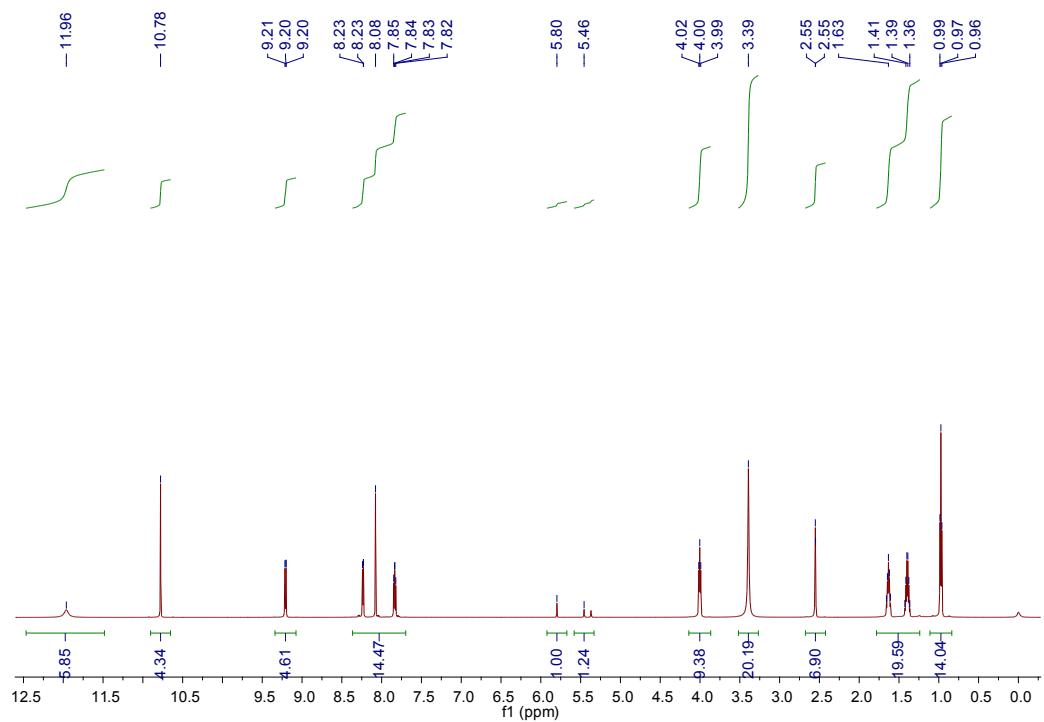


Fig. S1 ¹H NMR of NA (600 MHz, DMSO-*d*₆).

