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Review Article

Telomeres rather than telomerase a key target for anti-cancer therapy?

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Telomere shortening is a prerequisite of all replicating cells

Telomeres, the outermost ends of the chromosomes, are essential for chromosomal stability and integrity. They consist of many thousand repeats of the hexanucleotide TTAGGG giving rise to 4–15 kb of repetitive non-coding DNA in humans and up to 50 kb in mice. Due to replication deficiencies [endreplication problem (1,2)] and telomere end processing (3) the telomeres shorten progressively with replication in normal somatic cells in vitro and eventually trigger senescence, i.e. irreversible growth arrest. This telomere length-dependent growth inhibition which prevents critically short telomeres and thereby potentially unprotected chromosomes is thought to be a barrier for unlimited cellular proliferation (4, reviewed in 5).

Unrestricted proliferation requires telomere maintenance mechanisms

Due to replication-dependent telomere erosion, unrestricted proliferation, characteristic of germ line cells, immortal and tumor cells, requires a mechanism that is able to counteract telomere shortening and indeed, telomere shortening is overcome by activating telomere maintenance mechanisms. Whereas the majority (>80%) of cells (germ line and tumor cells) have an active telomerase, 10–20% of the tumor cells, especially sarcomas and astrocytomas (6,7), utilize alternative lengthening (ALT) of telomeres (8), and only a few have no known telomere maintenance mechanism (7).

Telomerase is an RNA-dependent DNA polymerase containing several components. The core enzyme consists of an RNA component, termed hTR in humans, containing the template that is utilized by the catalytic subunit, the telomerase reverse transcriptase hTERT for telomere repeat addition (9). Furthermore, a number of proteins are essential for a functional telomerase complex (for review, see 10). Active telomerase adds de novo telomeric sequences to the outermost ends of the telomeres and thus compensates for replication- or damage-dependent loss of telomere sequences, or can even elongate the telomeres.

ALT-positive cells, on the other hand, are characterized by very long telomeres (>40 kb) as well as an extremely large variation in telomere length within the same nucleus. Another hallmark of the ALT mechanism is the presence of ALT-associated promyelocytic leukaemia protein (PML) nuclear bodies, the APBs, subnuclear structures containing PML protein, telomeric DNA, telomere-binding proteins, and several proteins involved in DNA synthesis and recombination (11). Although the molecular mechanism of ALT is still largely unknown, it is believed that individual telomeres undergo steady telomere attrition upon which sudden lengthening and shortening events are superimposed (12). These recombination-dependent changes are not due to a

global increase in recombination in ALT vs. telomerase-positive cells but to increased rates of telomeric recombination (reviewed in 13).

Telomerase is thought to be a universal tumor marker

Telomerase is expressed during embryonic development but repressed in most adult tissues. Only regenerative tissues continue to show some level of activity (reviewed in 14). Most tumor cells, on the other hand, are thought to have high activity implicating that telomerase is an excellent tumor marker (reviewed in 15). The most frequently used method to evaluate telomerase activity is the Telomere Repeat Amplification Protocol (TRAP assay), originally described by Kim et al. (16) and extensively used thereafter to demonstrate a positive relationship between telomerase activity and tumor growth (for reviews see 17–19). However, as a drawback, TRAP activity can only be determined from fresh tissue, requires pieces of tumor material and thus may suffer from a rate of false-positive results when contaminating telomerase-positive non-tumor cells are included or false negative when inhibitory factors (e.g. heparin) are present (summarized in 20). Consequently, in situ hybridization for hTR and hTERT have been performed and correlated with telomerase activity (summarized in 21). Although this allows a more detailed analysis throughout the tumor and thus discloses intratumoral heterogeneity (22,23), it is time- and labour-consuming and as a drawback has to remain inconclusive about the functionality of the telomerase complex.

