A ratiometric fluorescence sensor for HOCl based on FRET platform and application in living cells

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Scheme S1 Synthesis of CRSH
Fig. S1 HRMS spectra of one fluorescent compound separated from the reaction of rhodamine thiohydrazide and HOCl.

Fig. S2 The red line is the donor (1 μM) and the green line is CRSH (1 μM) after addition of HOCl (4 μM) Condition: NaH$_2$PO$_4$ (0.05 M, pH = 5) : EtOH = 5 : 5 ( v/v), $\lambda_{ex}$: 350 nm (slit widths: 10 nm/5 nm).

Energy transfer efficiency (CRSH) = $1 - \frac{F_{DA}}{F_D} = 1 - \frac{46.3481}{140.4140} = 67.0\%$
Fig. S3 pH-dependent fluorescence intensity ratio changes of CRSH, Condition: \([\text{CRSH}] = 1 \, \mu\text{M}, \text{NaH}_2\text{PO}_4 \,(0.05 \, \text{M}, \text{pH} = 4-10) : \text{EtOH} \,(5 : 5, \, \text{v/v}), \lambda_{\text{ex}}: 350 \, \text{nm} \,(\text{slit widths: 10 nm/10 nm}), \text{HOCl} \,(4 \, \mu\text{M}).

Fig. S4 (a) The ratio \(I_{580}/I_{470}\) of CRSH with addition of ROS/RNS. 1. free CRSH, 2. CRSH+HOCl, 3-10. CRSH+other ROS/RNS \((^1\text{O}_2, \text{H}_2\text{O}_2, \text{HO}^-, \text{NO}, \text{NOOO}^-, \cdot\text{O}_2, \cdot\text{BuO}, \cdot\text{BuOOH})+\text{HOCl}\). (b) Absorption spectra of CRSH toward \text{HOCl} \,(4 \, \mu\text{M}), other ROS/RNS \,(20 \, \mu\text{M for HO}^-, \text{ONOO}^-, \text{NO}, \text{H}_2\text{O}_2, \cdot\text{BuOOH}, \cdot\text{BuOO}^-, ^1\text{O}_2, \cdot\text{O}_2),\) Condition: Condition: \([\text{CRSH}] = 1 \, \mu\text{M}, \text{NaH}_2\text{PO}_4 \,(0.05 \, \text{M}, \text{pH} = 5) : \text{EtOH} \,(5 : 5, \, \text{v/v}).\)
Fig. S5 Absorption spectra of CRSH upon addition of HOCl (0–4 μM), Condition: [CRSH] = 1 μM, NaH₂PO₄ (0.05 M, pH = 5) : EtOH (5 : 5, v/v).

Fig. S6 Time-dependent fluorescence intensity ratio changes of CRSH, Condition: [CRSH] = 1 μM, NaH₂PO₄ (0.05 M, pH = 5) : EtOH (5 : 5, v/v), λₑₓ: 350 nm (slit widths: 10 nm/10 nm).
Fig. S7 Viability of RAW264.7 cells after treatment with CRSH for 6 h (a), 12 h (b) at the different concentration (1, 5, 10 μM).