***Supplementary Information***

# Beyond Telomeric G-quadruplexes: Remarkable Binding of PhenQE8 to Promoter Sequences

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## Spectral Data of Synthesized Compounds

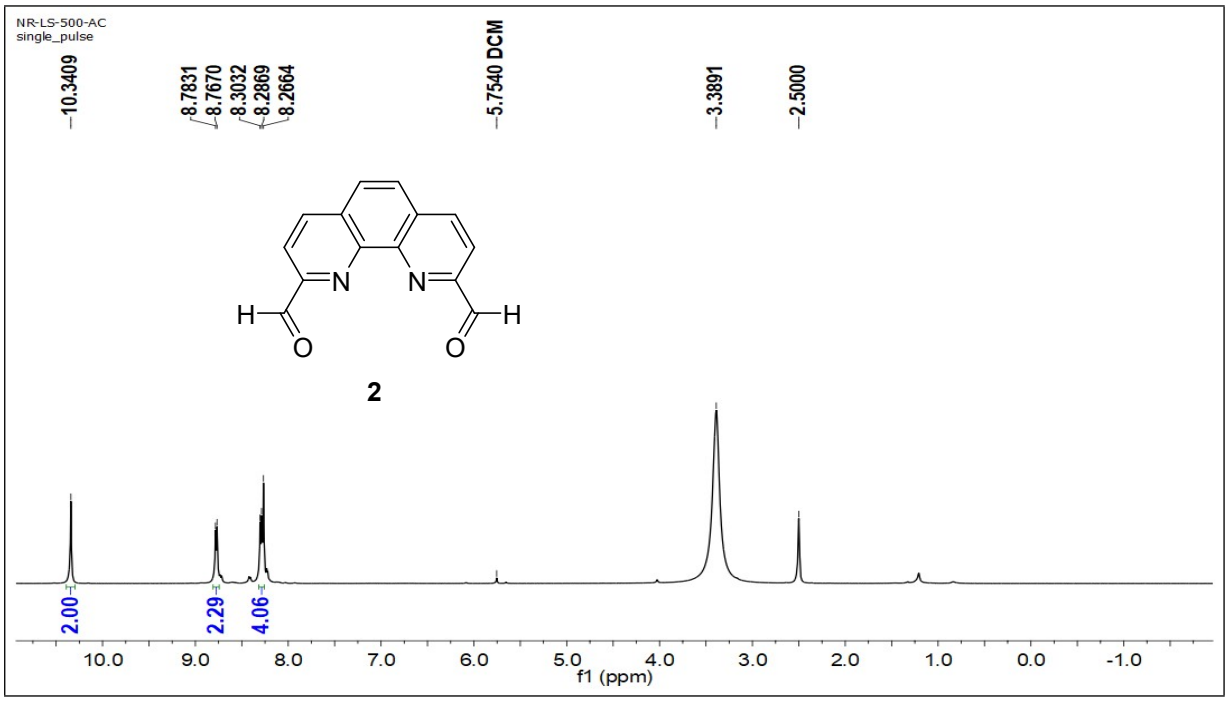



**Figure S1.** $^{1}$H NMR spectrum (500 MHz, DMSO-$d_6$) of compound **2**

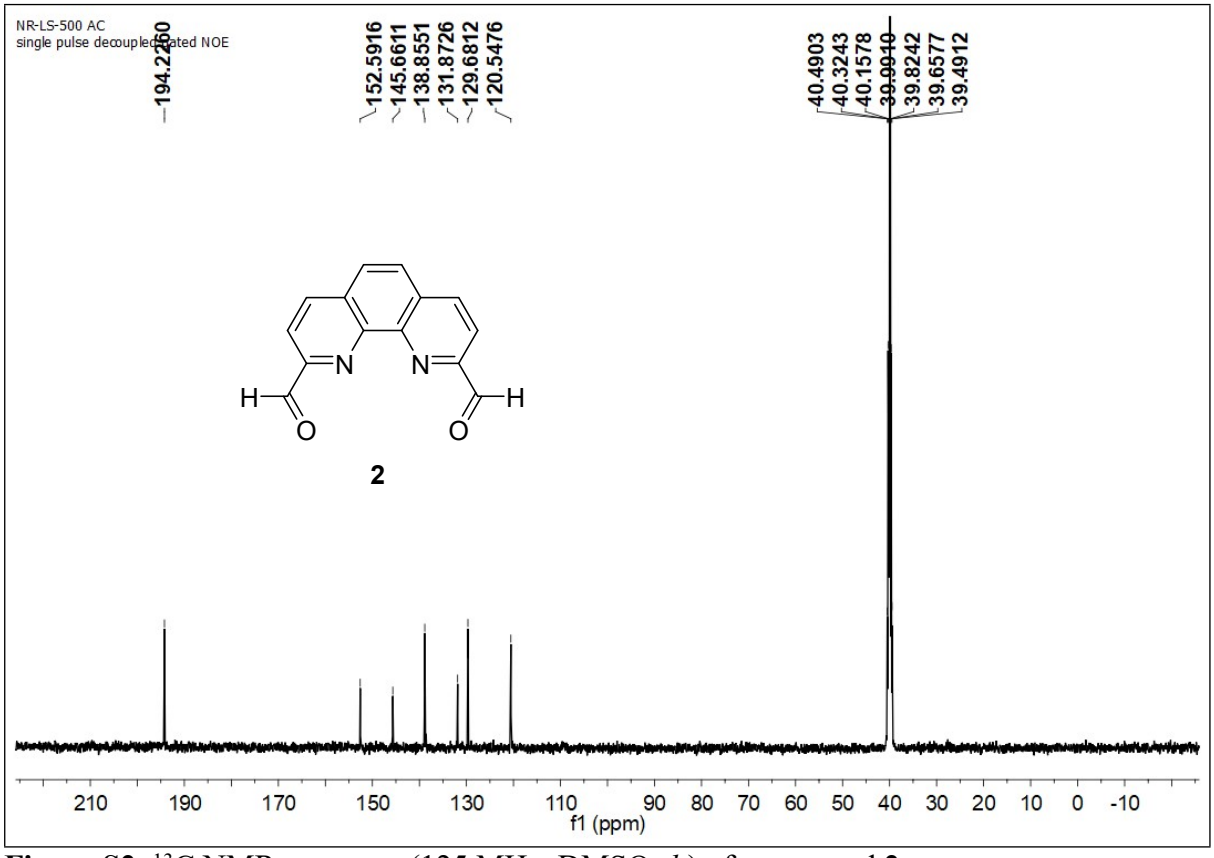



**Figure S2.** $^{13}$C NMR spectrum (125 MHz, DMSO-$d_6$) of