

## Supporting Information

### Highly specific and ultrasensitive probe for the imaging of inflammation-induced endogenous hypochlorous acid<sup>‡</sup>

Xinjian Song<sup>a,b</sup>, Wei Hu<sup>a</sup>, Donghua Wang<sup>b</sup>, Zhiqiang Mao<sup>a</sup>, Zhihong Liu<sup>a\*</sup>

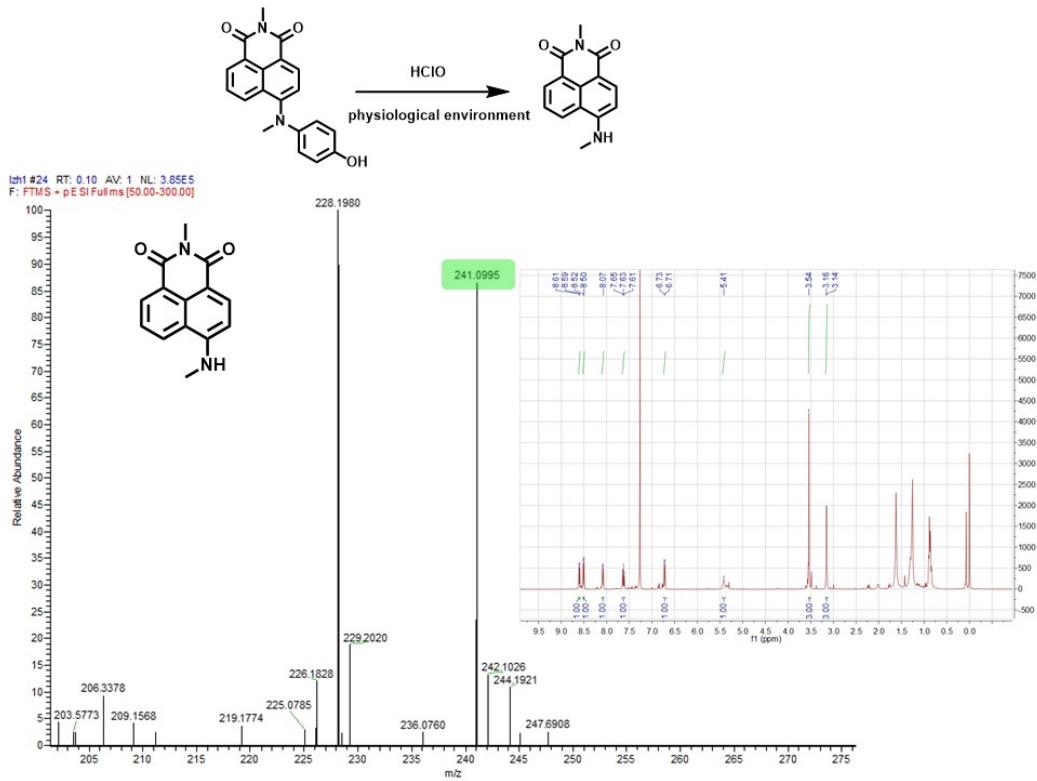
<sup>a</sup> Key Laboratory of Analytical Chemistry for Biology and Medicine (Ministry of Education), College of Chemistry and Molecular Sciences, Wuhan University, Wuhan 430072, China

<sup>b</sup> Hubei Key Laboratory of Biological Resources Protection and Utilization, Hubei University for Nationalities, Enshi 445000, China