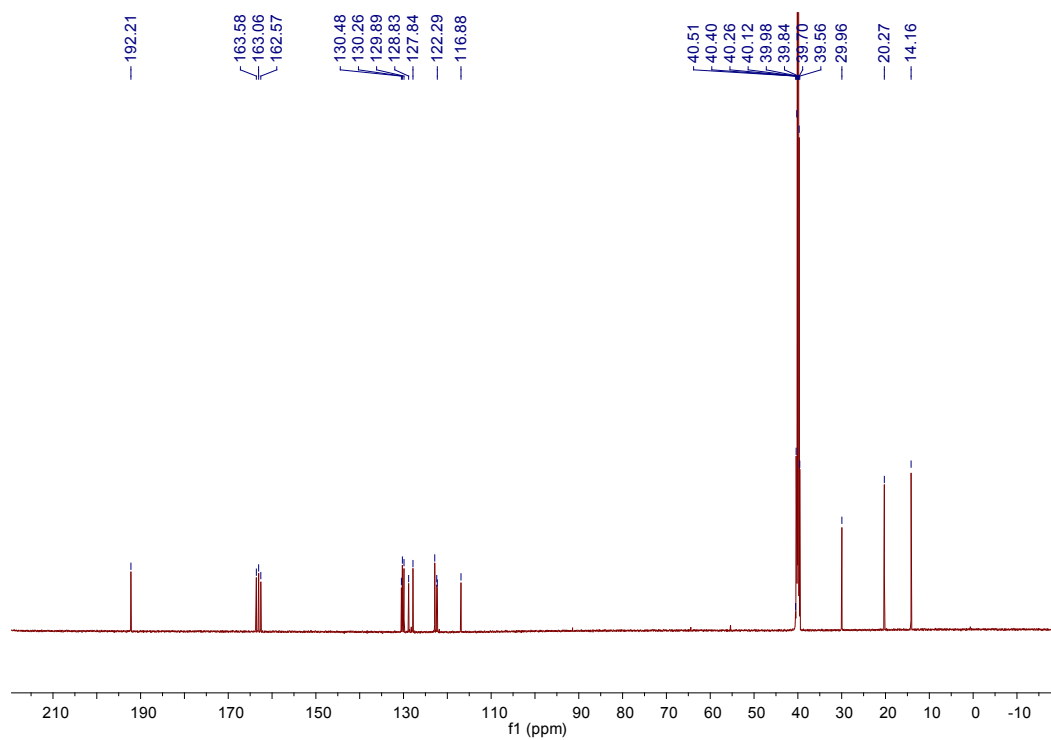


Fig. S2 ¹³C NMR of NA (150 MHz, DMSO-*d*₆).

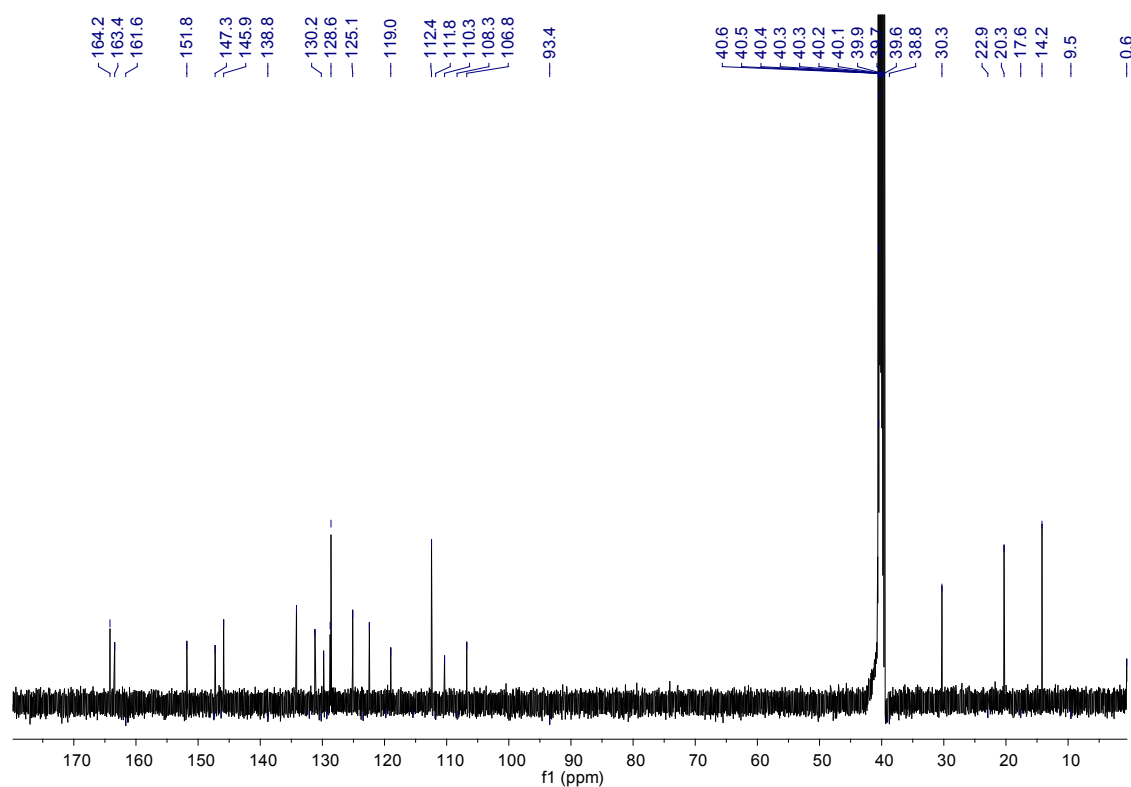


Fig. S5 ^{13}C NMR of **DNPH-NA** (150 MHz, $\text{DMSO-}d_6$).

