

Electronic Supplementary Information

A graphene oxide platform for assay of DNA 3'-phosphatases and their inhibitors based on hairpin primer and polymerase elongation

Wenping Zhu, Ziwei Zhao, Zhen Li, Jianhui Jiang*, Guoli Shen and Ruqin Yu*

*State Key Laboratory of Chemo/Bio-Sensing and Chemometrics, College of
Chemistry and Chemical Engineering, Hunan University, Changsha, 410082, P. R.
China*

To whom correspondence should be addressed.

Tel: +86 731 88822577.

Fax: +86 731 88822577.

E-mail address: rgyu@hnu.edu.cn (R.-Q. Yu)

jianhuijiang@hnu.edu.cn (J.-H.Jiang),

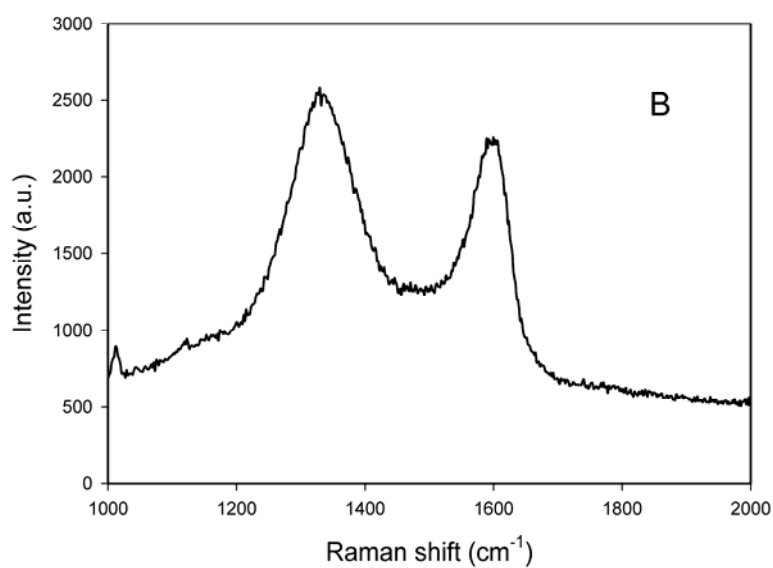
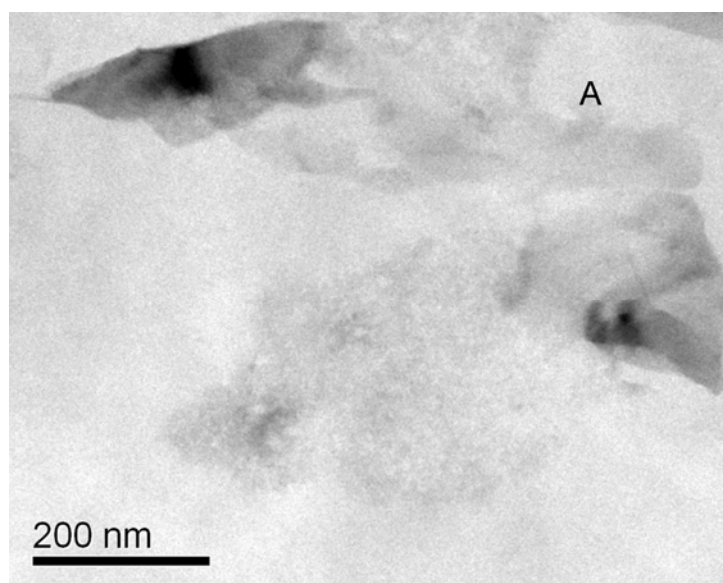


Figure S1. HRTEM image (A), Raman spectra (B) of GO. As shown in Figure S1B, two distinct peaks at 1329 and 1600 cm^{-1} correspond to the D and G bands, respectively.