With the same limitations, namely uncertainty about functional activity, antibody staining was used. Although the hTERT antibodies presently available are still a matter of debate, Lantuejoul et al. (21) reported on a high concordance between hTERT protein expression and detection of hTERT mRNA and telomerase activity. Even more so, they could attribute distinct hTERT staining patterns to different histopathological classes of lung cancer. This said, Volpi et al. (20) showed positive staining not only in the tumor tissue but also in all normal tissues analysed and most intriguingly, not only nuclear but also cytoplasmic staining. In tumors, the cytoplasmic TERT staining was mostly attributed to disruption of the normal hTERT nuclear translocation process during malignant transformation while post-transcriptional and/or post-translational modes of regulation were proposed for the normal cells (24,25). Hines et al. (26) found hTERT expression in both normal and malignant breast tissue and unexpectedly, the level of expression in tumor cells did not appear any greater than the maximum fluorescence contained within the normal samples, suggesting that the normal cells contained a subset with high hTERT expression.

The epidermis differs from other commonly analysed tissues

The epidermis is one of the few regenerative tissues that constitutively expresses telomerase activity (27,28). While telomerase activity was attributed to the presence of stem cells – requiring telomere maintenance mechanisms to maintain their lifelong replication capacity – we provided evidence that telomerase is not a stem cell marker but is most prominent in the actively proliferating successor cells, the so-called transit amplifying cells, and inhibited with differentiation in a step-wise fashion (29) (Rosenberger et al., in revision). Thus, telomerase is tightly regulated in the epidermis in situ. Furthermore, telomerase activity from freshly isolated epidermal keratinocytes can be as high as in immortal skin keratinocytes (Moshir et al., 2006, in revision) arguing that in skin carcinomas telomerase upregulation may not be required as an extra step during the multi-stage carcinogenesis process. Instead, it is tempting to speculate that the constitutive expression of telomerase and with that lack of 'aberration-dependent' de novo expression may be one reason for the high frequency of skin carcinomas.

Accordingly, nuclear hTERT protein expression was seen in normal epidermis, though the intensity was increased in epithelia formed by immortal HaCaT skin keratinocytes and further augmented in epithelia of hTERT over-expressing HaCaT cells (30). A comparative study of keratoacanthomas, benign and spontaneously regressing skin tumor, and skin squamous cells carcinomas, on the other hand, showed very similar staining patterns (31). Most notably, in most tumors expression was not uniform but restricted to focal areas (Fig. 1). Undoubtedly, this needs verification by other means. However, with all caution it could suggest that hTERT and with that also telomerase is not upregulated in all tumor cells and therefore not evenly expressed throughout the tumor. The reason could be twofold. So far the time of telomerase upregulation during tumor progression is still elusive and only certain populations of the tumor cells may have yet gained the ability to express substantial levels of telomerase. A second and for epidermal tumors perhaps most likely explanation would be that telomerase remains sensitive to regulation. In normal keratinocytes telomerase is upregulated with proliferation and inhibited with differentiation and this may similarly be maintained in tumors – as also suggested from the fact that positive staining is most prominent in tumor areas with stromal contact, i.e. areas of high proliferation (see Fig. 1).

How does this relate to studies measuring telomerase activity in human skin tumors? Different from our previous and present findings (28) (Moshir et al., 2006, in revision), other studies suggest that telomerase activation in normal

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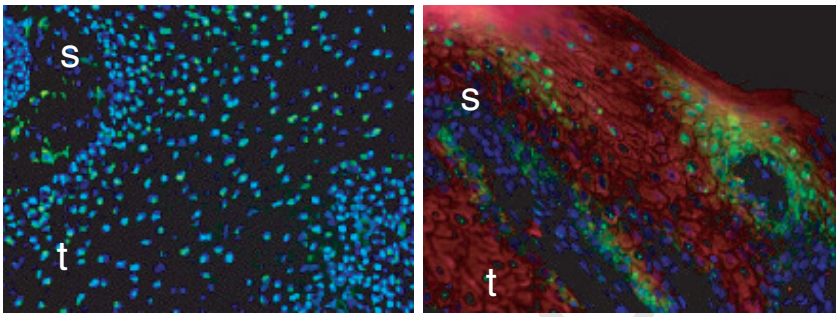

Fig. 1 Immunofluorescence staining of skin SCCs with an antibody against hTERT showing a tumor with nearly a 100% labelled nuclei (left) and a tumor with only focal expression of hTERT. Keratin is stained in red, hTERT in green, and nuclei are counterstained with Hoechst in blue (s = stroma, t = tumor).

