Electronic Supplementary Information

Sensitive and Selective Detection of Uracil-DNA Glycosylase Activity with a

New Pyridinium Luminescent Switch-on Molecular Probe

Yu-Jing Lu^{a,*}, Dong-Ping Hu^a, Qiang Deng^a, Zheng-Ya Wang^a, Bao-Hua Huang^a,

Yan-Xiong Fang^a, Kun Zhang^{a,b,*}, Wing-Leung Wong^{c,*}

^a Institute of Natural Medicine and Green Chemistry, School of Chemical Engineering and Light Industry, Guangdong University of Technology, Guangzhou 510006, P.R. China.

^b School of Chemical and Environment Engineering, Wuyi University, Jiangmen 529020, P.R. China.

^c Department of Science and Environmental Studies, Centre for Education in Environmental Sustainability, The Hong Kong Institute of Education, 10 Lo Ping Road, Tai Po, Hong Kong SAR, P.R. China.





Fig. S1 The ¹H NMR (A), ¹³C NMR (B) and ESI-MS (C) spectra of **DID-VP**.



Fig. S2 Fluorescence spectra of 2.5 μM DID-VP binding with: ON1 (G-quadruplex DNA),

ON1-ON2 (duplex DNA) and ON2 (single stranded DNA) at 2.5 $\mu M.$



Fig. S3 The melting profiles of 400 nM F21T G-quadruplex DNA (A) and 400 nM duplex DNA F10T (B) with increasing concentrations of **DID-VP**.



Fig. S4 Luminescence intensity of the system ([DID-VP] = $2.0 \ \mu$ M, [UDG] = $5.0 \ U/mL$) in the presence of duplex substrate or ON1m-ON2 duplex. Error bars represent the standard deviations of the results from three independent experiments.



Fig. S5 Luminescence response of the system with the complex alone ([DID-VP] = $2.0 \ \mu$ M) and in the presence of increasing concentrations of UDG (0, 10, 20, and 30 U/mL).



Fig. S6 Non-denaturing gel electrophoresis of samples in 20% polyacrylamide gel. Lane 1 contains 15.0 μ M ON1. Lane 2 contains 10 μ M ON2. Lanes 3 contains 15.0 μ M ON1-ON2 duplex substrate without UDG. Lane 4 contains 15.0 μ M ON1-ON2 duplex substrate with 5.0 U/mL UDG. The separated products were visualized by SYBR green I staining (A); and **DID-VP** staining (B).



Fig. S7 Circular dichroism (CD) spectrum of blank (red) or 5.0 U/mL UDG (black) recorded in Tris buffer (50 mM Tris, pH 7.0).



Fig. S8 Circular dichroism (CD) spectrum of 2.5 μ M ON1-ON2 duplex substrate in the absence (red) or presence (black) of 5.0 U/mL UDG recorded in Tris buffer.



Fig. S9 Relative luminescence intensity at 585 nm of the system ([ON1-ON2] = 1.5μ M, [UDG] = 2.5 U/mL) in the presence of different concentrations of **DID-VP** (0.25, 0.5, 1.0, 2.0 and 3.0 μ M) in aqueous buffer solution (50 mM Tris, 20 mM KCl, 50 mM NH₄OAc, pH 7.0). Error bars represent the standard deviations of the results from three independent experiments.



Fig. S10 Relative luminescence intensity at 585 nm of the system ([**DID-VP**] = 2.0 μ M, [UDG] = 2.5 U/mL) at various concentrations of ON1-ON2 (0.25, 0.5, 1.5 and 3 μ M) in aqueous buffer solution (50 mM Tris, pH 7.0). Error bars represent the standard deviations of the results from three independent experiments.



Fig. S11 Relative luminescence intensity at 585 nm of the system ([ON1-ON2] = 0.25μ M, [UDG] = 2.5 U/mL, [**DID-VP**] = 2.0 μ M) at various concentrations of KCl (0, 20, 50, 100 mM). Error bars represent the standard deviations of the results from three independent experiments.



Fig. S12 Relative luminescence intensity at 585 nm of the system ([ON1-ON2] = 0.25μ M, [UDG] = 2.5 U/mL, [DID-VP] = 2.0 μ M) at various concentrations of NH₄OAc ([NH₄OAc] = 0, **50**, 150, 300 mM). Error bars represent the standard deviations of the results from three independent experiments.



Fig. S13 Relative luminescence intensity at 585 nm of the system ([ON1-ON2] = 0.25μ M, [UDG] = 2.5 U/mL, [DID-VP] = 2.0 μ M) by varying the pH of the buffered system (50 mM Tris, 20 mM KCl, 50 mM NH₄OAc). Error bars represent the standard deviations of the results from three independent experiments.