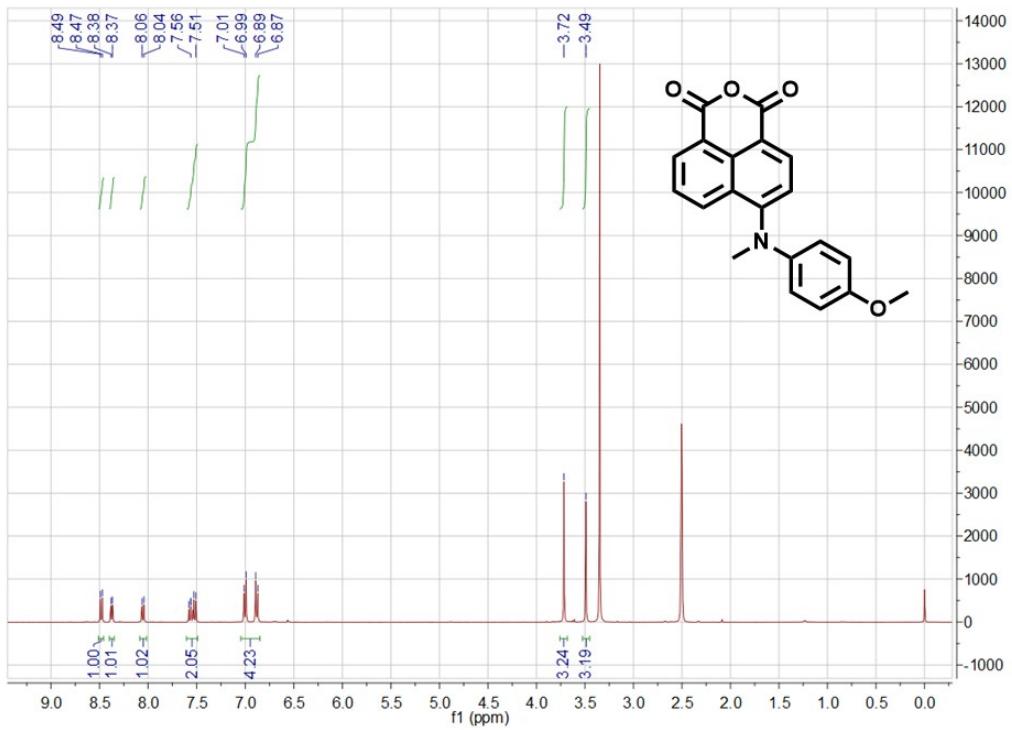
\* To whom correspondence should be addressed.

Tel: +86 27 87217886; Fax: +86 27 87217886;

