Supporting Information

First evidence of a functional interaction between DNA quadruplexes and Poly(ADP-ribose) polymerase-1

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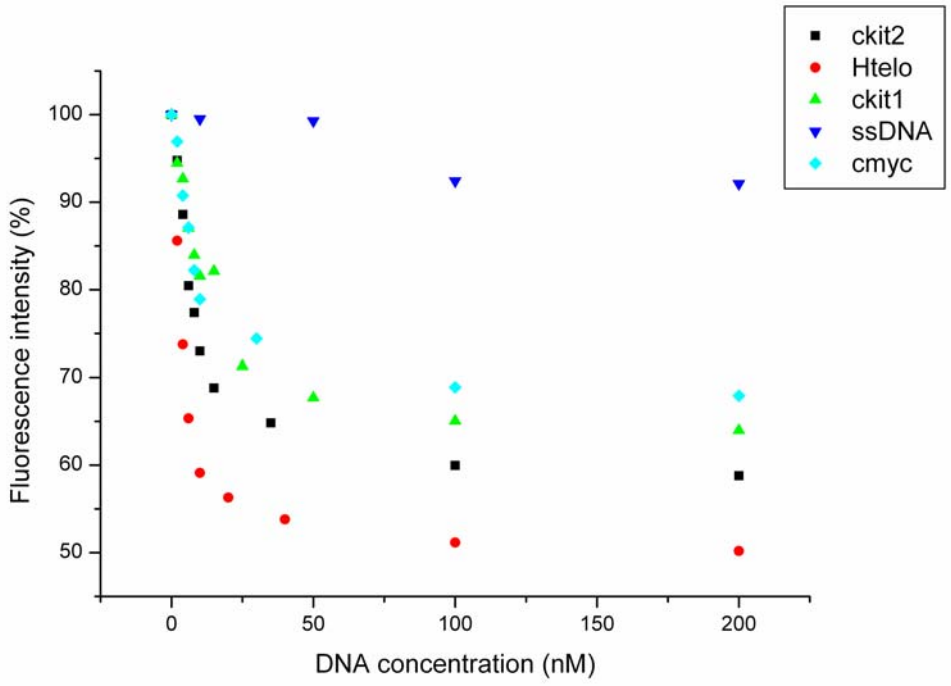
Fluorescence titration experiments of a hPARP-1 solution with single-stranded DNA and three different quadruplexes (c-kit-1, c-kit-2 and Htelo). To a 40 nM solution of hPARP-1 in 50 mM TRIS buffer pH 7.4 containing 100 mM KCl, 50 µM $Zn(OAc)_2$ and 1 mM DTT was titrated in a solution of DNA (single-stranded or quadruplex) in buffer and the fluorescence of hPARP-1 tryptophans was recorded at 350 nm (when exciting at 295 nm). The plots of the fluorescence intensity *vs* DNA concentration are shown in Supplementary Figure 1 below.



Supplementary Figure 1 *Fluorescence titration experiment of hPARP-1 (40 nM) with intramolecular DNA quadruplexes and single-stranded DNA. A specific fluorescence quenching is observed with DNA quadruplexes.*

The oligonucleotides sequences used for these fluorescence titration experiments are as indicated below:

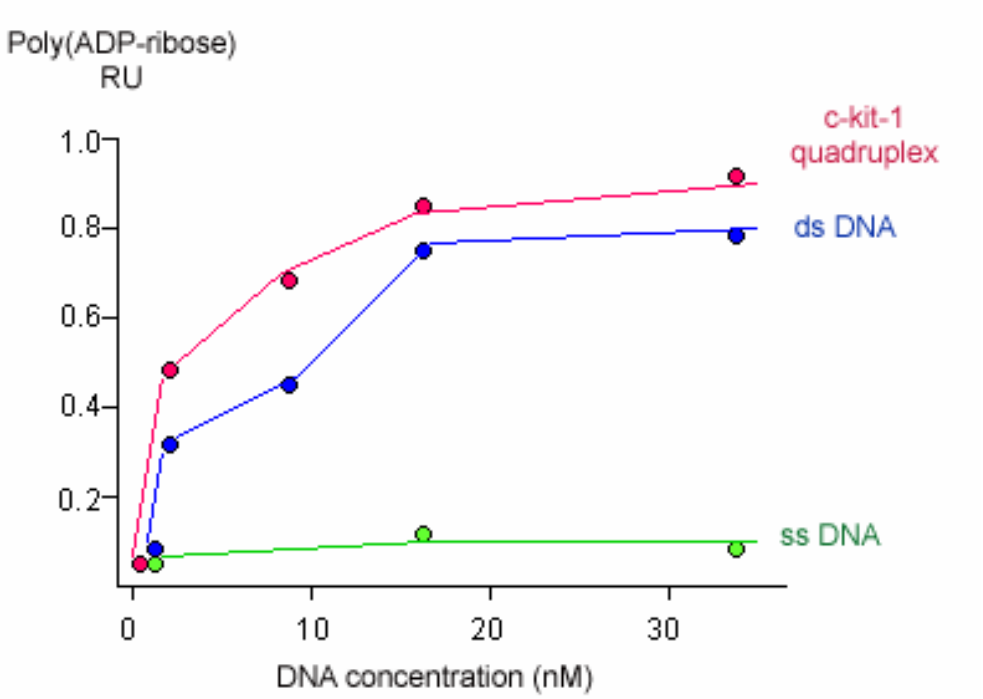
c-kit-2: AGG GAG GGC GCT GGG AGG AGG G

Htelo: GGG TTA GGG TTA GGG TTA GGG

ss-DNA: AGT GAG TGC TCT GTG AGG AGT G

c-myc: GGG TGG GGA GGG TGG G

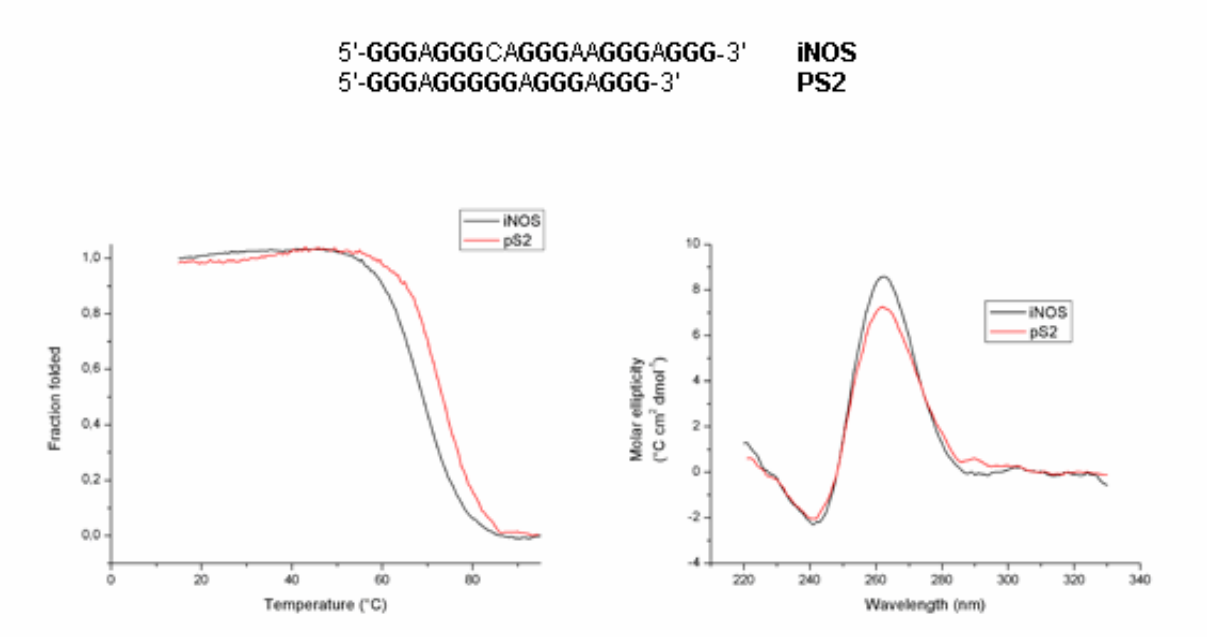
hPARP-1 activation upon binding to DNA quadruplexes and B-form duplex DNA ends. hPARP-1 activity is known to be stimulated by different types of DNA ligands including both non-B DNA structures and DNA strand interruptions. Using the reaction of hPARP-1 auto-modification, we compared the co-enzymatic efficiency of a double stranded DNA break (B-form of DNA) with that of the c-kit-1 DNA quadruplexes (non-B DNA). Supplementary Figure 2 shows that quadruplex DNA is an equally potent hPARP-1 activator as a B-form duplex DNA end. These data are in line with previously reported observations on high co-enzymatic efficiency of other non-B DNA structures.