1. Optimization of the concentration of HP

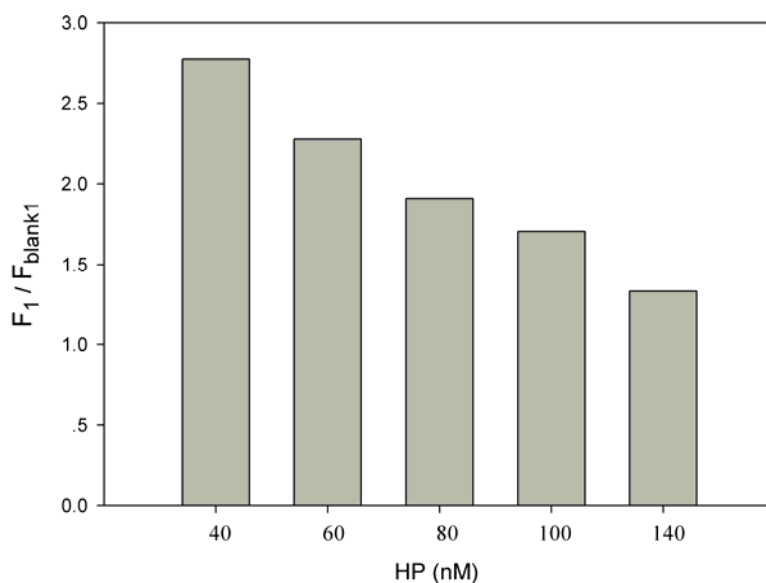


Figure S2. Dependency of F_1/F_{blank1} values on the concentration of HP for assaying T4 PNKP. F_1/F_{blank1} is defined as the ratio of fluorescence peak intensity at 524 nm from 4 U/mL T4 PNKP (F_1) to that from no addition of T4 PNKP (F_{blank1}).

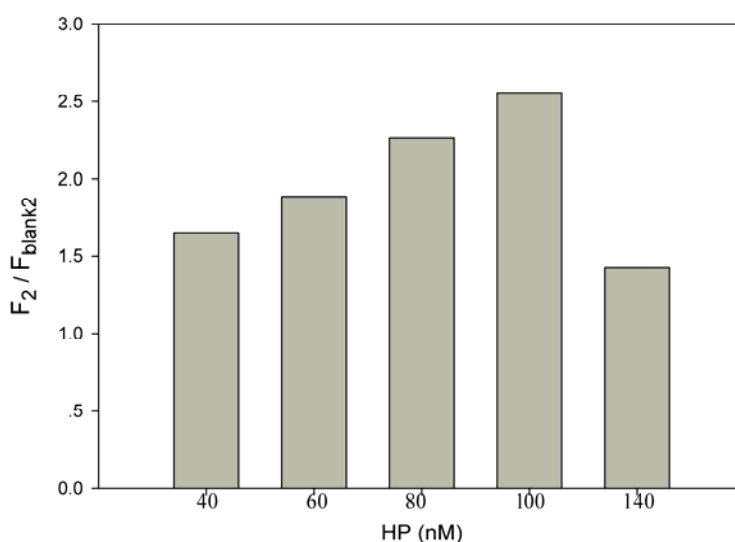
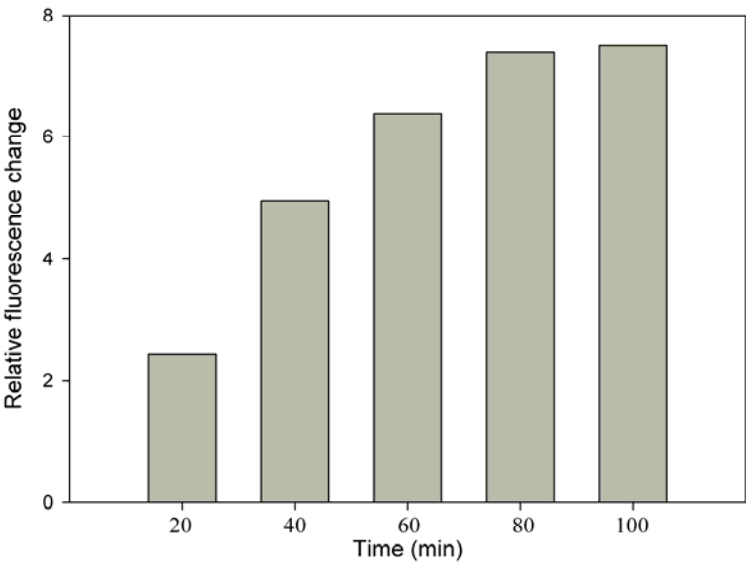
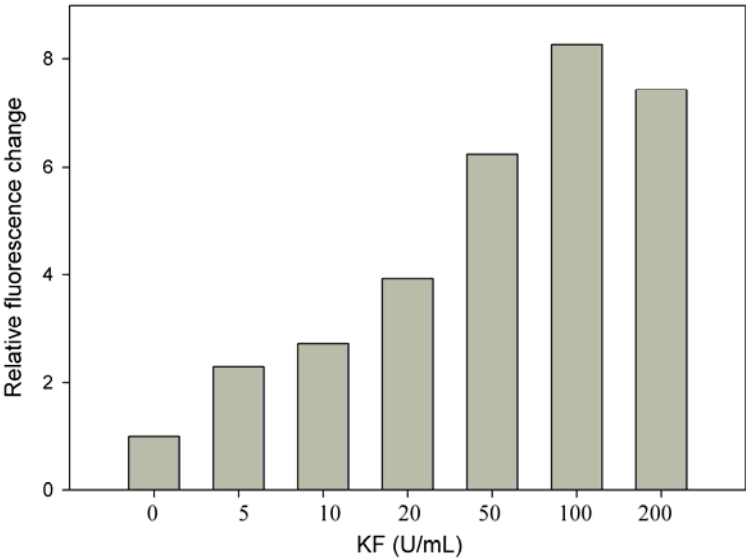