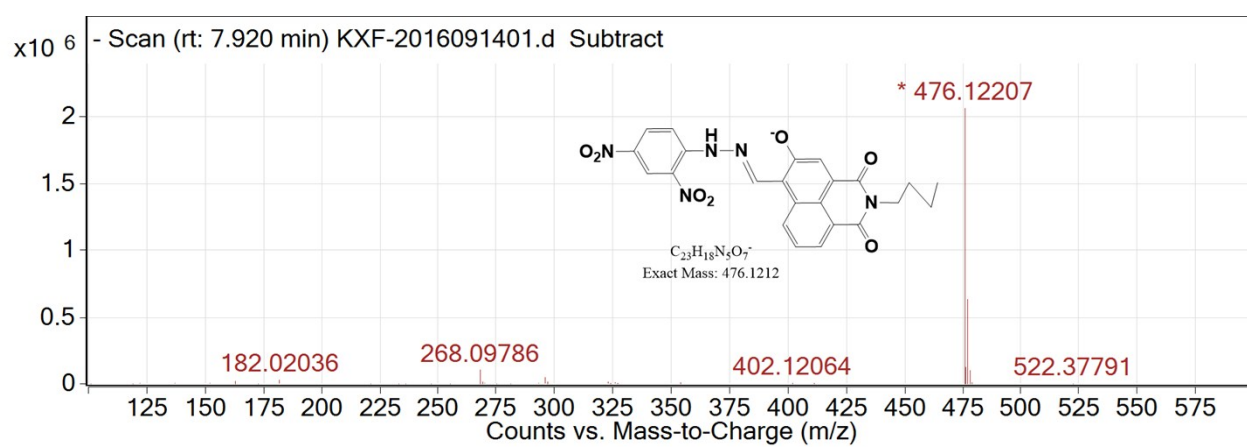


Fig. S6 HR MS of **DNPH-NA**.

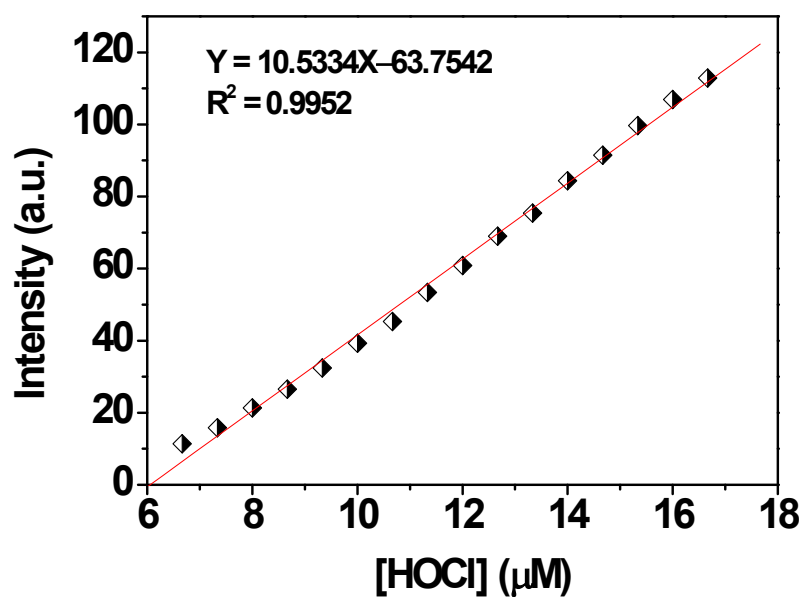


Fig. S7 Fluorescence spectra of **DNPH-NA** (3 μM) in (THF:PBS=3:7, v/v; pH 7.4) in titration point line fitting.



Fig. S8 Changes in fluorescence color of **DNPH-NA** (10 μM) in the presence of various ROS and anions (120 μM): (1) **DNPH-NA** only, (2) HOCl, (3) H₂O₂, (4) ¹O₂, (5) ONOO⁻, (6) ·OH, (7) ·O₂⁻, (8) NO₂⁻, (9) NO₃⁻, (10) PO₄³⁻, (11) Pi, (12) SO₄²⁻, (13) HCO₃⁻, (14) Cl⁻, (15) F⁻.