compound **2**

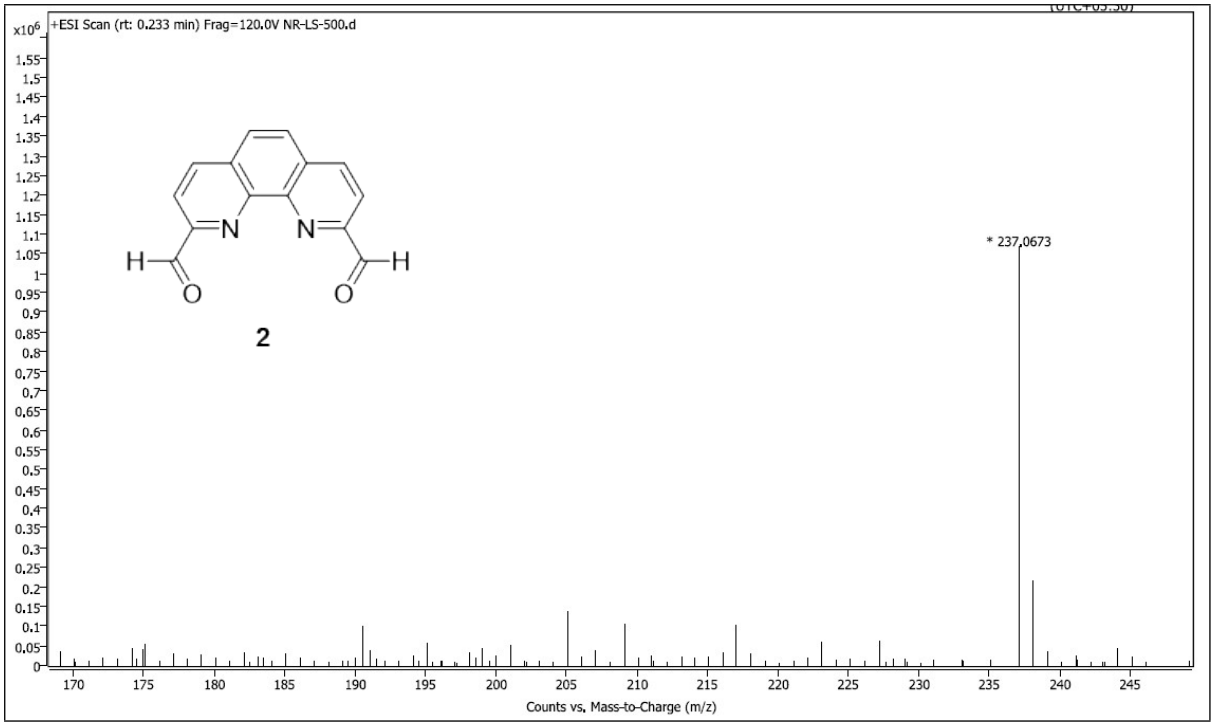



**Figure S3.** ESI-HRMS spectrum of compound **2**

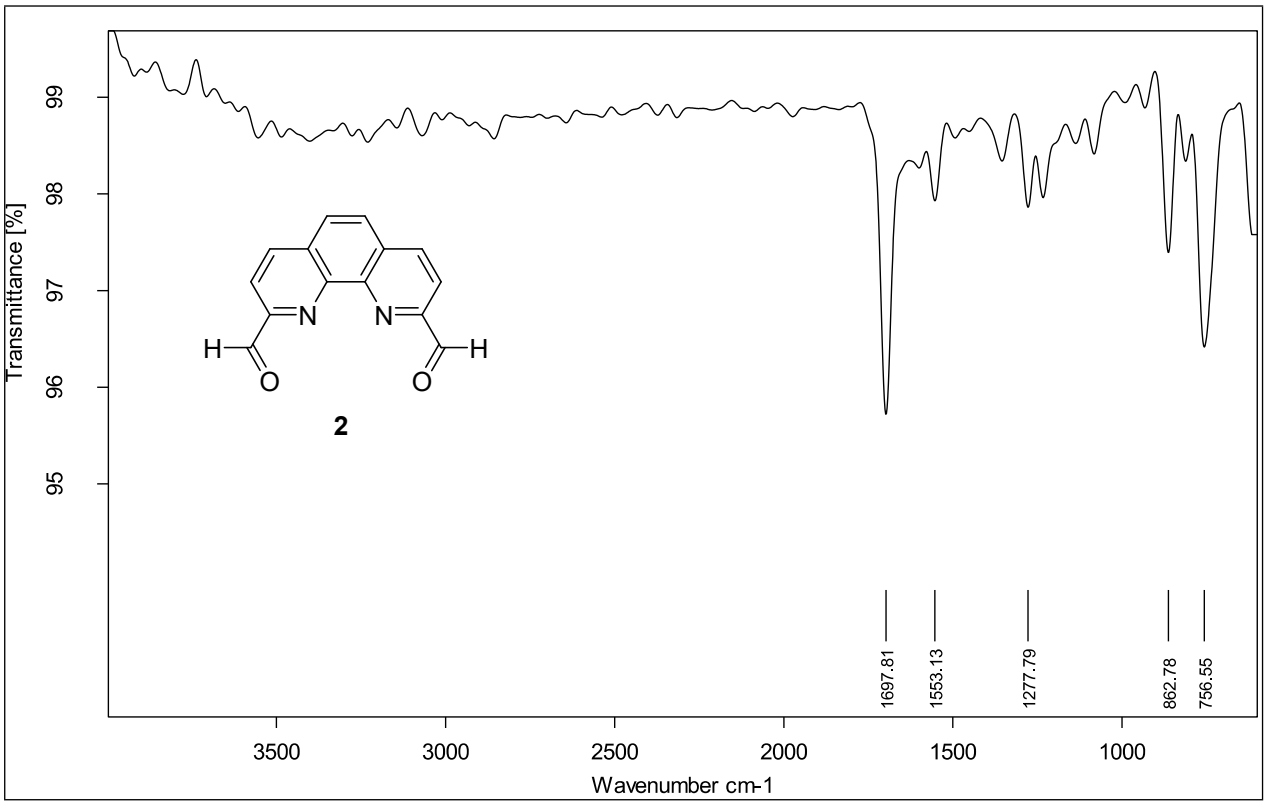



**Figure S4.** IR spectrum of compound **2.**

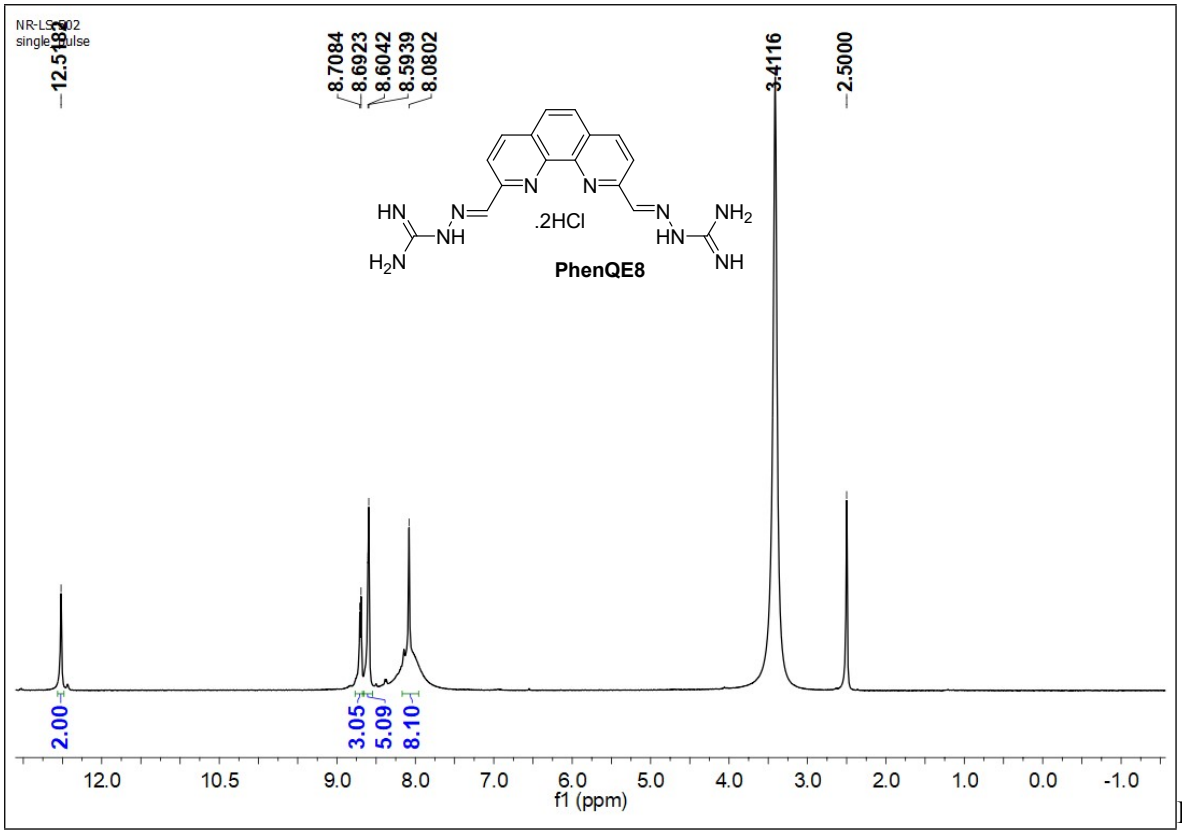