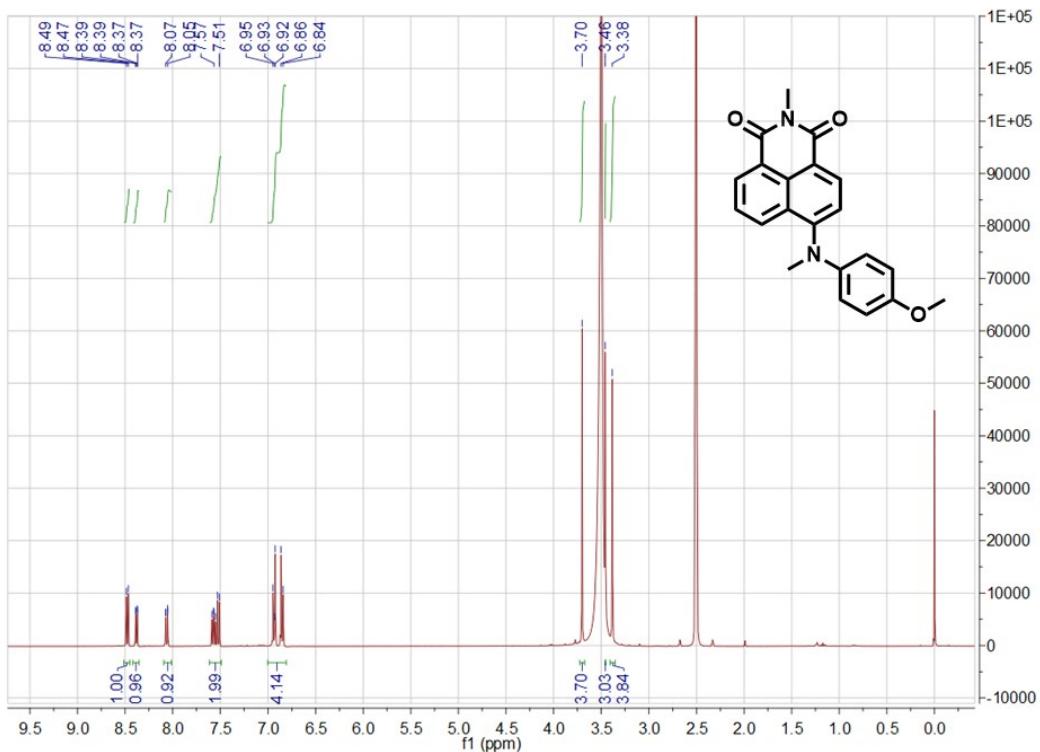
E-mail address: zhhl@whu.edu.cn



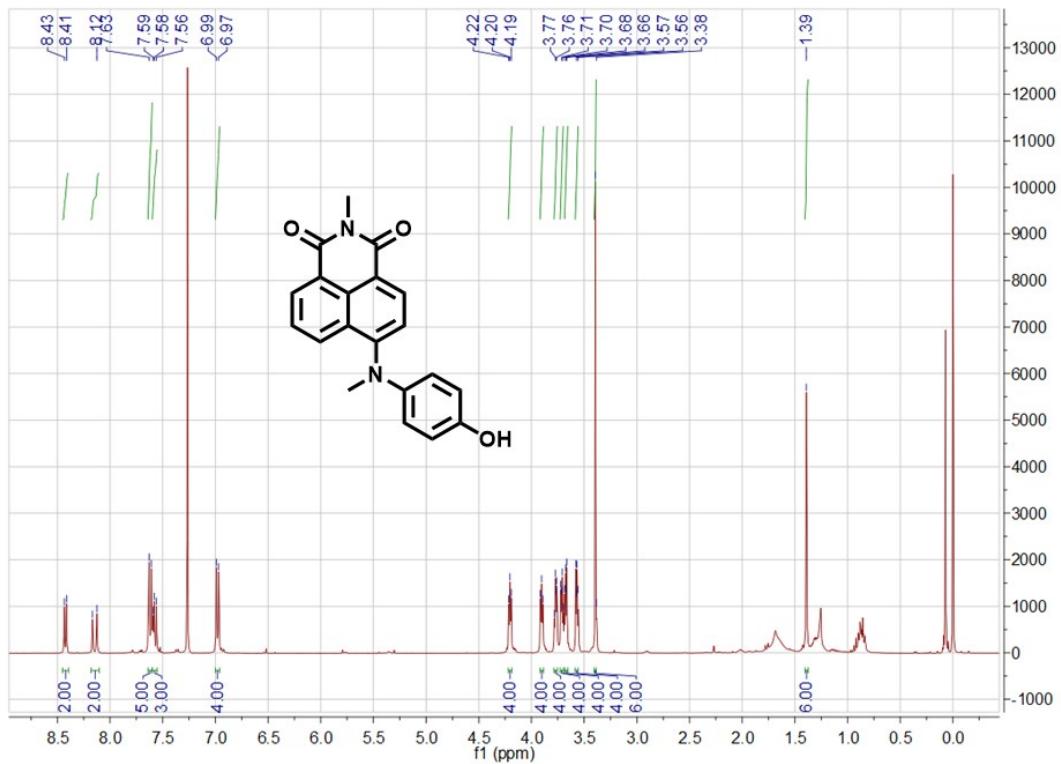
**Fig. S1** Proposed mechanism and mass spectrum of **HCA-Green** after reaction with 5 equivalents of NaClO.



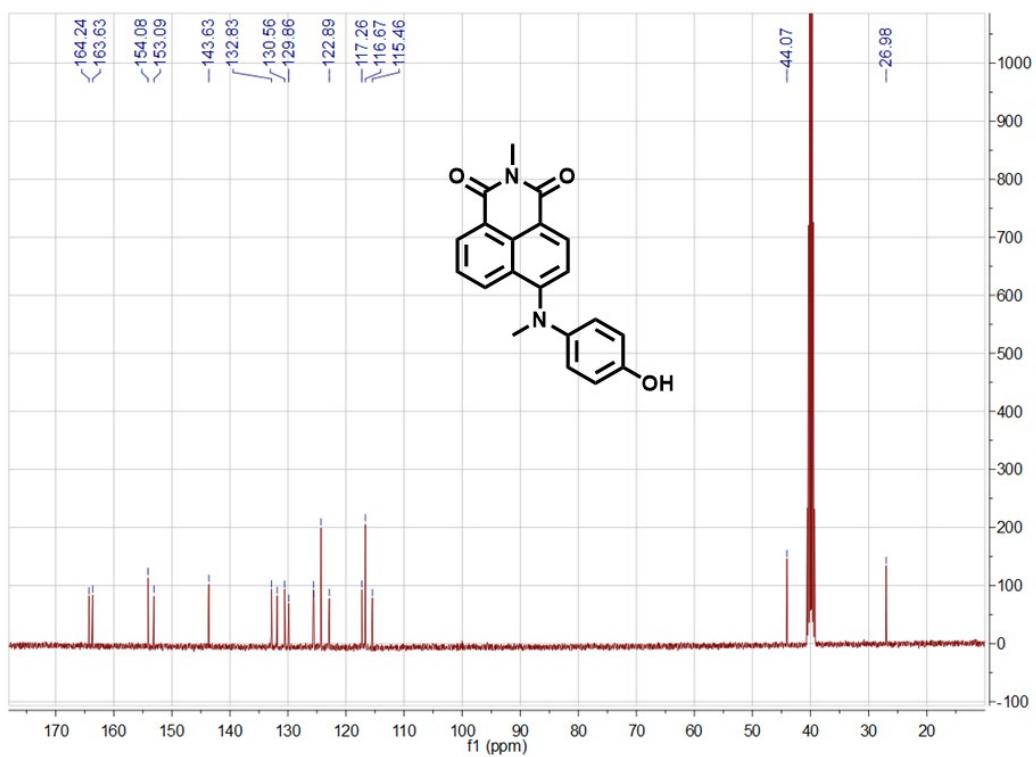
**Fig. S2**  $^1\text{H}$ -NMR spectrum of **1** (400 MHz,  $\text{DMSO}-d_6$ )