skin is a rare event (32,33). In addition, in benign lesions – including viral and seborrhoeic warts – telomerase activity was rarely detected while 42% of the actinic keratoses and Bowens's diseases showed TRAP activity. In the same study a good number of BCCs and MMs, but only few SCCs were telomerase-positive (32). On the other hand, Boldrini et al. (34) reported on high telomerase activity in SCCs but only low levels in BCC. Using telomerase enzyme-linked immunosorbent assays, a third study in turn found high levels of telomerase activity in BCCs (35). Even more so and comparing tumors and their tumor margins, shorter relapse-free periods correlated with telomerase-positive margins. As the tumors were not further specified – and if not due to technical reasons – these discrepancies in telomerase expression in both SCCs and BCCs may indeed reflect their overall spectrum of telomerase expression.

Telomerase is also expressed by other regenerative epithelia

Comparing telomerase activity in different types of epithelia, we previously showed that the oesophagus, stomach and colon express telomerase. However, a level of expression similar to that observed in the epidermis was only seen in the multilayered tissue of the oesophagus. In stomach and colon, TRAP activity was detectable, though clearly reduced (14). Furthermore, Barrett's oesophagus, a transition from a multilayered to a simple-type epithelium, correlated with a similarly reduced level of telomerase activity. A significant increase in telomerase activity only occurred late in tumor progression as also confirmed recently by others (36). Whether this is a result of de novo expression of telomerase in the tumor cells or selection for already actively expressing telomerase-positive precursor cells still remains unsolved.

Little is known about telomerase regulation in tumors in situ

If skin carcinomas are not an exception, one has to assume that at least in certain tumors telomerase activity remains prone to regulation and, therefore, as described above, telomerase may not be expressed constitutively throughout the tumor. However, little is known about its regulation in tumors in situ. With the exception of the haematopoietic system (reviewed in 37), most studies dealing with solid tumors are performed with telomerase-positive tumor cell lines, thereby demonstrating various mechanisms and levels of regulation including differentiation-dependent inhibition of telomerase for several cell types and regulation by growth factors in culture (for review, see 38).

Accordingly, only few studies addressed the possibility of telomerase regulation through the environment. As shown for normal and immortal keratinocytes, telomerase is regulated in a tissue-dependent manner. This was demonstrated by changes in the hTERT splicing pattern (39) and can now also be extended to histone acetylation and with that hTERT promoter activation in vivo vs. in vitro (Moshir et al., 2006, in revision). Besides these global changes caused by growing cells in the three-dimensional situation of a multicellular tumor vs. monolayer culture, minor micro-milieu-based variations also have to be considered. Along that line, hypoxia was shown to regulate telomerase (discussed in 40) and may thus also be responsible for local telomerase modulations in the tumor.

Finally, evidence is increasing that tumors contain stem cell populations (for review, see 41). It is anticipated that these tumor stem cells have characteristics similar to that observed in normal somatic ones and may thus only proliferate rarely. If, as suggested for epidermal stem cells (29), they express less telomerase activity than the more rapidly proliferating successor transit amplifying cells, which are likely to represent the majority of the tumor mass, the tumor stem cells may well be protected against many treatment regimes. These would include conventional chemotherapy targeting the rapidly proliferating cells as well as the newly anticipated anti-telomerase tumor therapy targeting cells with high telomerase expression.

Anti-telomerase therapy is the future challenge

With the finding that telomerase is absent in most normal tissues but highly upregulated in cancer cells, telomerase

quickly became a target for the development of novel anti-cancer strategies (extensively reviewed in 42). Furthermore, it was proposed that 'the unique biology and function of telomerase, together with the complexity of its regulation, means that therapeutic targeting is possible at various points of the cell signalling and transcriptional machinery' (43). As a first approach, telomerase activity was inhibited in cancer cells by dominant-negative mutants of the hTERT gene, antisense oligonucleotides, or ribozymes directed against hTR or hTERT (Fig. 2a). These studies provided proof of principle that inhibition of telomerase was causing telomere shortening and consequently a delayed onset of apoptosis or senescence. One such compound GRN163L is a 13-mer N3-P5′ oligonucleotide thio-phosphoramidate complementary to the template region of hTR and thus a potent telomerase antagonist and is now ready for phase I/II clinical trials (44) (see Fig. 2a). Depending on telomere length and growth rate of the tumor the onset of apoptosis or senescence may take long, perhaps even too long to be effective for the patient. Therefore, only tumors with very short telomeres appear as appropriate candidates for this approach.