Supplementary Figure 2. *hPARP-1 enzymatic activation by c-kit-1 DNA quadruplexes and duplex DNA end*. The hPARP-1 auto-poly(ADP-ribosyl)ation reactions were carried out at fixed hPARP-1 protein concentration (10 nM) in the presence of 133 µM NAD^+ and stimulated by increasing concentrations of either DNA quadruplex (**c-kit-1**), ssDNA oligonucleotide carrying mutated c-kit-1 sequence (**ssDNA**) to prevent quadruplex formation, or a linear ds DNA fragment (301-bp PvuII-PvuII fragment of pUC8) modified with streptavidin to carry only one dsDNA end unprotected as a single hPARP-1 binding site (see reference 27 from main text). The

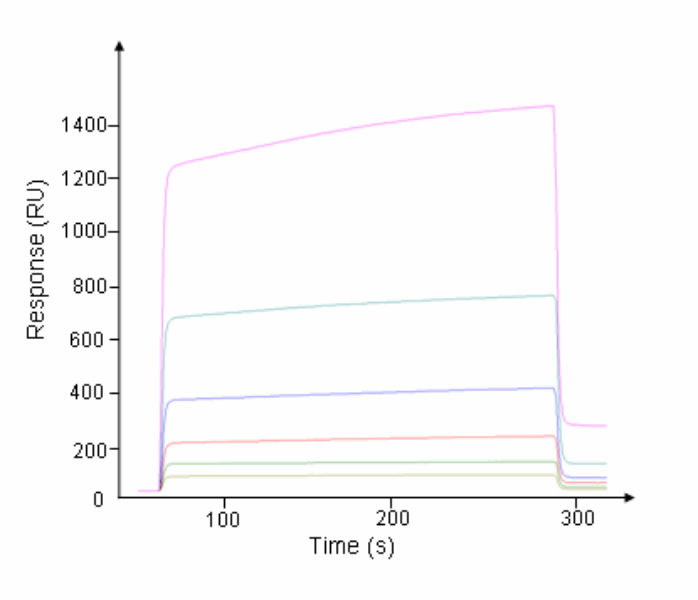
extend of hPARP-1 auto-poly(ADP-ribosyl)ation was determined by Western blot analysis and the levels of poly(ADP-ribose) polymer (PAR) were quantified densitometrically (Epson 4870 photo, scion image program) and then normalized to the hPARP-1 protein content. Data are expressed in relative units of PAR levels of *in vitro* automodified PARP-1 standard (Trevigen), which was arbitrary set as 1.0.

CD and UV characterization of the pS2 and iNOS quadruplexes. Putative quadruplex forming sequences have been identified in the region of the promoters of the pS2 and iNOS genes. To demonstrate the capacity for these sequences to form G-quadruplexes *in vitro*, we have used CD and UV spectroscopy. The DNA sequences of both putative quadruplexes as well as the CD and UV spectra of a 5 µM DNA solution are reported in Supplementary Figure 3 below.



Supplementary Figure 3 *Biophysical analyses of iNOS and pS2 promoter quadruplexes*. (**top**) DNA sequences of iNOS and pS2 DNA quadruplexes. (**bottom left**) Melting UV profiles of iNOS and pS2 quadruplexes at 10 µM strand concentration in 50 mM TRIS-HCl, pH 7.4 containing 100 mM KCl. (**bottom right**) CD spectra of iNOS and pS2 quadruplexes at 10 µM strand concentration in 50 mM TRIS-HCl, pH 7.4 containing 100 mM KCl.

hPARP-1 binding to the intramolecular human telomeric quadruplex. In order to demonstrate that hPARP binding to the c-kit-1 quadruplex was not sequence specific but structure specific, we used SPR to measure the affinity of purified hPARP-1 for the intramolecular human telomeric quadruplex. The sensorgram overlay is reported in Supplementary Figure 4 below and show that hPARP-1 also binds Htelo although with a weaker affinity than c-kit-1.



Supplementary Figure 4 *hPARP-1 binds to the human telomeric quadruplex.* Sensorgram overlay for hPARP-1 binding to immobilized human telomeric quadruplex of sequence d(biotin-[$GT_2A(G_3T_2A)_4G_2$]) at 6 different hPARP-1 concentrations (0.98 to 31.25 nM bottom to top)