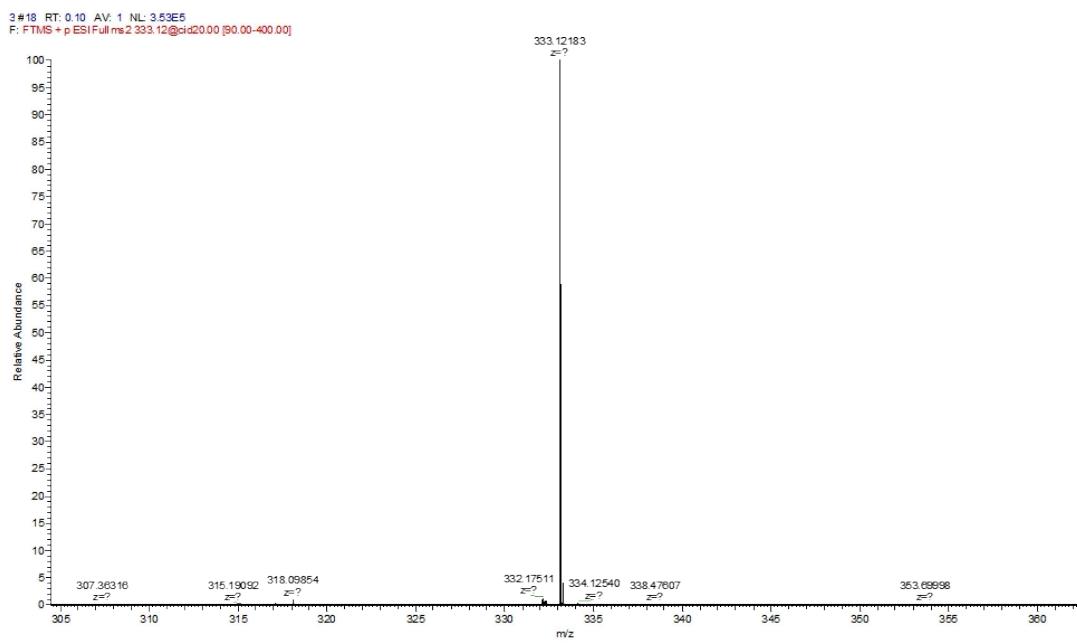
**Fig. S3**  $^1\text{H}$ -NMR spectrum of **2** (400 MHz, DMSO- $d_6$ )