Figure S3. Dependency of F_2/F_{blank2} values on the concentration of HP for assaying SAP. F_2/F_{blank2} is defined as the ratio of fluorescence peak intensity at 524 nm from 0.1 U/mL SAP (F_2) to that from no addition of SAP (F_{blank2}).

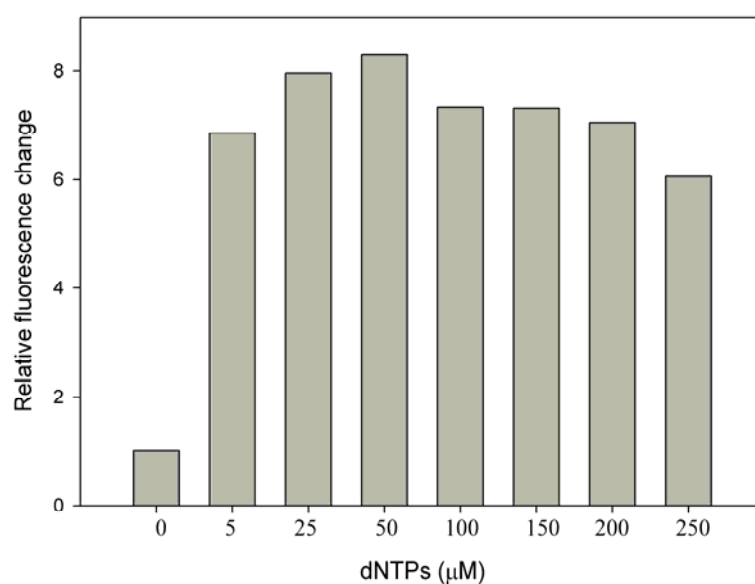
43 **2. Optimization of experimental conditions for assaying T4 PNKP**



(a)



