

## **Telomere localization and dynamics in normal and in cancer cells**

**Hans J. Tanke<sup>1</sup>, Anneke Brouwer<sup>1</sup>, Tsion Abraham<sup>1</sup>, Vered Raz<sup>1</sup>, Nico Verwoerd<sup>1</sup>, Roland Eils<sup>2</sup>, B.J. Vermolen<sup>3</sup> and Roeland W. Dirks<sup>1</sup>**

**<sup>1</sup>Department Molecular Cell Biology, Leiden University Medical Centre, Leiden;  
<sup>2</sup>Intelligent Bioinformatics Systems, DKFZ, Heidelberg; <sup>3</sup>Delft University of Technology, Department of Imaging Science & Technology, Delft.**

**Address: Leiden University Medical Center  
Department of Molecular Cell Biology  
PO Box 9600-Postal zone S1-P  
2300 RC Leiden  
The Netherlands  
Email: h.j.tanke@lumc.nl**

Telomeres are large nucleoprotein complexes at the ends of eukaryotic chromosomes that consist of highly repetitive hexanucleotide sequences and associated proteins. These complexes play an important role in maintaining chromosome stability and integrity, in regulating the replicative life span of somatic cells and in chromosome pairing processes during the first meiotic prophase. Furthermore, telomeres are suggested to contribute to maintenance of chromosome topology in the cell nucleus and thereby to play an important role in establishing a functional nuclear organization. We wished to test the hypothesis that telomeres are static complexes in the cell nucleus because of their association with the nuclear matrix. To this end we used a fluorescently labeled peptide nucleic acid (PNA) probe with a sequence complementary to telomeric DNA. Upon introduction into cells by a glass bead loading technique these probes were shown to specifically bind to telomeres. Alternatively, fusion proteins of the telomere binding proteins TRF1, TRF2 and POT with YFP were expressed in living cells. The dynamic behavior of telomeres was analyzed by time-lapse imaging and quantified using computer software. In a telomerase negative osteosarcoma cell line (U2OS) that maintains telomeres by the ALT mechanism, telomeres were shown to interact dynamically with PML bodies. The nature of this interaction is investigated further by analyzing protein-protein interactions using a fluorescence resonance energy transfer approach.

Using newly developed algorithms developed at Delft University of Technology (BJV) 3D spatial reorganization of centromeres and telomeres was studied in mesenchymal stem cells, in which apoptosis was induced by caspase-8.