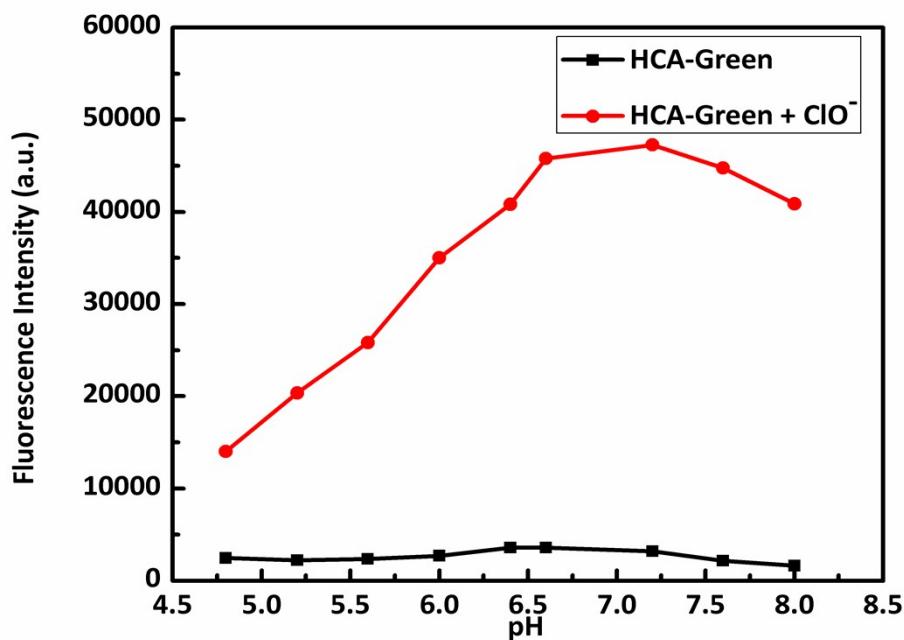
**Fig. S4**  $^1\text{H}$ -NMR spectrum of HCA-Green (400 MHz,  $\text{DMSO}-d_6$ )



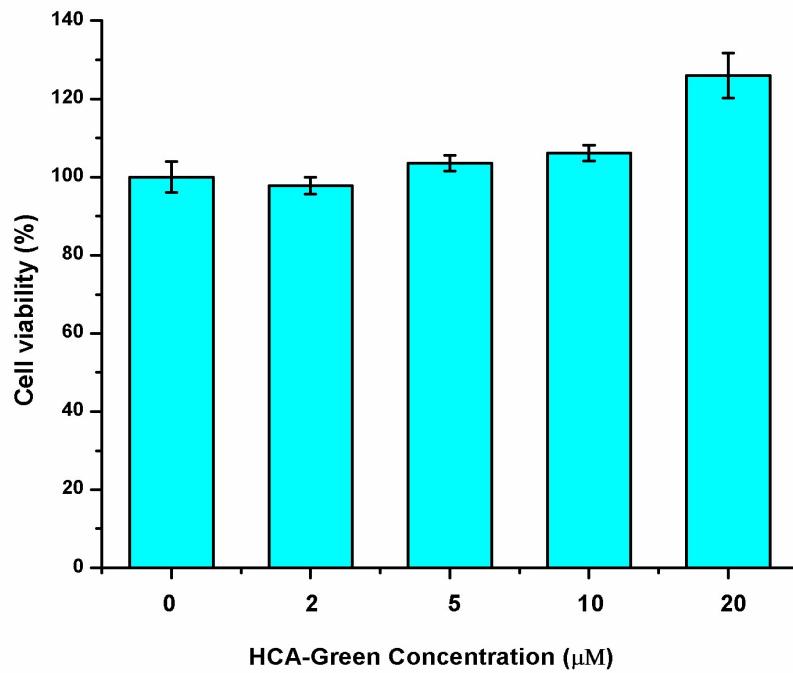
**Fig. S5** <sup>13</sup>C-NMR spectrum of **HCA-Green** (100 MHz, DMSO-*d*<sub>6</sub>)



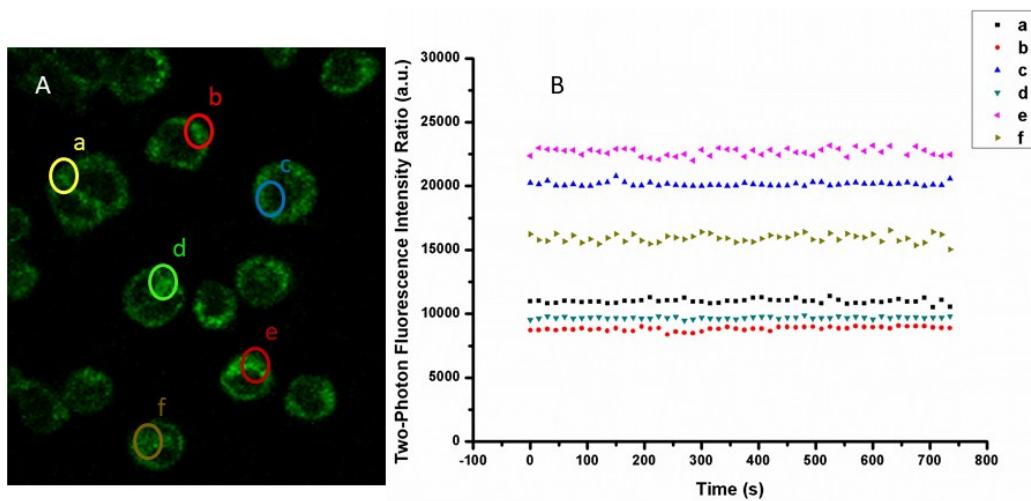
**Fig. S6** HRMS spectrum of **HCA-Green**