Telomerase immunotherapy is another promising approach. In three different studies vaccination is presently performed with hTERT peptides in a variety of advanced tumor patients in order to induce functional cytotoxic T lymphocytes (CTL). These CTL recognize hTERT-specific peptides that are expressed on the cell surface of tumor but not normal cells and were shown to kill hTERT-positive tumor cells in vitro (45) (Fig. 2b). Furthermore, tumor necrosis was observed and dose escalations resulted in enhanced immunological response. In a Norwegian study two different hTERT peptides were co-injected with granulocyte macrophage colony-stimulating factor (GM-CSF) intradermally into about 100 advanced pancreatic, lung and melanoma patients during the past 3 years. So far no serious adverse effects have been observed with respect to bone marrow stem cells or autoimmune disease, even not in long-term survivors (>2 years) (presented in 44). The latter is an interesting finding because it was recently shown for a skin cancer model that GM-CSF can act as a paracrine growth factor for the tumor cells and increase their invasive capacity (46). In particular, the long-term follow-up study reveilles hope. Thus, it is now the efficacy of the treatment that needs to be proved.

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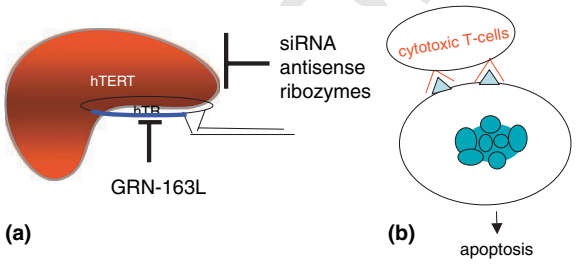

Fig. 2 Telomerase as an anti-cancer target: (a) The two major components of the telomerase complex, the RNA component (hTR) and the catalytic subunit (hTERT), are inhibited by compounds such as specific siRNAs, anti-sense oligonucleotides or by GRN-163L complementary to the template region of hTR. (b) Cytotoxic T-cells recognizing hTERT on tumor cells and inducing apoptosis.

Other recent approaches are based on gene therapy strategies (summarized in 43). Most frequently, the hTR or hTERT promoter is placed in front of a cell killing agent (versatile suicide gene therapy). Furthermore, oncolytic viruses are used where genes, that are crucial for replication, are placed under the control of the hTERT gene promoter. These viral vectors only replicate in tumor cells and even more so spread to adjacent tumor cells upon cell lysis. Interestingly, small interfering RNAs (siRNA) targeting hTR rapidly inhibited growth of human cancer cells not by causing the expected effect on telomere shortening but by inducing changes in the global gene expression profile (47). As genes were involved that are responsible for cell-cycle progression, tumor growth, angiogenesis and metastasis, the authors suggested that 'cancer cells are telomerase-addicted and uncover functions of telomerase in tumor growth and progression in addition to telomere maintenance'. Thus, telomerase upregulation in tumor cells is indicative of novel response pathways and anti-telomerase tumor therapy is supposed – as mentioned above – to interfere with many steps of tumor growth. This said, many predictions derived from studies with cancer cell lines. If skin carcinomas are not the exception and if telomerase regulation is maintained to a certain degree also in tumors, successful targeting of all tumor cells as well as oncolytic restriction to the tumors may not be achievable.

The most radical hypothesis was proposed by de Grey et al. (48): 'The deletion (not merely inhibition) of a gene whose function is essential for cancers to progress would present a major challenge to cancer cells'. As the gene of question they suggested telomerase in order to obtain 'whole body interdiction of lengthening of telomeres', shortly termed WILT. The argument for inhibiting telomere elongation in order to avoid cancer cell growth stems from the early studies with telomerase knock out mice providing evidence that short telomeres suppressed tumor progression (reviewed in 49). Thus depleting telomere elongation mechanisms body-wide is thought to be the ultimate cure. Of course, this requires a number of considerations. The most serious one is that certain tissues, such as the haematopoietic system, the gastrointestinal tract or the epidermis of the skin, proliferate continuously. Their cells need to be replaced recurrently in order to avoid telomere-length-dependent deficiencies. For this, the authors proposed an ex vivo telomere elongation in the respective

stem cells and reconstitution of the cells in vivo. Whether this will ever be practicable needs to be seen.

