

Supplementary data

Some fluorescent HOCl probes developed since 2016 and their characteristics are presented in Table S1 (see articles [1–17]).

Table S1. Comparison of fluorescent probes for HOCl¹

Probe	Type of fluorescent signal	λ (nm)	LOD (nM)	Plato time (s) ²	References
CMOS	turn-off	405, F480↓	21	5	[1]
HKOCl-3	turn-on	490, F527↑	0.33	60	[2]
BCO	ratiometric	372, F460↑/430	154	n/a	[3]
BETC	turn-on	350, F440↑	32	n/a	[3]
FHZ	turn-on	490, F520↑	n/a	60	[4]
NDS	turn-on	420, F525↑	105	16	[5]
RT-1	turn-on	550, F587↑	2.18	180	[6]
BR-O	turn-on	610, F670↑	19	420	[7]
HKOCl-4	turn-on	530, F577↑	9	30	[8]
BRT	ratiometric	525, F580↑/540↓	38	15	[9]
BC-3	turn-on	620, F669↑	11	30	[10]
HDI-HClO	turn-on	440, F520↑	8.3	8	[11]
HQMN	ratiometric	370, F468↑/572↓	787	100	[12]
Dcp-EPtz	turn-on	475, F618↑	39	300	[13]
RO610	turn-on	535, F577↑	29	30	[14]
CR-Ly	ratiometric	420, F582↑/479↓	12	50	[15]
FD-301	turn-on	620, F686↑	44	10	[16]
FN-1, FN-2	turn-on	490, F529↑	210, 230	300, 120	[17]
CB	turn-on	430, F590↑	32	30	Current work

¹The scope of comparison includes only work published between 2016 and 2021 that involved cell or tissue imaging of HOCl.

²Plateau time means the time to reach a plateau in the change in fluorescence intensity after HOCl addition.

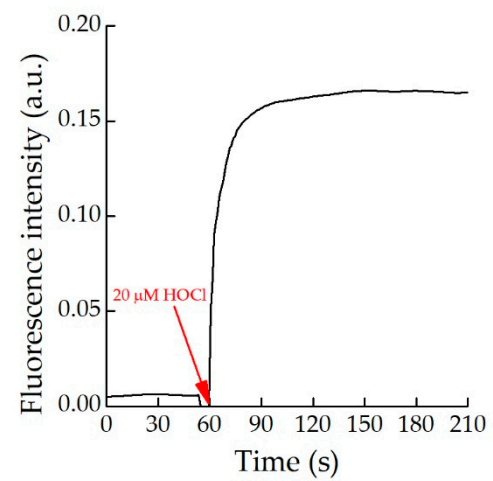
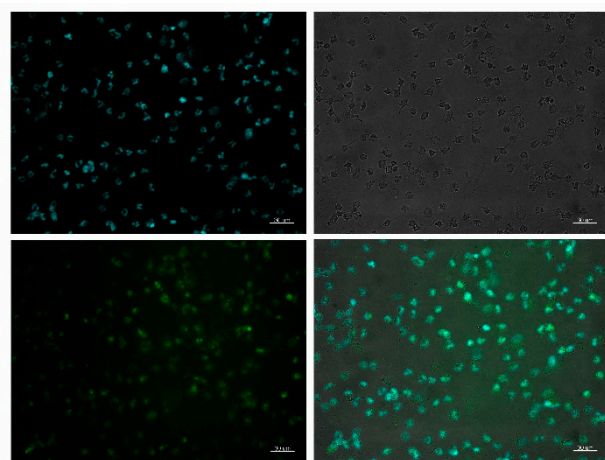
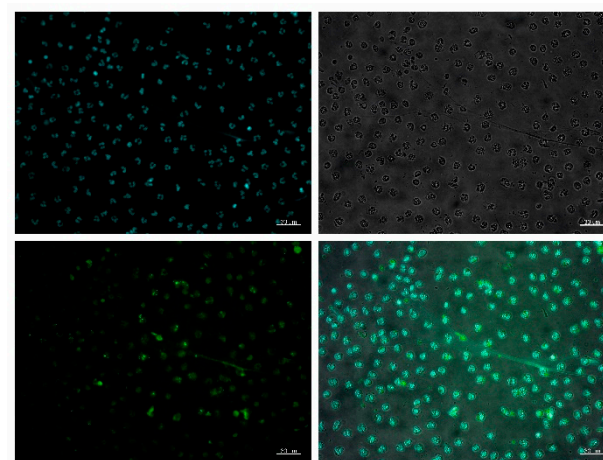