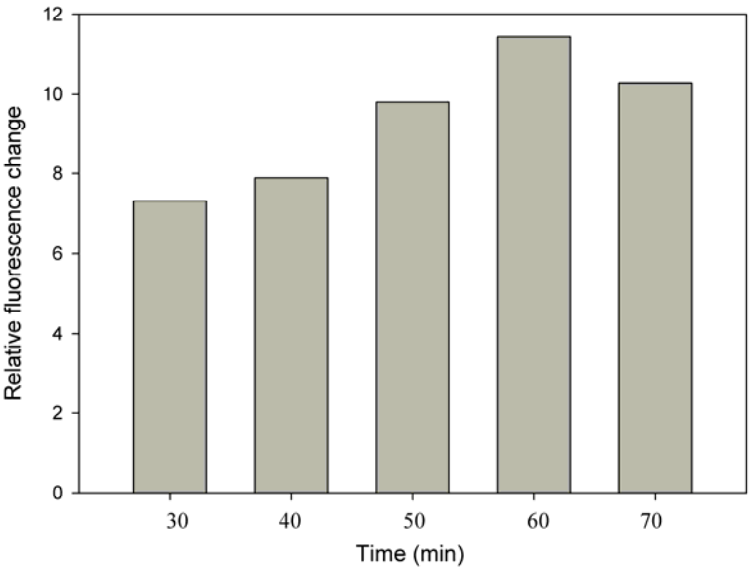
(b)



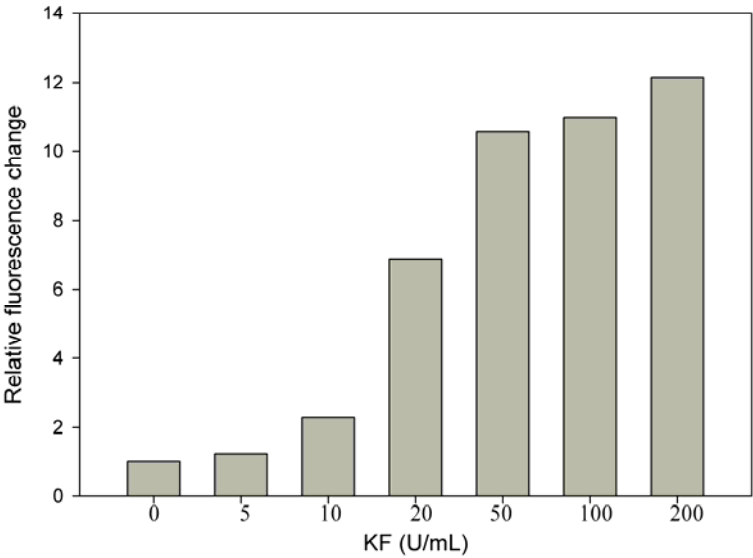
(c)

Figure S4. (a) The effect of reaction time on the relative fluorescence change of the sensing system. (b) The effect of the concentration of KF polymerase on the relative fluorescence change of the sensing system. (c) The effect of the concentration of dNTPs on the relative fluorescence change of the sensing system. The relative fluorescence change is defined as the ratio of fluorescence peak intensity at 524 nm from T4 PNKP (40 U/mL) to that from no addition of T4 PNKP.

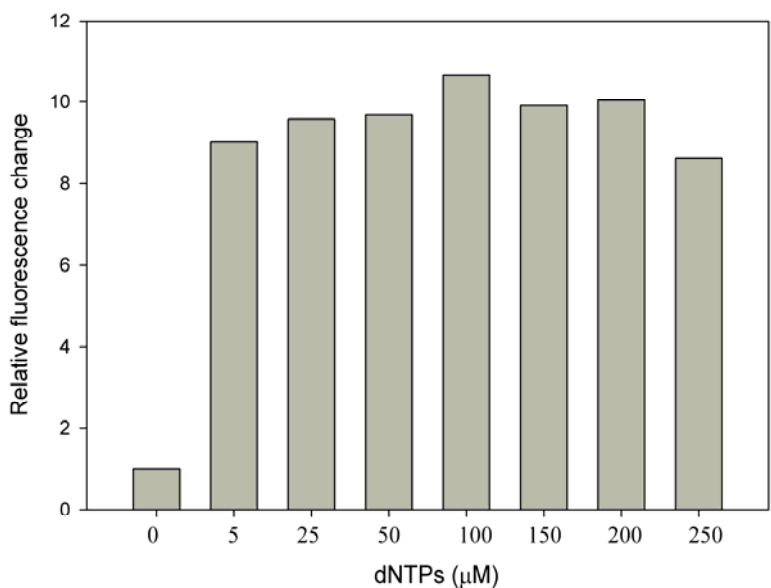
57 **3. Optimization of experimental conditions for assaying SAP**