One important step in the preclinical evaluation of these approaches are animal studies. As telomerase is thought to be less tightly regulated in mouse than in man (50,51), telomerase gene therapy may, however, be difficult to prove conclusively in mouse models. With the novel data on telomerase being present in more and more normal human tissues, on the other hand, these arguments may be put into perspective. Nevertheless, prominent differences remain between mouse and man. One example is that the human hTERT gene is characterized by several splice variants while so far splice variants were not described for the mouse mTERT gene (52). This said, it could be shown that only the full-length variant gives rise to telomerase activity while the shorter variants are inactive or even act in a dominant negative fashion (53,54). As the splicing pattern is prone to regulation in vivo (39), splicing is also now extensively analysed in tumors and may even provide potential for interference. The fact that splice variants are so far only detected in humans may indicate that splicing has evolved only late in evolution and is required for the special needs to tightly control telomerase activity in humans.

The proposed mechanism of an anti-telomerase therapy is that telomerase inhibition leads to telomere shortening and that this, depending on the cell type, results in apoptosis or senescence. However, telomere reduction requires replication. As discussed above, if the hypothesis of tumor stem cells is correct, and a 'stem cell hierarchy' exists, with stem cells only replicating rarely, telomeres in these tumor stem cells would only shorten very little in spite of telomerase inhibition, implicating that these cells can survive for a long time. Thus, understanding the molecular mechanism of stem cell quiescence and through that being able to force the cells into proliferation will be essential to effectively attack them. Furthermore, the old idea by Cairns, inducing tumor cell differentiation, may be as effective as an anti-tumor therapy because, in addition to downregulating telomerase activity, it causes the cells to irreversibly exit cell cycle and finally die.

Telomere inhibition provides an alternative therapeutic intervention

9 Another approach involves folding of the 3′ overhang into a four-stranded G-quadruplex structure (55). The double-stranded (ds) telomeric TTAGGG repeats end in a 3′ single-stranded overhang. While the ds DNA can fold back into a loop structure, the T-loop (56), and is stabilized by a number of proteins, the single-stranded overhang is incorporated into the loop (57) (Fig. 3a,b). Uncapping of the telomere ends, on the other hand, leads to free exposure of the overhang and with that to senescence or apoptosis (58,59). Due to the stacking of multiple planar G·G·G·G tetrads, G-rich structures can form G-quadruplex structures (60) and concerning the telomeric 3′ overhang, G-quadruplex structures have been implicated in elongation of telomeres by telomerase (61). While the non-folded single-stranded telomere is required for optimal telomerase action, G-quadruplex structures hinder telomere elongation by telomerase. Accordingly, small molecules selectively stabilizing the telomeric G-quadruplex structures cause telomere shortening and replicative senescence (summarized in 62). One such agent is telomestatin, a G-quadruplex-interacting agent derived from Streptomyces nanalatus that was described as a potent telomerase inhibitor (63). In certain leucemic cells, telomestatin led to telomere shortening and consequently to telomere dysfunction (64). Recently, Tahara et al. (65) reported that short-term treatment of telo-