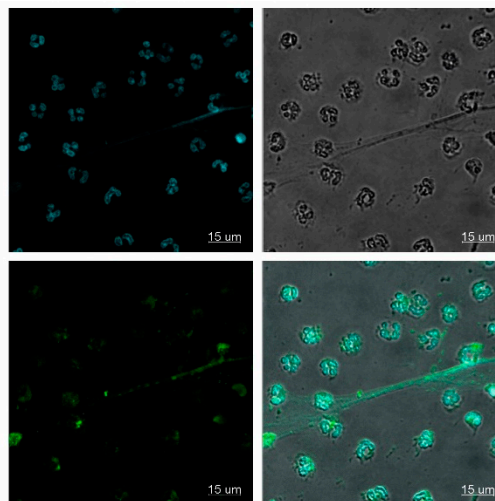
Figure S1. The time course of fluorescence intensity of CB (20 μ M) at 590 nm (λ_{ex} =430 nm) after treatment with 1 eqv. of HOCl (time range 0–210 s). PBS (pH7.4), T=25°C.



(a)



(b)



(c)

Figure S2. Visualization of neutrophil extracellular traps using APF and CB. (a) Images obtained by confocal microscopy for preparations of neutrophils incubated with 5 μ M APF in the presence of 50 nM PMA; (b), (c) with 20 μ M CB and in the presence of 50 nM PMA. 1 – DAPI (blue), 2 – differential interference contrast, 3 – fluorescence of APF or CB (green) obtained upon excitation with mercury-vapor lamp at 465–505 nm, emission at 515–750 nm (a), (b) or laser at 488 nm emission at 515–750 nm (c), 4 – merge of 1, 2 and 3.

References

1. Liu, Z.; Li, G.; Wang, Y.; Li, J.; Mi, Y.; Guo, L.; Xu, W.; Zou, D.; Li, T.; Wu, Y. A Novel Fluorescent Probe for Imaging the Process of HOCl Oxidation and Cys/Hcy Reduction in Living Cells. *RSC Advances* **2018**, *8*, 9519–9523, doi:10.1039/C7RA13419C.
2. Hu, J.J.; Wong, N.-K.; Lu, M.-Y.; Chen, X.; Ye, S.; Zhao, A.Q.; Gao, P.; Yi-Tsun Kao, R.; Shen, J.; Yang, D. HKOCl-3: A Fluorescent Hypochlorous Acid Probe for Live-Cell and in Vivo Imaging and Quantitative Application in Flow Cytometry and a 96-Well Microplate Assay. *Chemical Science* **2016**, *7*, 2094–2099, doi:10.1039/C5SC03855C.
3. Jin, L.; Tan, X.; Zhao, C.; Wang, Q. Two Highly Sensitive and Selective Coumarin-Based Fluorometric Probes for the Detection of ClO⁻ and Cell Imaging. *Analytical Methods* **2019**, *11*, 1916–1922, doi:10.1039/C9AY00105K.
4. Zhang, R.; Zhao, J.; Han, G.; Liu, Z.; Liu, C.; Zhang, C.; Liu, B.; Jiang, C.; Liu, R.; Zhao, T.; Han, M.-Y.; Zhang, Z. Real-Time Discrimination and Versatile Profiling of Spontaneous Reactive Oxygen Species in Living Organisms with a Single Fluorescent Probe. *Journal of the American Chemical Society* **2016**, *138*, 3769–3778, doi:10.1021/jacs.5b12848.
5. Jiao, C.; Liu, Y.; Pang, J.; Lu, W.; Zhang, P.; Wang, Y. A Simple Lysosome-Targeted Probe for Detection of Hypochlorous Acid in Living Cells. *Journal of Photochemistry and Photobiology A: Chemistry* **2020**, *392*, 112399–112405, doi:10.1016/j.jphotochem.2020.112399.
6. Choi, M.G.; Lee, Y.J.; Lee, K.M.; Park, K.Y.; Park, T.J.; Chang, S.-K. A Simple Hypochlorous Acid Signaling Probe Based on Resorufin Carbonodithioate and Its Biological Application. *Analyst* **2019**, *144*, 7263–7269, doi:10.1039/C9AN01884K.
7. Yang, J.; Yao, Y.; Shen, Y.; Xu, Y.; Lv, G.; Li, C. A Novel Phenoxazine-Based Fluorescent Probe for the Detection of HOCl in Living Cells. *Zeitschrift für anorganische und allgemeine Chemie* **2020**, *646*, 431–436, doi:10.1002/zaac.202000127.