(a)



(b)



(c)

Figure S5. (a) The effect of incubation time of the dephosphorylation on the relative fluorescence change of the sensing system. (b) The effect of the concentration of KF polymerase on the relative fluorescence change of the sensing system. (c) The effect of the concentration of dNTPs on the relative fluorescence change of the sensing system. The relative fluorescence change is defined as the ratio of fluorescence peak intensity at 524 nm from SAP (2 U/mL) to that from no addition of SAP.

4. Study of GO as an efficient signal-to-background enhancer

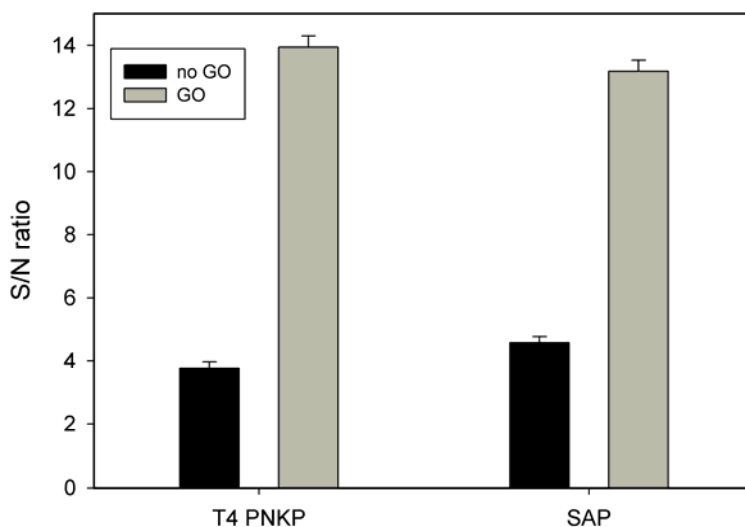
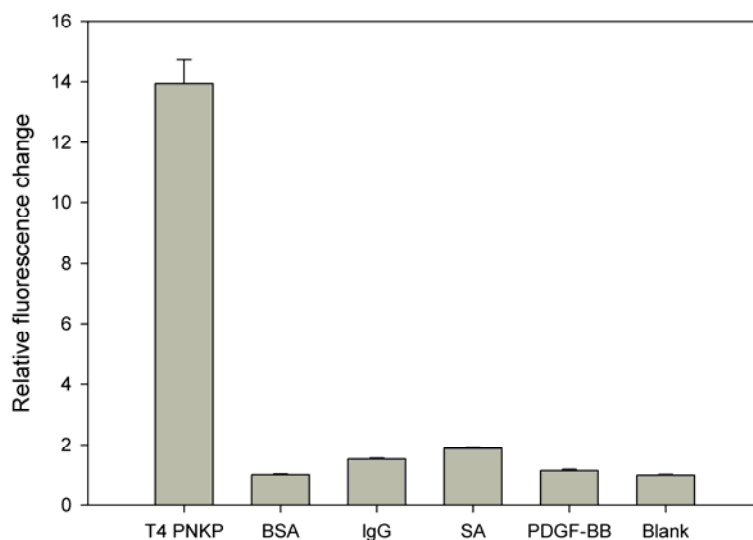