**Figure S5.** $^{1}$H NMR spectrum (500 MHz, DMSO-$d_6$) of PhenQE8.

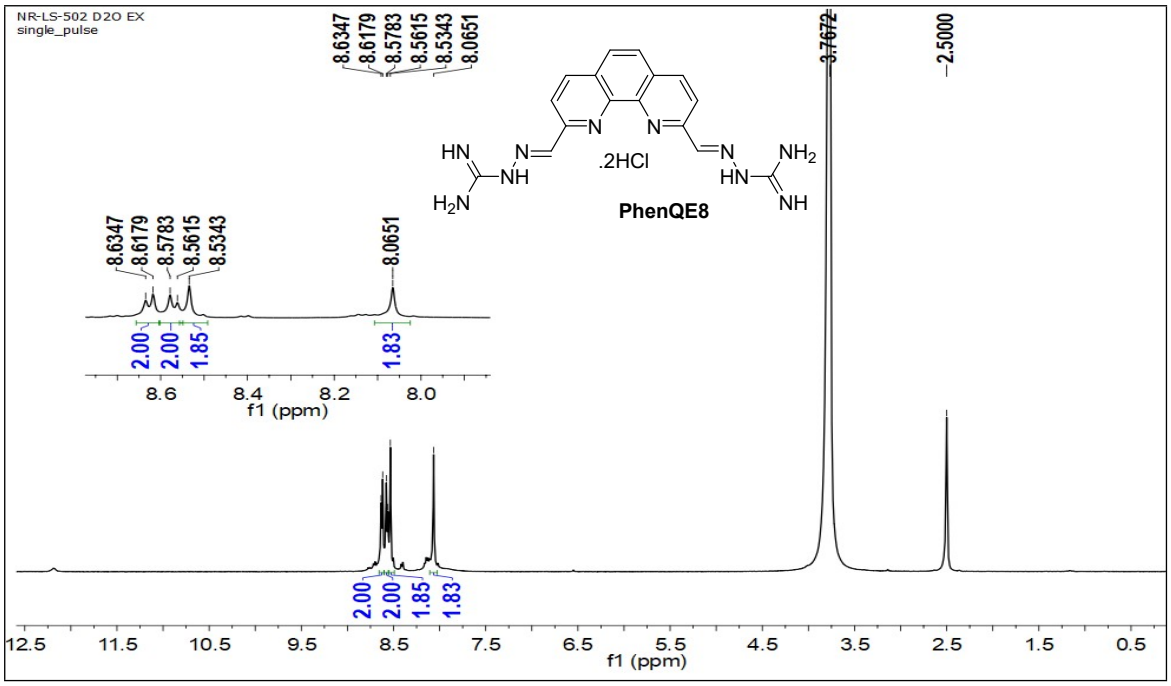



**Figure S6**. $D_2O$ Exchange $^{1}$H NMR spectrum (500 MHz, DMSO-$d_6$) of