## Monitoring and Inhibition of Plk1: Amphiphilic porphyrin conjugated Plk1 specific peptides for its imaging and anti-tumor function

Hongguang Li,<sup>*a*</sup> Chi-Fai Chan,<sup>*a*</sup> Wai-Lun Chan,<sup>*a*</sup> Sam Lear,<sup>*b*</sup> Kwok-Keung Shiu,<sup>*a*</sup> Steven L. Cobb,<sup>*b*</sup> Nai-Ki Mak,<sup>*c*</sup> Terrence Chi-Kong Lau,<sup>*d*</sup> Rongfeng Lan,<sup>*e*\*</sup> Wai-Kwok Wong,<sup>*a*\*</sup> and Ka-Leung Wong <sup>*a*\*</sup>

<sup>*a*</sup> Department of Chemistry, Hong Kong Baptist University, Kowloon Tong, Hong Kong SAR; <sup>*b*</sup> Department of Chemistry, Durham University, Durham, DH1 3LE, UK; <sup>*c*</sup> Department of Biology, Hong Kong Baptist University, Kowloon Tong, Hong Kong SAR; <sup>*d*</sup> Department of Biology and Chemistry, City University of Hong Kong, Kowloon Tong, Hong Kong SAR; <sup>*e*</sup> Hong Kong Baptist University Institute of Research and Continuing Education, Shenzhen, P. R. China.

## Supporting Information



Figure S1. The synthetic scheme for Por-P1 and Por-P2.



**Figure S2**. HPLC trace of **P1**- condition 10% A (ACN+0.1%TFA) + 90% B (H<sub>2</sub>O+0.1%TFA).



**Figure S3.** HPLC trace of **P2**- condition 10% A (ACN+0.1%TFA) + 90% B (H<sub>2</sub>O+0.1%TFA).



Figure S4. ESI-MS spectrum of P1.



Figure S5. ESI-MS spectrum of P2.



FigureS7. <sup>13</sup>C-NMR spectrum of Por-NH<sub>2</sub>.







FigureS9. <sup>13</sup>C-NMR spectrum of Por-COOH.



Figure S10. The binding fitting via molecular modeling for the comparisons of interactions between (a) P1, (b) P2, (c) Por-P1, (d) Por-P2 and Plk1 structure.



FigureS11. Purified Plk1 (PBD domain) in protein gel stained with coomassie blue.



**Figure S12**. The UV absorption spectra of Por-COOH, Por-**P1** and Por-**P2** (10<sup>-5</sup>M) in HEPES buffer (10 mM HEPES, pH=8.0, 150 mM NaCl).



Figure S13. The emission spectra of Por-COOH, Por-P1 and Por-P2 in HEPES buffer. ( $\lambda_{ex} = 430 \text{ nm} \text{ and } 10^{-6}\text{M}$ )

![](_page_7_Figure_0.jpeg)

**FigureS14**. The emission titration of Por-P1 (1  $\mu$ M) upon addition of Plk1 (2 nM - 200 nM) in HEPES buffer. ( $\lambda_{ex} = 427$  nm)

![](_page_7_Figure_2.jpeg)

Figure S15. The emission change of Por-P1 (1  $\mu$ M) upon addition of Zn<sup>2+</sup>, Cu<sup>2+</sup> and HSA in HEPES. ( $\lambda_{ex} = 427 \text{ nm}$ )

![](_page_8_Figure_0.jpeg)

**Figure S16**. The emission change of Por-**P2** (1  $\mu$ M) upon addition of Zn<sup>2+</sup>, Cu<sup>2+</sup> and HSA in HEPES buffer. ( $\lambda_{ex} = 427 \text{ nm}$ )

![](_page_8_Figure_2.jpeg)

**Figure S17**. Representative Half-Offset histograms of HeLa cells analyzed by Flow cytometry after treated with Por-**Pn**. Figures were processed by using FlowJo 7.6.1. Por-**P1** or Por-**P2** cause the HeLa cells arrested in G2/M phase in a concentration-dependent manner, with an obvious G2/M peak in the concentration of 20  $\mu$ M.

![](_page_9_Figure_0.jpeg)

**Figure S18**. Raw data of MRC-5 cells analyzed using Flow cytometry and presented by Half-Offset histograms from FlowJo 7.6.1. Parallel performed as in **Figure S17**.

![](_page_9_Figure_2.jpeg)

**Figure S19**. Por-**P1** and Por-**P2** treatments had induced Plk1 inhibition and succeeding G2/M arrest of HeLa cells. Experimentally, HeLa cells were microscopically imaged by Zeiss Axio Observer A1 (10 ×).Upon 10 or 20  $\mu$ M of Por-Pn treatments, parts of cells showed mitotic arrested, and cell growth was inhibited.

![](_page_10_Figure_0.jpeg)

**Figure S20**. Human normal lung fibroblasts MRC-5 cells were treated with Por-**P1** and Por-**P2** and then microscopically imaged. No obvious effects were detected under Por-**Pn** treatments on the MRC-5 cells.