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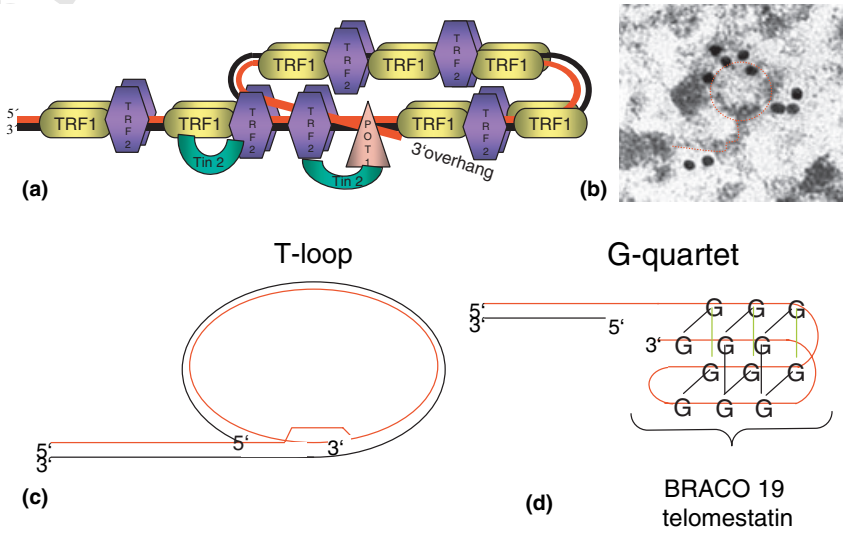

18 **Fig. 3** Telomere models. (a) T-loop structure occupied by the major protein components, the telomere repeat binding factors TRF1 and TRF2, TIN2, and the single strand (ss)-binding protein POT1. (b) Immuno-electron micrograph of a telomere loaded with TRF2-tagged gold particles, resembling a T-loop structure. (c) Schematic view of the T-loop. (d) Schematic view of the G-quadruplex structure of the ss overhang stabilized by BRACO 19 Or telomestatin.

mestatin caused cancer cells to die but not normal cells. As a mechanism, they proposed that the telomere repeat binding factor 2 (TRF2), one of the two telomeric proteins that directly bind to telomeric DNA and are essential for stabilization of the telomeric loop and thus for the capping function of the telomere, is rapidly dissociated from the telomeres. It was shown earlier that inhibition of TRF2 by overexpressing a dominant-negative mutant resulted in a high frequency of end-to-end fusions and activation of the ATM (Ataxia-Telangiectasia-mutated)/p53 DNA damage response pathway (66). As telomestatin induced the same massive telomere dysfunction, uncapping of the telomeres is obviously its primary effect. Accordingly, BRACO-19, a 11 trisubstituted acridine, has been shown to induce long-term growth arrest and replicative senescence in beast carcinoma cells and some in vivo activity against a tumor xenograft (67). Using a prostate cancer cell line, the authors showed that, similar as with telomestatin, BRACO 19 caused uncapping of the telomeres resulting a high degree of end-to-end fusions.

Interesting in this context is also the fact that overexpression of a dominant-negative mutant TRF2 caused apoptosis in tumor cells while normal fibroblasts went into senescence (66). Accordingly, telomestatin treatment also caused a rapid growth inhibition and early cell death in cancer cells while non-cancer cells remained viable for much longer (65). While it is difficult to imagine how telomeres should differ between normal and tumor cells it may suggest that mechanisms causing the slow growth of normal vs. the rapid proliferation of tumor cells may be involved. However, we also cannot exclude at present that the protein composition at the telomeres may differ (quantitatively or qualitatively) and thus provide normal cells with a higher degree of telomere stability. Whatever the reason will be, this differential response is intriguing and may open new avenues of interference.

Can telomere stabilization be the ultimate cure?

Unfortunately, long-term follow-up is not yet possible. Thus, it remains to be seen whether such a treatment causes all cells to suffer from end-to-end fusion and to die or whether some cells will be able to escape. Why could this be possible? Critically short and therefore dysfunctional telomeres are prone to end-to-end fusion which can result in chromosomes with two or more active centromeres. When these attach to the opposite poles during mitosis they give rise to anaphase bridges followed by breakage during further chromosome separation. This process also known as fusion-bridge-breakage cycle is now used extensively to explain the origin of translocation chromosomes as well as gain and loss of chromosomal material (for review, see 68). Thus, depending on the damage of the cells, dysfunctional telomeres likely contribute to chromosomal changes.

