Two independent regions of human telomerase reverse transcriptase (hTERT) are important for its oligomerization and telomerase activity

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Human telomerase reverse transcriptase (hTERT), the catalytic subunit of telomerase, contains motifs conserved among reverse transcriptases. Several nucleic acid-dependent polymerases (NdNPs) that shared a "fingers, palm and thumb substructure" were shown to oligomerize. Here, we demonstrate that hTERT also has this ability using partially purified recombinant hTERTs and mammalian cells coexpressing different tagged hTERTs. Human-TR, by contrast, has no effect on the structural oligomerization of hTERTs. Therefore, hTERT by itself has an intrinsic ability of oligomerization in the absence of hTR. We identified two separate regions as essential for the oligomerization. The regions, aa 301-538 (amino-terminal region) and aa 914-928 (carboxy-terminal region) outside the fingers and palm substructure covering motif A to D, interact with each other *in vivo*. A dominant negative form of hTERT bound to the wild-type hTERT and inhibited its telomerase activity transiently expressed in telomerase-negative finite normal human fibroblasts. The catalytically inactive truncated forms of hTERT, which contained the binding region to the wild-type hTERT, inhibited the telomerase activity partially, probably by preventing the wild-type hTERT from forming an oligomer. These results imply that the hTERT oligomers having active catalytic centers are critical for telomerase activity.
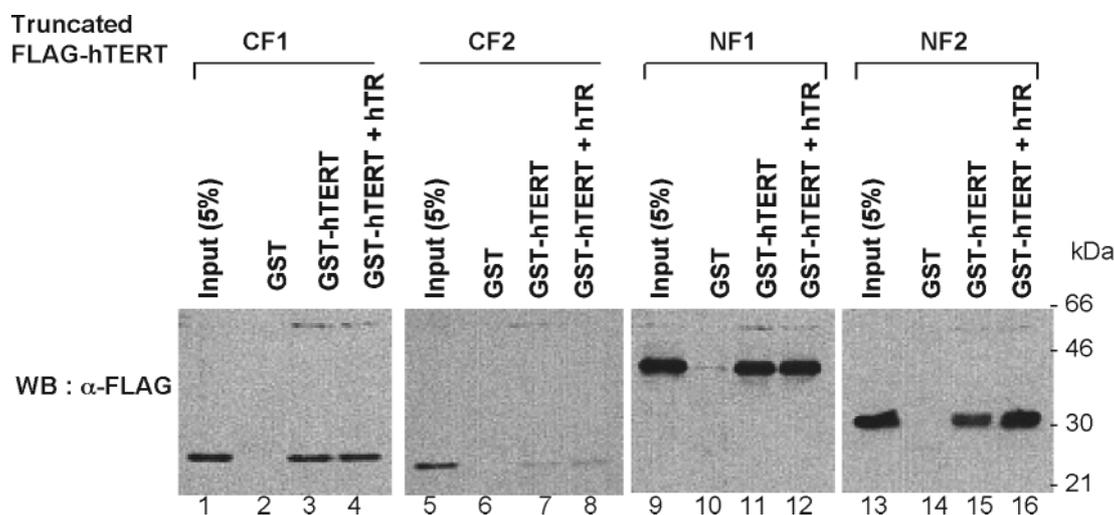
Figure Two independent regions of hTERT exhibited homomeric interaction.
Partially purified truncated forms of FLAG-hTERT were inbucated with GST, GST-hTERT, or GST-hTERT and hTR, then subjected to GST pull-down. Recovered FLAG-hTERT was visualized by western blotting with anti-FLAG antibody. CF1, CF2, NF1 and NF2 harbor aa 914-1132, aa 928-1132, aa 201-538, and aa 301-538, respectively.