8. Bai, X.; Yang, B.; Chen, H.; Shen, J.; Yang, D. HKOCl-4: A Rhodol-Based Yellow Fluorescent Probe for the Detection of Hypochlorous Acid in Living Cells and Tissues. *Organic Chemistry Frontiers***2020**, 7, 993–996, doi:10.1039/D0QO00081G.
9. Liu, Y.; Zhao, Z.-M.; Miao, J.-Y.; Zhao, B.-X. A Ratiometric Fluorescent Probe Based on Boron Dipyrromethene and Rhodamine Förster Resonance Energy Transfer Platform for Hypochlorous Acid and Its Application in Living Cells. *Analytica Chimica Acta***2016**, 921, 77–83, doi:10.1016/j.aca.2016.03.045.
10. Zheng, W.; Yang, J.; Shen, Y.; Yao, Y.; Lv, G.; Hao, S.; Li, C. The Near-Infrared Fluorescent Probes Based on Phenoxazine for the Rapid Detection of Hypochlorous Acid. *Dyes and Pigments***2020**, 179, 108404–108410, doi:10.1016/j.dyepig.2020.108404.
11. Luo, P.; Zhao, X. A Sensitive and Selective Fluorescent Probe for Real-Time Detection and Imaging of Hypochlorous Acid in Living Cells. *ACS Omega***2021**, 6, 12287–12292, doi:10.1021/acsomega.1c01102.
12. Das, S.; Aich, K.; Patra, L.; Ghoshal, K.; Gharami, S.; Bhattacharyya, M.; Mondal, T.K. Development of a New Fluorescence Ratiometric Switch for Endogenous Hypochlorite Detection in Monocytes of Diabetic Subjects by Dye Release Method. *Tetrahedron Letters***2018**, 59, 1130–1135, doi:10.1016/j.tetlet.2018.02.023.
13. Zheng, D.; Qiu, X.; Chang, L.; Xiaojie Jiao; Song He; Liancheng Zhao; Xianshun Zeng Synthesis and Bioapplication of a Highly Selective and Sensitive Fluorescent Probe for HOCl Based on a Phenothiazine-Dicyanoisophorone Conjugate with Large Stokes Shift. *New Journal of Chemistry***2018**, 42, 5135–5141, doi:10.1039/C8NJ00279G.
14. Zhang, Y.; Ma, L.; Tang, C.; Pan, S.; Shi, D.; Wang, S.; Li, M.; Guo, Y. A Highly Sensitive and Rapidly Responding Fluorescent Probe Based on a Rhodol Fluorophore for Imaging Endogenous Hypochlorite in Living Mice. *Journal of Materials Chemistry B***2018**, 6, 725–731, doi:10.1039/C7TB02862H.
15. Meng, H.; Huang, X.-Q.; Lin, Y.; Yang, D.-Y.; Lv, Y.-J.; Cao, X.-Q.; Zhang, G.-X.; Dong, J.; Shen, S.-L. A New Ratiometric Fluorescent Probe for Sensing Lysosomal HOCl Based on Fluorescence Resonance Energy Transfer Strategy. *Spectrochimica Acta Part A: Molecular and Biomolecular Spectroscopy***2019**, 223, 117355–117360, doi:10.1016/j.saa.2019.117355.
16. Liu, L.; Wei, P.; Yuan, W.; Liu, Z.; Xue, F.; Zhang, X.; Yi, T. Detecting Basal Myeloperoxidase Activity in Living Systems with a Near-Infrared Emissive “Turn-On” Probe. *Anal. Chem.***2020**, 92, 10971–10978, doi:10.1021/acs.analchem.9b04601.
17. Lv, J.; Wang, F.; Wei, T.; Chen, X. Highly Sensitive and Selective Fluorescent Probes for the Detection of HOCl/OCl⁻ Based on Fluorescein Derivatives. *Industrial & Engineering Chemistry Research***2017**, 56, 3757–3764, doi:10.1021/acs.iecr.7b00381.