PhenQE8

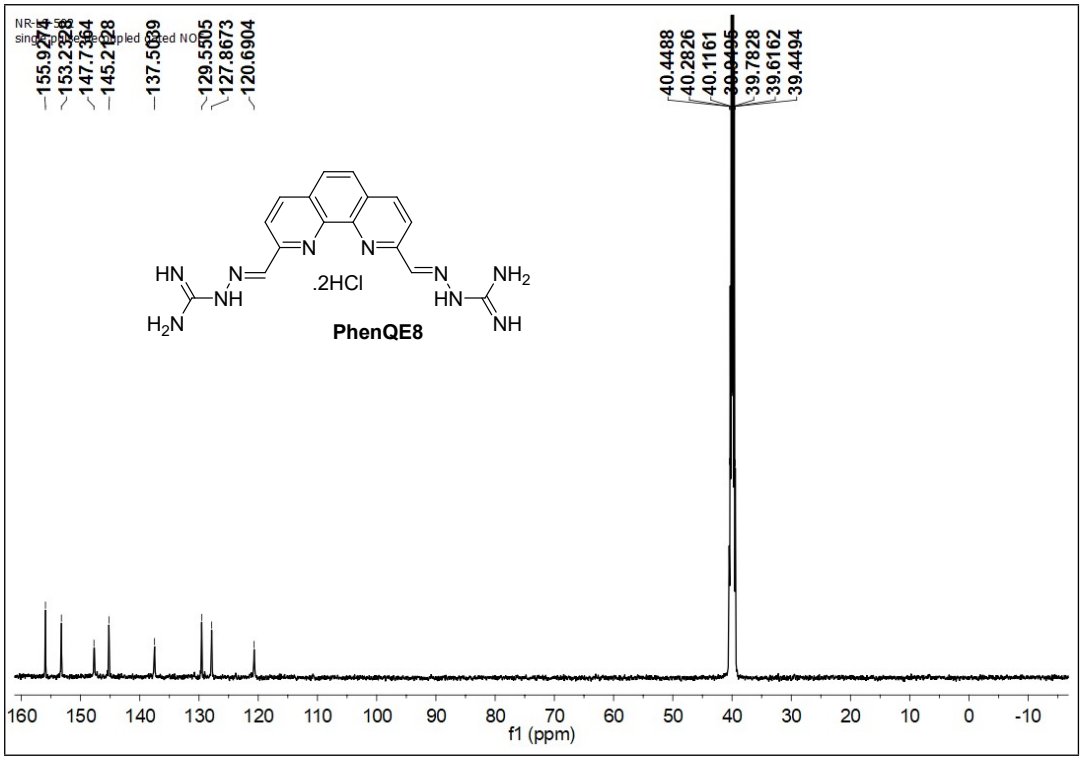



**Figure S7.** $^{13}$C NMR spectrum (125 MHz, DMSO-$d_6$) of PhenQE8**.**

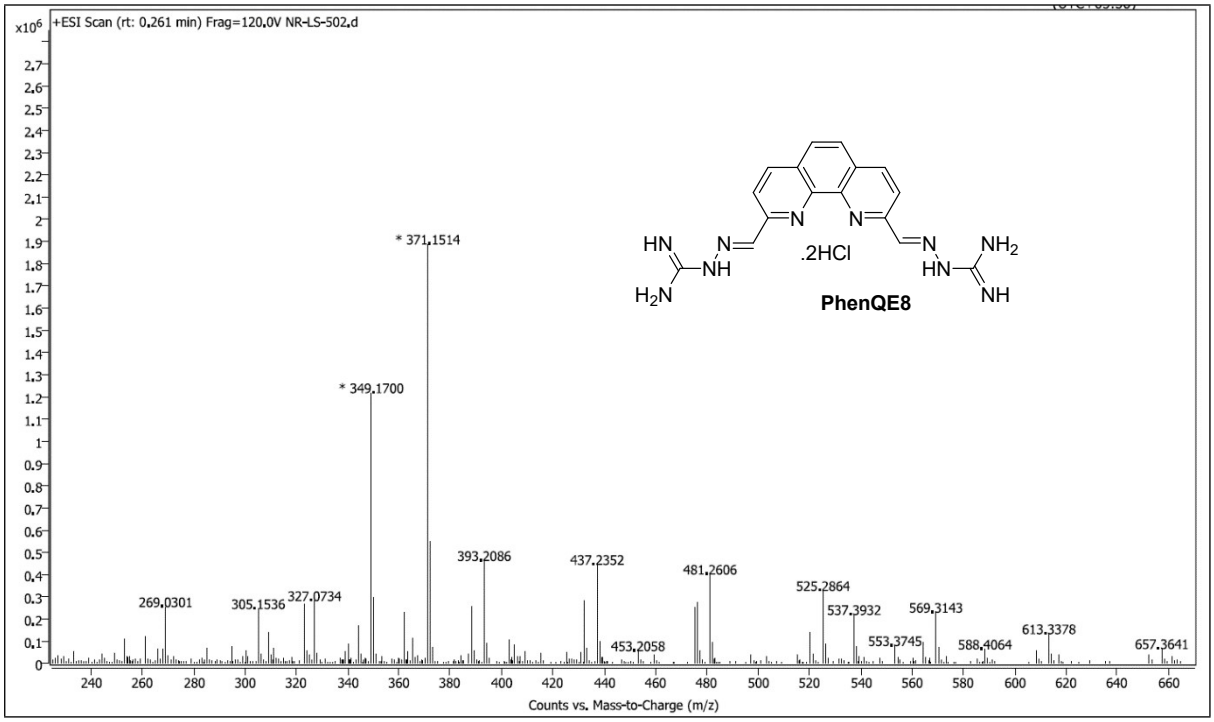
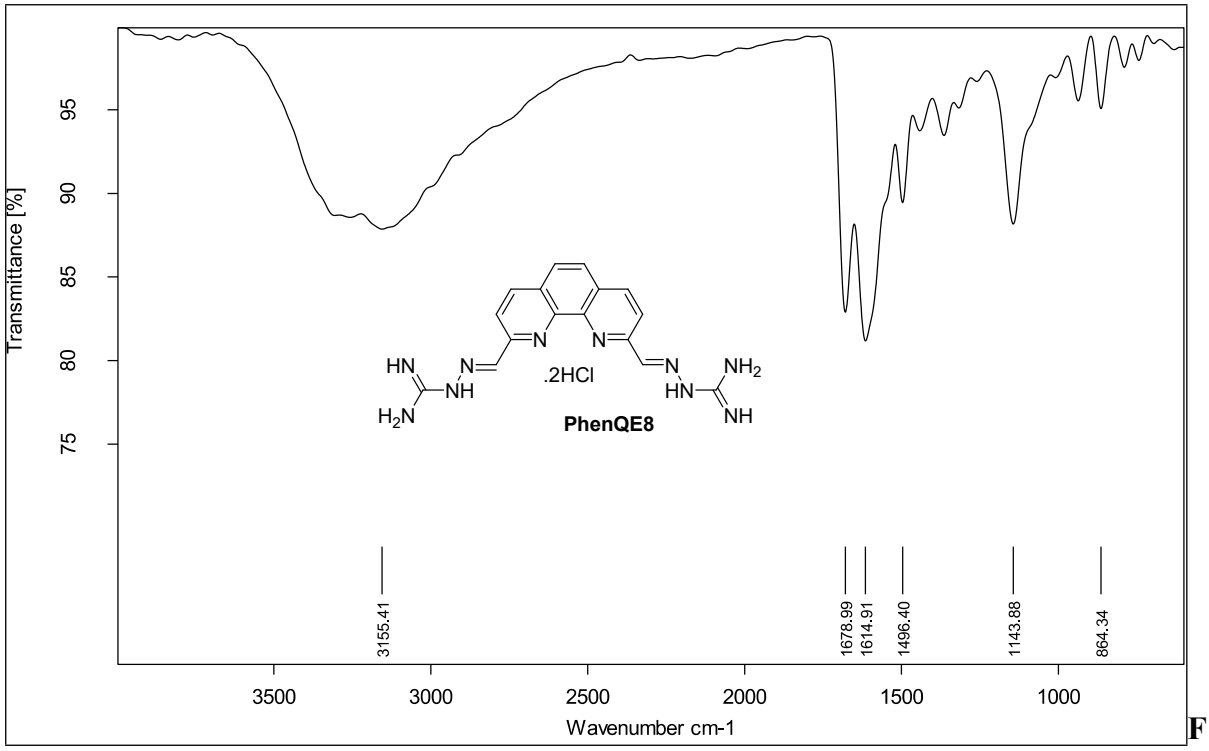