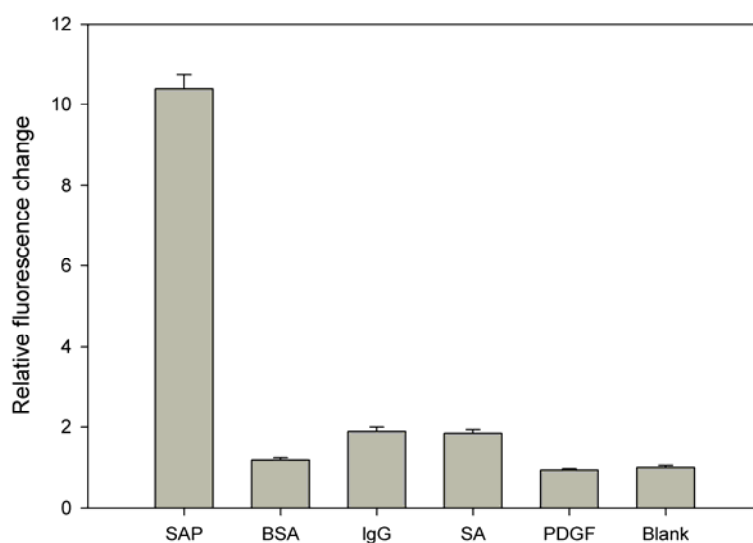
Figure S6 Bar graphic of the S/N ratio without addition of GO compares to the addition of GO (under the optimized experimental conditions). The S/N ratio is defined as the ratio of fluorescence peak intensity at 524 nm from T4 PNKP (100 U/mL) or SAP (10 U/mL) to that from no addition of DNA 3'-phosphatases.

78 5. Selectivity of the proposed strategy



79
80 **Figure S7.** Selectivity of the sensing platform towards T4 PNKP compared to other
81 interfering proteins. The concentration of T4 PNKP is 100 U/mL whereas the
82 concentration for other interfering proteins like BSA, IgG, PDGF-BB and SA are all
83 0.2 μ M. The “Blank” at the X-axis is defined as the relative fluorescence change
84 without addition of T4 PNKP.

85



86

87 **Figure S8.** Selectivity of the sensing platform towards SAP compared to other
88 interfering proteins. The concentration of SAP is 2 U/mL whereas the concentration
89 for other interfering proteins like BSA, IgG, PDGF-BB and SA are all 0.2 μ M. The
90 “Blank” at the X-axis is defined as the relative fluorescence change without addition
91 of SAP.

92

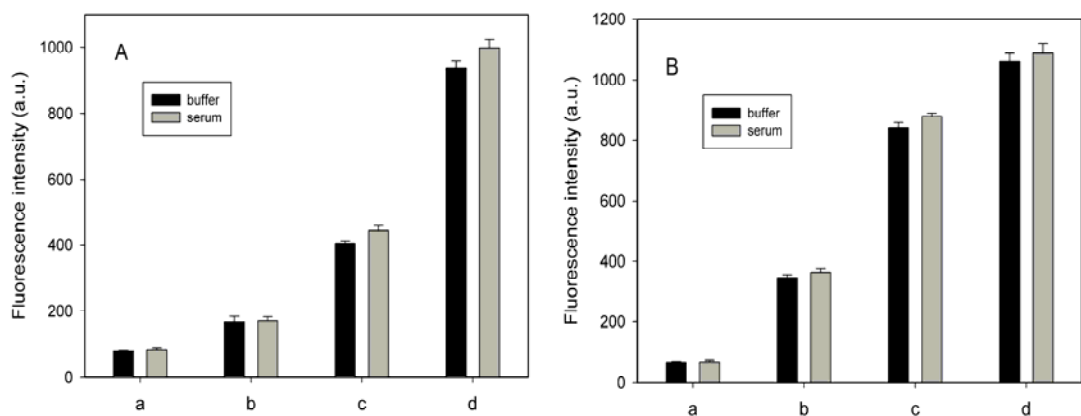


Figure S9 Bar plot of corresponding fluorescence responses at 524 nm of the DNA 3'-phosphatase assay in diluted serum (gray) or in 1× NEBuffer 2 (black). (A) T4 PNKP assay in 0.5% serum (200 fold diluted) or 1× NEBuffer 2 under its optimal experimental conditions in the absence of T4 PNKP (a) or in the presence of three random concentrations of T4 PNKP (b), (c) and (d) followed by incubating with 8 µg/mL GO. (B) SAP assay in 1% serum (100 fold diluted) or 1× NEBuffer 2 under its optimal experimental conditions in the absence of SAP (a) or in the presence of three random concentrations of SAP (b), (c) and (d) followed by incubating with 14 µg/mL GO.