**Fig. S7** Effect of pH on the fluorescence ( $\lambda_{\text{ex}}/\lambda_{\text{em}} = 448/556 \text{ nm}$ ) of **HCA-Green** (10  $\mu\text{M}$ ) after reacting with sodium hypochlorite (50  $\mu\text{M}$ ). Reaction time: 5 min.



**Fig. S8** Cell survival rate of control groups (without **HCA-Green**) and experimental group (with 2, 5, 10, 20  $\mu\text{M}$  of **HCA-Green**). All groups contain 1% DMSO in 100  $\mu\text{L}$  DMEM).



**Fig. S9** (A) TPM images of HeLa cells labeled with 1  $\mu\text{g}/\text{mL}$  LPS and 1  $\mu\text{g}/\text{mL}$  PMA for 1 h and further incubated with **HCA-Green** for 30 min; (B) Two-photon fluorescence intensity from circle a-f as a function of time. The two-photon fluorescence intensity was collected with 15-sec intervals for the duration of 12.5 min under *xyt* mode. Scale bar: 20  $\mu\text{m}$ .

**Table S1.** Comparison of fluorescent probes for HClO. (OP: One-photon, TP: Two-photon)

Structure of probe	$\lambda_{\text{ex}}/\text{em}$ (nm)	Detection Limit (nM)	Response Time	Reference
	375/500	16.6	Within seconds	[1]
	490/527	0.33	< 1 min	[2]
	426/492, 562	89	60 s	[3]

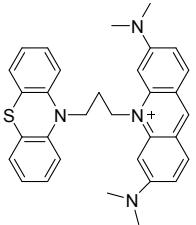
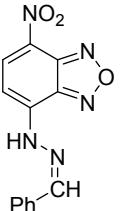
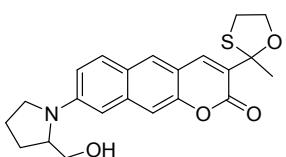
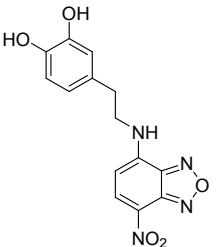
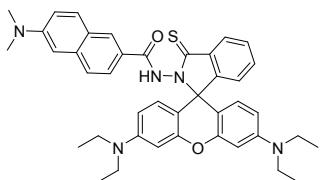
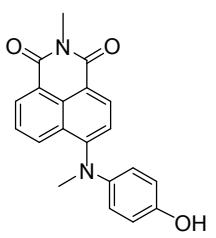
---

	405/505	674	2.5 min	[4]
	OP: 550/630 TP: 800/630	40	90 s	[5]
	425/480, 554	590	30 s	[6]
	468/557	1.37	2 min	[7]
	360/435	80	30 s	[8]
	498/523	200	Not mentioned	[9]
	488/538, 589	850	15 s	[10]

---



---

	475/540	2.7	5 s	[19]
	495/560	2.7	50 s	[20]
	460/598, 633	34.8	Within seconds	[21]
	470/540	9.7	<30 s	[22]
	350/440, 585	100	50 s	[23]
	OP: 454/556 TP: 810/556	42.3	50 s	<b>This work</b>

---

## References

- [1] L. Yuan, L. Wang, B. K. Agrawalla, S. J. Park, H. Zhu, B. Sivaraman, J. J. Peng, Q. H. Xu and Y. T. Chang, *J. Am. Chem. Soc.* 2015, **137**, 5930–5938.
- [2] J. J. Hu, N. K. Wong, M. Y. Lu, X. M. Chen, S. Ye, A. Q. Zhao, P. Gao, R. Y. Kao, J. Shen and D. Yang, *Chem. Sci.*, 2016, **7**, 2094–2099.
- [3] Z. Q. Mao, M. T. Ye, W. Hu, X. X. Ye, Y. Y. Wang, H. J. Zhang, C. Y. Li and Z. H. Liu, *Chem. Sci.*, 2018, **9**, 6035–6040.
- [4] B. B. Zhang, X. P. Yang, R. Zhang, Y. Liu, X. L. Ren, M. Xian, Y. Ye and Y. F. Zhao, *Anal.*