**Figure S8.** ESI-HRMS spectrum of PhenQE8.




**igure S9.** IR spectrum of PhenQE8**.**

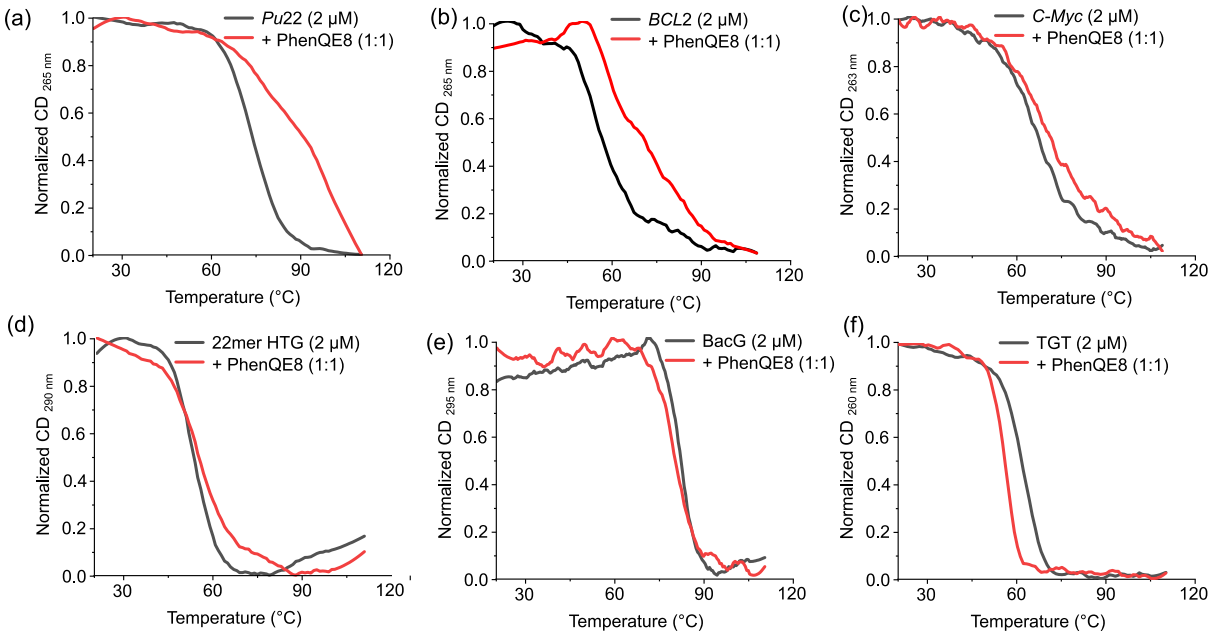



**Figure S10.** CD thermal melting profile of (a) *Pu*22 (b) *BCL2* (c) *c-Myc* (d) 22mer HTG (e) BacG, and (f) TGT in the absence and the presence of PhenQE8 at concentration of DNA and ligands as shown in the graph.

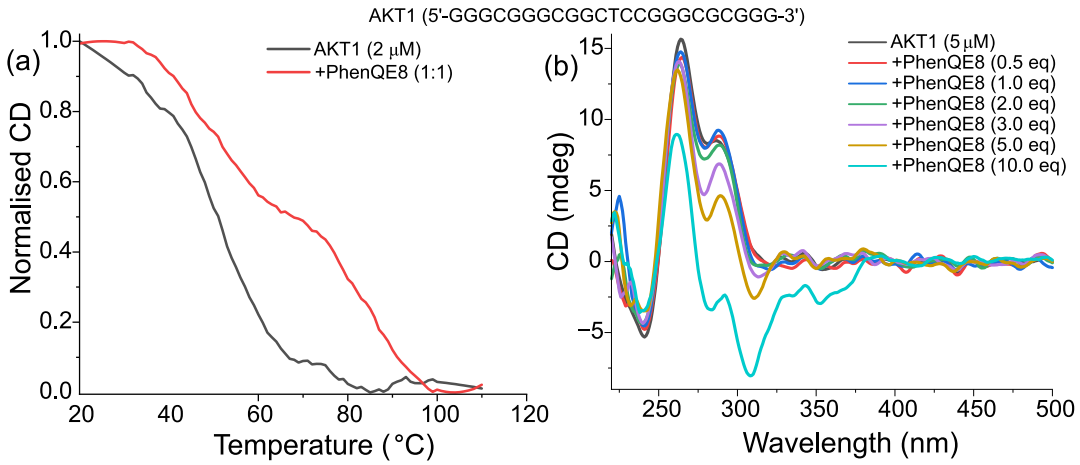



**Figure S11.** CD-spectroscopy study (a) CD thermal melting profile of AKT1, (b) CD titration profile of AKT1 with PhenQE8.

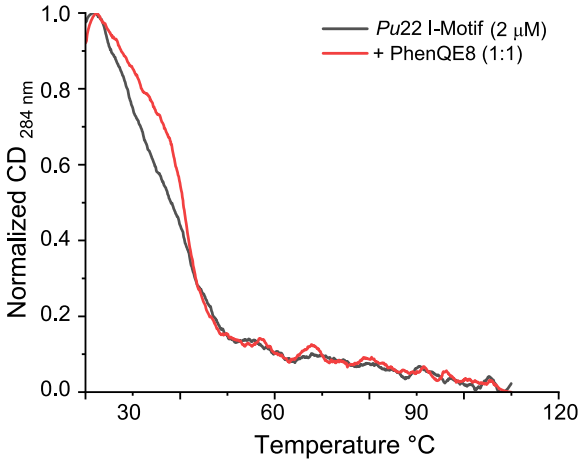



**Figure S12.** CD thermal melting profile of *Pu*22 i-motif in the absence and presence of PhenQE8 at different concentration of DNA and ligand as shown in the graph. Thermal denaturation of DNA sequences was performed from temperature 20-110 ºC at a heating rate of 1.0 ºC/minute. Melting experiments were performed in buffer 10 mM potassium phosphate, 0.5 mM EDTA, and 100 mM KCl at pH 5.5.

