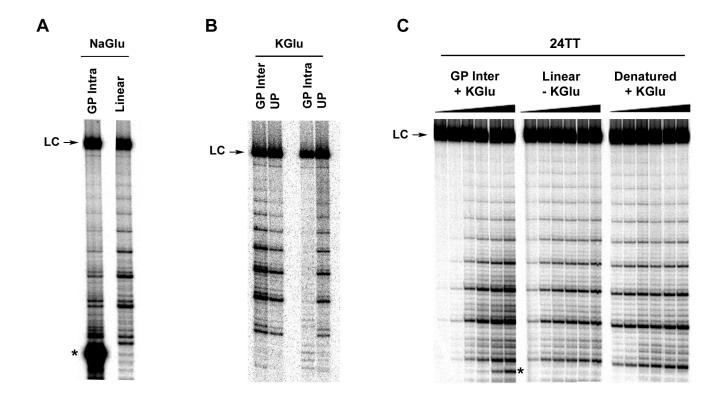
## **Supplementary Figure 3**



Telomerase activity assays using *in vitro* reconstituted recombinant *Tetrahymena* telomerase and G-quadruplexes formed from oligonucleotide 24TT stabilised in 50 mM KGlu. For all panels, LC = loading control (<sup>32</sup>P-labelled 100mer) and \* represents the unextended <sup>32</sup>P-labelled gel-purified G-quadruplex. All assays were conducted at 25°C.

- (A)  $^{32}$ P-labelled gel-purified intramolecular 24TT ("GP Intra") at a final concentration of 7.7  $\mu$ M (93% purity) stabilised in the presence of NaGlu was used a substrate in a 60 min telomerase activity assay. Unlabelled unpurified 24TT primer of the same concentration folded in the presence of 50 mM NaGlu was used as a control ("UP").
- (B) Telomerase activity assays using unlabelled inter-  $(7.7 \,\mu\text{M})$  and intramolecular  $(1.3 \,\mu\text{M})$  24TT G-quadruplex stabilised in the presence of 50 mM KGlu (lanes 1 and 3 respectively). Unpurified 24TT primer was folded in the presence of 50 mM KGlu at either 7.7  $\mu$ M or 1.3  $\mu$ M and used as a control substrate (lanes 2 and 4 respectively) in a 60 min telomerase activity assay.
- (C) Telomerase extension of increasing concentrations (0.04-18  $\mu$ M) of <sup>32</sup>P-labelled intermolecular 24TT G-quadruplex (86% purity) stabilised in 50 mM KGlu. The control is unlabelled linear 24TT over the same concentration range in the absence of KGlu. As an additional control for the effect of KGlu, linear 24TT in the presence of 50 mM KGlu was denatured at 95°C for 5 min and immediately placed on ice prior to extension. The extension was carried out for 10 min.