Accordingly, we recently showed that telomeres can form aggregates and that these aggregates are induced when the *c-myc* oncogene is activated (69–71). We further showed that aggregate formation in these cells was correlated with chromosomal rearrangements (71) underlining the role of abnormal telomeres for genomic instability and thus genomic variability. Although the mechanism of aggregate formation is still unclear, preliminary evidence suggests that TRF2 is involved, further strengthening the role for TRF2 as one of the key factors responsible for genomic stability (70) (Ermler and Boukamp, in preparation). On the other hand, we yet have to investigate whether uncapped 3′ overhangs are involved. One telomere-binding protein that seems particularly prone to this process is POT1 (protection of telomeres 1), a protein binding to the single stranded 3′ overhang (72). It was recently shown that POT1 is required for normal processive elongation of telomeres by telomerase. hPOT1 does not act catalytically but forms a stoichiometric complex with the DNA, freeing its 3′ tail (73). As this suggests that hPOT1 functions by trapping the unfolded forms in order to allow proper elongation of the telomeres by telomerase, the 3′ single-stranded overhangs in cooperation with POT1 may be promising candidates in the search of the molecular players in aggregate formation. Thus, similar to length-dependent or treatment-dependent telomere uncapping, telomere aggregation may also allow for genomic instability. This can result in new genetic aberrations which in turn may provide the cells with a selective growth advantage and thus help to escape from further treatment.

More sophisticated preclinical human models are required to test anti-telomerase treatment regimes

The models for testing anti-telomerase agents are either monolayer cultures of human tumor cell lines or human xenograft tumors. In both cases the cell systems only reflect part of the tumor entirety, namely those that adapt most easily to tissue culture conditions. These cells are propagated and characterized in vitro and after reinjection into immune-deficient mice often form rapidly growing tumors. How does this relate to tumor growth in the patient? Besides the discrepancies described above concerning the different regulation of telomerase in vitro and in vivo (39), the composition – rather homogeneous population in vitro vs. tumor heterogeneity in vivo – may vary significantly. Most importantly, different stages of tumor progression, including those that may not have yet an upregulated telomerase, are present in solid tumors next to the invasively growing tumor cells. This heterogeneity is not recapitulated in tumors derived after subcutaneous injection of cell lines,

and the question also remains whether tumor stem cells establish in such tumors. Another aspect concerns the species specificity of oncolytic viruses. The human adenovirus serotype 5 that is the basis of e.g. the tropism-modified telomerase-specific replication-selective adenoviral agent 'Telomelysin-RGD' (74,75) only infects human cells and correspondingly side effects in tissues other than the human tumor can not be investigated. Thus, in addition to designing new approaches for treating cancer, improved and more sophisticated (natural) models are needed in order to better test the efficacy of novel therapeutic approaches.

Recently a number of complex three-dimensional culture systems have attracted much attention including organotypic cultures to analyse stratified epithelia and their corresponding tumors (76,77) or spheroid cultures for a variety of tumor cells (77,78). In addition, organotypic hippocampal slice cultures are used to investigate mechanisms and treatment strategies of neurodegenerative disorders (reviewed in 79). Similarly, mouse tumor models need to be included that address the problems of tumor heterogeneity and slow replication of the tumor stem cells in order to obtain a more realistic proof of the success of the respective treatment regime.

Taken together, much effort is put into designing specific telomerase- and telomere-dependent anti-cancer treatment regimes and it is thought to be a highly promising approach. However, reports demonstrate and argue for a function of telomerase in normal tissues. If our early results can be generalized, that telomerase is not necessarily active in the stem cells but required in the more rapidly proliferating transit amplifying cells (29) then telomerase inhibition, particularly when administered over a long time, is likely to affect normal cells as well. Approaches causing tumor-specific and rapid cell death should, therefore, have precedence. Only this way, adaptation and resistance (e.g. a switch from telomerase to the ALT mechanism of telomere elongation) can largely be excluded. Furthermore, instead of ignoring the effects on normal tissues, it might be favourable to also design strategies to better protect them and/or interfering with mechanisms causing early upregulation of telomerase in tumor cells. Targeting telomeres, the actual target structure, has opened up another avenue for potential anti-tumor therapies. Intriguing in this context is the treatment with telomestatin. If future experiments will prove that only telomeres from tumor cells but not normal cells are affected and thus only tumor cells die, telomere-dependent strategies may provide one of the most promising anti-cancer treatment strategies.

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