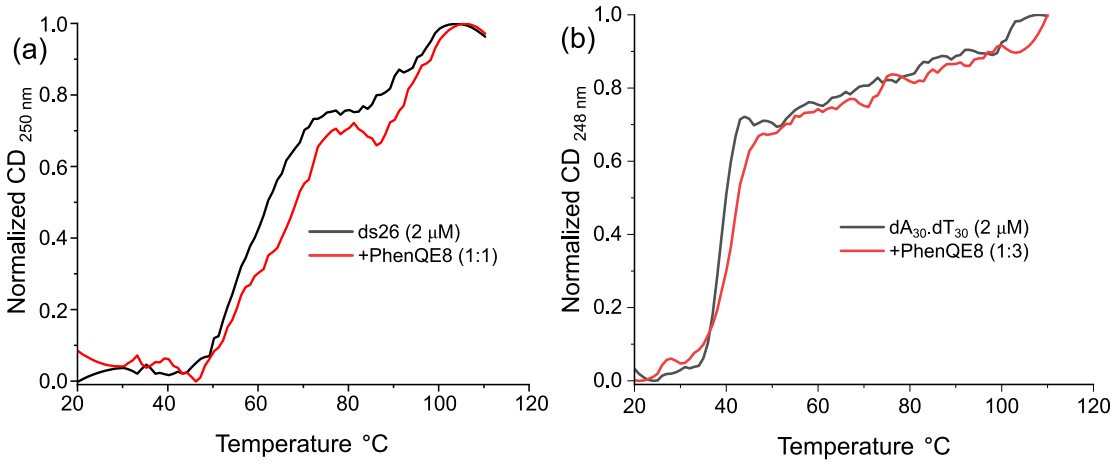



**Figure S13.** CD thermal melting profile of (a) ds26-duplex (b) $dA_{30}.dT_{30}$ in the absence and presence of PhenQE8 at different concentration of DNA and ligand as shown in the graph. Thermal denaturation of DNA sequences was performed from temperature 20-110 ºC at a heating rate of 1.0 ºC/minute. Melting experiments were performed in buffer 2 mM potassium phosphate, 0.1 mM EDTA, and 12 mM KCl at pH 7.0.

**Table S1. Melting temperature of nucleic acids in the presence and absence of PhenQE8.**

| Nucleic acids | DNA: Ligand Ratio | PhenQE8 | |
|---|---|---|---|
| | | $T_m$ (°C) | $\Delta T_m$ (°C) |
| **AKT1** | Control | 50.3 | - |
| | 1:1 | 69.9 | 19.6 |
| **ds26** | Control | 61.8 | - |
| | 1:1 | 68.1 | 6.3 |
| **$dA_{30}.dT_{30}$** | Control | 38.6 | - |
| | 1:3 | 40.6 | 2.0 |
| ***Pu*22 i-motif** | Control | 33.1 | - |
| | 1:1 | 38.3 | 5.2 |

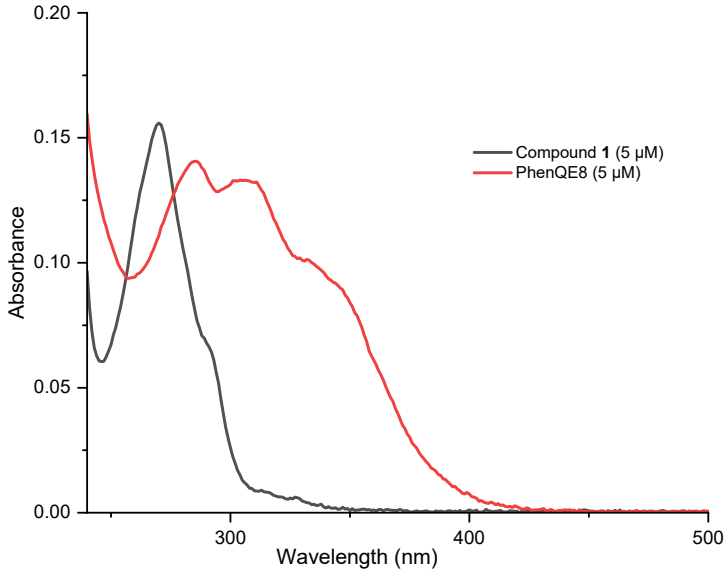



**Figure S14.** UV absorption spectra of compound **1** and PhenQE8 in 2 mM potassium phosphate, 0.5 mM EDTA and 12 mM KCl, having pH 7.0 at 25 °C. Absorption spectra were acquired at compounds concentration of 5 μM each and recorded between 200-800 nm and relevant data was plotted as absorbance *versus* wavelength plot.

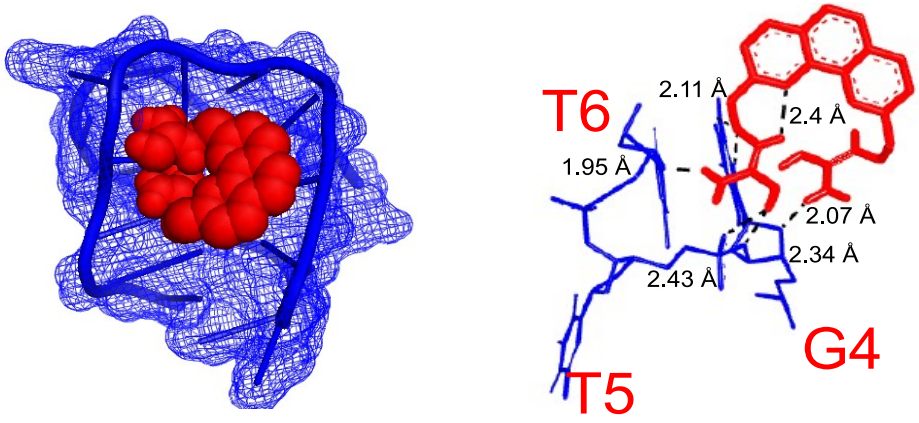



**Figure S15.** Molecular docking results showing (a) mode of binding and (b) key binding interaction of PhenQE8 with G-quadruplex (PDB ID: 143D).

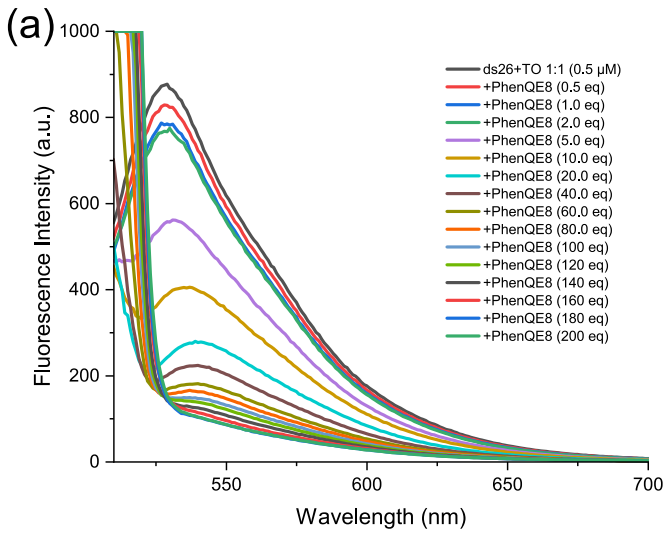



**Figure S16.** (a) Fluorescence intercalator displacement experiment of a complex of ds26 duplex DNA:TO (0.5 μM each) upon incremental addition of PhenQE8 **(**from 0.5 to 200 equivalents). The experiment was performed in buffer 2 mM potassium phosphate, 0.5 mM EDTA and 12 mM KCl having pH 7.0 at 20 °C. TO was excited at 501 nm and the emission spectrum was recorded between 510-700 nm.