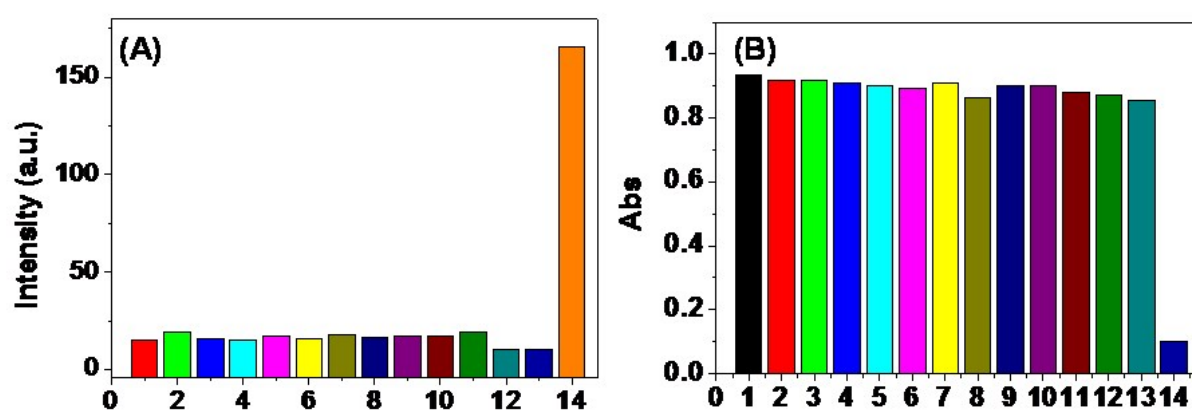


Fig. S9 Fluorescence response of **DNPH-NA** (10 μM) towards various biological cations (150 μM) in PBS buffer (THF:H₂O=3:7, v/v; pH 7.4). The cations include 1. Li⁺, 2. Na⁺, 3. K⁺, 4. Ca²⁺, 5. Mg²⁺, 6. Ba²⁺, 7. Al³⁺, 8. Fe³⁺, 9. Cr³⁺, 10. Zn²⁺, 11. Co²⁺, 12. Mn²⁺, 13. Cu²⁺, 14. HOCl. The excitation and emission wavelength are 430/518 nm for **DNPH-NA**.

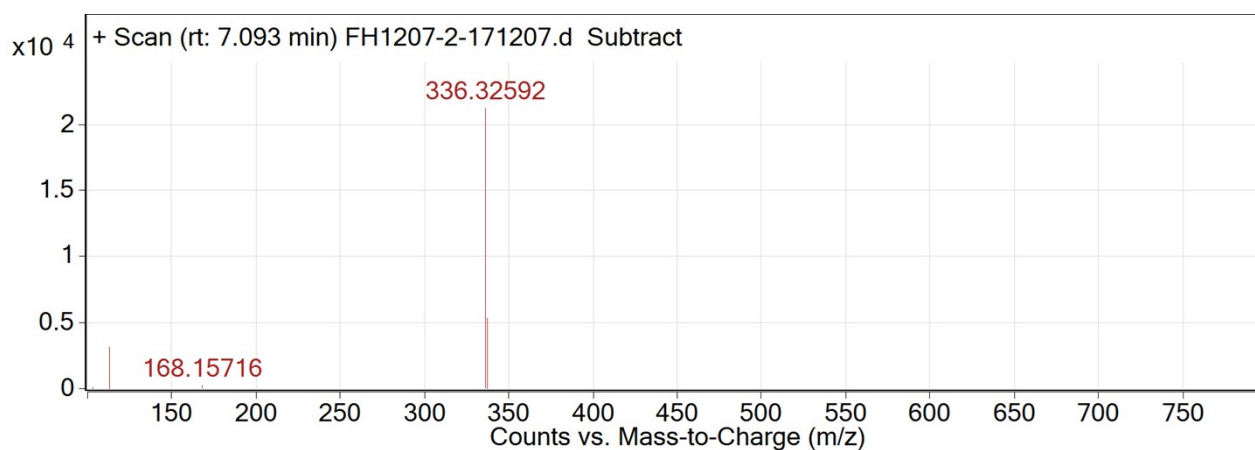


Fig. S 10 HR MS of **DNPH-NA** in the presence of HOCl.

Table S1. Some recently reported fluorescent probes for HOCl detection^{S1-9}.