- Chem.* 2017, **89**, 10384–10390.
- [5] Y. J. Gong, M. K. Lv, M. L. Zhang, Z. Z. Kong and G. J. Mao, *Talanta*, 2019, **192**, 128–134.
  - [6] J. T. Hou, H. S. Kim, C. Duan, M. S. Ji, S. Wang, L. T. Zeng, W. X. Ren and J. S. Kim, *Chem. Commun.*, 2019, **55**, 2533–2536.
  - [7] P. Jia, Z. H. Zhuang, C. Y. Liu, Z. K. Wang, Q. X. Duan, Z. L. Li, H. C. Zhu, B. Du, B. C. Zhu, W. L. Sheng and B. T. Kang, *Anal. Chim. Acta*, 2019, **1052**, 131–136.
  - [8] L. Y. Chen, S. J. Park, D. Wu, H. M. Kim and J. Y. Yoon, *Dyes and Pigments*, 2018, **158**, 526–532.
  - [9] Q. L. Xu, K. A. Lee, S. Y. Lee, K. M. Lee, W. J. Lee and J. Y. Yoon, *J. Am. Chem. Soc.*, 2013, **135**, 9944–9949.
  - [10] J. Kang, F. J. Huo, Y. K. Yue, Y. Wen, J. B. Chao, Y. B. Zhang and C. X. Yin, *Dyes and Pigments*, 2017, **136**, 852–858.
  - [11] L. L. Xi, X. F. Guo, C. L. Wang, W. L. Wu, M. F. Huang, J. Y. Miao and B. X. Zhao, *Sens. Actuat. B-Chem.*, 2018, **255**, 666–671.
  - [12] B. Zhu, L. Wu, M. Zhang, Y. Wang, C. Liu, Z. Wang, Q. Duan and P. Jia, *Biosens. Bioelectron.*, 2018, **107**, 218–223.
  - [13] F. S. Tian, Y. Jia, Y. N. Zhang, W. Song, G. J. Zhao, Z. J. Qu, C. Y. Li, Y. H. Chen and P. Li, *Biosens. Bioelectron.*, 2016, **86**, 68–74.
  - [14] T. Guo, L. Cui, J. N. Shen, R. Wang, W. P. Zhu, Y. F. Xu and X. H. Qian, *Chem. Commun.*, 2013, **49**, 1862–1864.
  - [15] M. Emrullahoglu, M. Uçuncu and E. Karakus, *Chem. Commun.*, 2013, **49**, 7836–7838.
  - [16] G. H. Cheng, J. L. Fan, W. Sun, J. F. Cao, C. Hu and X. J. Peng, *Chem. Commun.*, 2014, **50**, 1018–1020.
  - [17] S. I. Reja, V. Bhalla, A. Sharma, G. Kaur and M. Kumar, *Chem. Commun.*, 2014, **50**, 11911–11914.
  - [18] J. Zhou, L. H. Li, W. Shi, X. H. Gao, X. H. Li and H. M. Ma, *Chem. Sci.*, 2015, **6**, 4884–4888.
  - [19] L. J. Liang, C. Liu, X. J. Jiao, L. C. Zhao and X. S. Zeng, *Chem. Commun.*, 2016, **52**, 7982–7985.
  - [20] Q. Fu, G. Chen, Y. X. Liu, Z. P. Cao, X. E. Zhao, G. L. Li, F. B. Yu, L. X. Chen, H. Wang and J. M. You, *Analyst*, 2017, **142**, 1619–1627.
  - [21] Y. W. Jun, S. Sarkar, S. Singha, Y. J. Reo, H. R. Kim, J. J. Kim, Y. T. Chang and K. H. Ahn, *Chem. Commun.*, 2017, **53**, 10800–10803.
  - [22] Y. Jiang, G. S. Zheng, N. Cai, H. T. Zhang, Y. Tan, M. J. Huang, Y. H. He, J. He and H. Y. Sun, *Chem. Commun.*, 2017, **53**, 12349–12352.
  - [23] Y. R. Zhang, N. Meng, J. Y. Miao and B. X. Zhao, *Chem. Eur. J.*, 2015, **21**, 19058–19063.