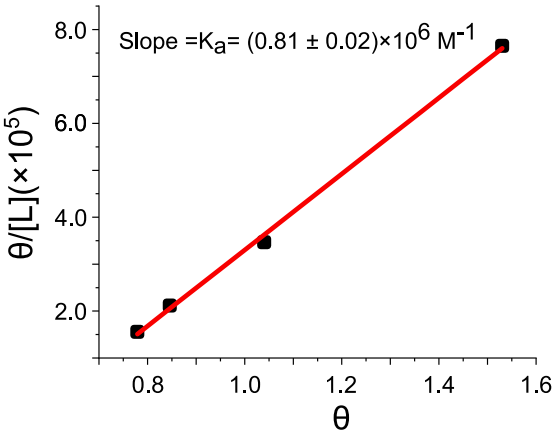



**Figure S17.** Scatchard plot analysis for determining the apparent binding constant ($K_a$). The $K_a$ can be calculated as the slope of plot between θ/[L] and θ. Where θ is ratio of change in UV-visible absorption upon binding to G-quadruplex DNA at saturation. i.e., $\theta = \Delta X/(\Delta X)_T$. where, X is the absorbance of PhenQE8 at 341 nm and $(\Delta X)_T$ the observed change at saturation.

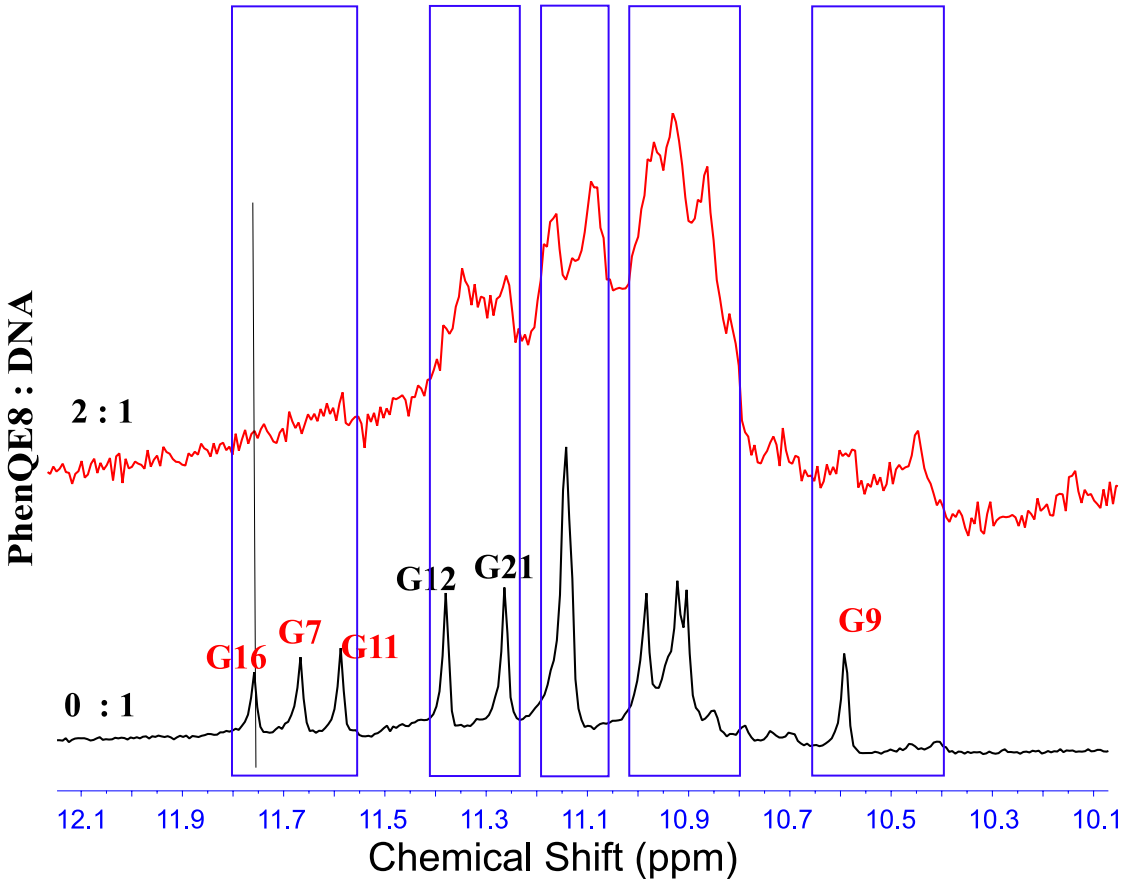



**Figure S18.** Imino proton region of the 1D $^{1}$H NMR titration spectrum of G-quadruplex with PhenQE8. The bases of *Pu*22 G-quadruplex upon addition of two equivalent PhenQE8, with clear perturbation are indicated in red and those with possibility of shift/merge are marked in black. Assay was performed in 10 mM $KH_2PO_4$ buffer (100 mM KCl, 10% $D_2O$, pH 7.4) at 25 °C. The final concentration of G-quadruplex DNA was 1 mM.