Probes	Detection limit	Solvent	Colour changes	Ex/Em (nm)	emission enhancement	Response time	Intracellular analysis	<i>In vivo/vitro</i> sensing	Ref
HQ	0.31 μ M	PBS–DMSO (1 : 9, v/v, 7.4).	--	495/370	23.5-fold	within 10 min	Exogenous HOCl in HeLa cells	--	1
MPhS e– BOD	--	PBS-CH ₃ CN (7 : 3, v/v, 7.4)	--	460/510	~5-fold	5 min	Endogenous HOCl in macrophage	--	2
1b	0.356 μ M	PBS-CH ₃ CN (8 : 2, v/v, 7.4)	--	398/455	42-fold	5 min	Exogenous HOCl in HepG2 cells	--	3
RSTP P	9.00 nM	PBS (7.4)	Colourless -pink more than	553/580	200-fold	2 min	Endogenous HOCl in mitochondria of macrophage (subcellular level)	--	4
AC-CIO	25 nM	DMF-PBS solution (4:1, v/v; 10 mM PBS, pH 7.4)	--	480/576	372-fold	within 2 min)	exogenous and endogenous ClO ⁻ in live Raw 264.7 cells	-	5
1	--	DMF-PBS solution (1:9, v/v; 10 mM PBS, pH 7.4)	--	480/542	--	1 min	Exogenous HOCl in HeLa cells and endogenous HOCl in macrophage (single cell level by flow cytometry analysis)	--	6
FBS	0.20 μ M	in KH ₂ PO ₄ buffer (50 mM, pH 7.4)	-	498/523	--	--	--	(<i>Ex vivo</i>) intestinal HOCl in <i>Drosophila melanogaster</i> (endogenous HOCl)	7
MMSi R	-	DMF-PBS solution (0.9:99.1, v/v; PBS, pH 7.4)	Colourless -blue	620/670	--	A few seconds	Endogenous HOCl in neutrophils	(<i>In vivo</i>) mouse peritonitis (endogenous HOCl)	8
PZ-Py	17.9 nM	in PBS (pH 7.3, 10 mM, containing	-	400/562	40.5-fold	A few seconds	Exogenous HOCl in mitochondria HeLa,	(<i>In vivo</i>) nude mice (exogenous HOCl)	9

0.5% DMSO)							endogenous HOCl in mitochondria of RAW 264.7 (subcellular level)		
Ptz-AO	2.7 nM	in H ₂ O	-	475/540	75-fold	5 s	Exogenous HOCl in INS-1 β -islet cells and endogenous HOCl in macrophage	--	10
FDOC I-1	2.62 nM	in PBS (pH 7.2, 10 mM, containing 0.1% EtOH)	Colourless -blue	620/686	78-fold	30 s	Endogenous HOCl in macrophage	(<i>In vivo</i>) HOCl in mice arthritis (endogenous HOCl)	11

References:

1. X. Wang, X. Wang, Y. Feng, M. Zhu, H. Yin, Q. Guo and X. Meng, *Dalton Trans.*, 2015, **44**, 6613–6619.
2. B. Wang, P. Li, F. Yu, P. Song, X. Sun, S. Yang, Z. Lou and K. Han, *Chem. Commun.*, 2013, **49**, 1014–1016.
3. L. Long, Y. Wu, L. Wang, A. Gong, F. Hu and C. Zhang, *Chem. Commun.*, 2015, **51**, 10435–10438
4. J. Zhou, L. Li, W. Shi, X. Gao, X. Li and H. Ma, *Chem. Sci.*, 2015, **6**, 4884–4888.
5. J. Fan, H. Mu, H. Zhu, J. Wang and X. Peng, *Analyst*, 2015, **140**, 4594–4598.
6. Q. Xu, K.-A. Lee, S. Lee, K. M. Lee, W.-J. Lee and J. Yoon, *J. Am. Chem. Soc.*, 2013, **135**, 9944–9949.
7. Koide, Y. Urano, K. Hanaoka, T. Terai and T. Nagano, *J. Am. Chem. Soc.*, 2011, **133**, 5680–5682.
8. H. Xiao, K. Xin, H. Dou, G. Yin, Y. Quan and R. Wang, *Chem. Commun.*, 2015, **51**, 1442–1445.
9. L. Liang, C. Liu, X. Jiao, L. Zhao and X. Zeng, *Chem. Commun.*, 2016, **52**, 7982–7985.
10. P. Wei, W. Yuan, F. Xue, W. Zhou, R. Li, D. Zhang and T. Yi, *Chem. Sci.*, 2018, **9**, 495–501.