

# Telomeres in Disease

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**Telomeres have been linked to numerous diseases over the years, but how exactly short telomeres cause diseases and how medicine can prevent telomere erosion are still up for debate.**

The ends of linear chromosomes have attracted serious scientific study—and Nobel Prizes—since the early 20th century. Called telomeres, these ends serve to protect the coding DNA of the genome. When a cell's telomeres shorten to critical lengths, the cell senesces. Thus, telomeres dictate a cell's life span—unless something goes wrong. Work over the past several decades has revealed an active, though limited, mechanism for the normal enzymatic repair of telomere loss in certain proliferative cells.[1. E.H. Blackburn et al., “Telomeres and telomerase: the path from maize, Tetrahymena and yeast to human cancer and aging,” *Nat Med*, 12:1133-38, 2006.] Telomere lengthening in cancer cells, however, confers an abnormal proliferative ability.

In addition to cancer, telomeres have been found to be involved in numerous other diseases, including liver dysfunction and aplastic anemia, a condition in which the bone marrow does not produce a sufficient supply of new blood cells.[2. R.T. Calado et al., “Telomere diseases,” *N Engl J Med*, 361:2353-65, 2009.] Inadequate telomere repair and accelerated telomere attrition can be molecular causes of these diseases, and targeting these processes may lead to the development of novel therapies.

## Chromosome tails

Telomeres consist of hexameric nucleotide sequences (TTAGGG in humans) that are repeated hundreds to thousands of times at each extremity of each chromosome. Telomeric DNA is coated by a group of proteins, collectively termed shelterin, which serves to protect telomere structure. Because DNA can only be synthesized in one direction, the RNA primers at the chromosome's ends cannot be filled in, and thus a small amount of DNA is lost with every cell division—a loss that occurs in the telomeres. During normal aging of an animal or in cell culture, cells divide and telomeres shorten. As telomeric sequences do not contain genes, no important genetic information undergoes erosion during DNA replication. When telomeres become critically short, the cell becomes senescent—it ceases to divide—or undergoes apoptosis—it dies.

Telomere attrition explains the “Hayflick limit,” the number of divisions a cell is capable of undergoing in tissue culture before the cell stops dividing. Telomere length is therefore a type of “mitotic clock,” a measure of a cell's proliferative history. Under circumstances in which cell proliferation continues despite critically short telomeres (usually about a few hundred hexanucleotide repeats), the telomere's protective function is lost. Subtelomeric genetic information can be lost, and more importantly, recombination between chromosomes occurs, leading to chromosome instability, aneuploidy, and possible transformation to a cancer phenotype.

Some proliferative cells can elongate telomeres enzymatically through the telomerase complex. Telomerase (TERT) is a reverse transcriptase that employs a small RNA molecule (TERC) as a template to extend telomeres in cells. In this way, telomerase counterbalances the effects of cell division and cellular genetic “aging,” preventing senescence, apoptosis, and genetic instability. Telomerase-dependent telomere repair occurs naturally in some cells, such as embryonic and adult stem cells and some cells of the immune system—cell types that divide regularly to support development, maintain tissues, and combat infections, respectively.

Telomere maintenance is also possible by other mechanisms, such as the alternative pathway (ALT), which uses recombination between chromosomes to maintain telomere length. In ALT, telomeres are not newly elongated, but rather transferred from one chromosome to another, resulting in some daughter cells whose chromosomes have shorter telomeres and others with longer telomeres. The details of ALT's components and regulation, however, are not well understood.

## Telomere Timeline

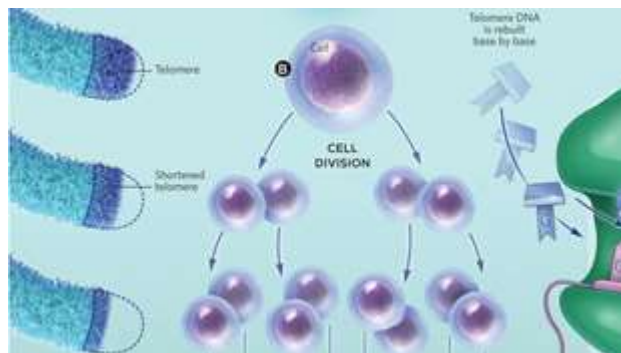
In the 1930s, Hermann Muller and Barbara McClintock observed that fragmented chromosomes tended to fuse with each other, while normal chromosomes were stable, and they predicated characteristics of the “natural ends” to explain this difference. Muller named these ends “telomeres.”

Almost 40 years later, Alexey Olovnikov, on theoretical grounds, and James Watson, based on phage experiments, recognized a fundamental problem regarding the mechanics of DNA replication, during which a small amount of genetic information is lost with every cell division. In the late 1970s, Elizabeth Blackburn and Joseph Gall discovered the structure of telomeres—short, highly repetitive noncoding nucleotide sequences—in the ciliated protozoan *Tetrahymena*. In 1982, Blackburn and Jack Szostak elucidated how telomeres in yeast could serve as a buffer for the coding DNA during replication, and postulated the existence of an enzyme that could rebuild telomeres—an enzyme discovered by Blackburn and Carol Greider in *Tetrahymena* 3 years later. By 1988, the sequence of the human telomere was known, and researchers began actively investigating its role in aging and cancer. Subsequent work showed that the telomeres in certain proliferative cells are actively repaired. For their work on telomeres, Blackburn, Greider, and Szostak were awarded the 2009 Nobel Prize in Physiology or Medicine.

## Telomere shortening and cancer

Most cells in which telomeres reach critically short lengths either die or enter senescence. In those few that survive, perhaps due to inadequate monitoring by p53 and related DNA damage-response safeguards, telomere repair would be subject to powerful selective pressure. Indeed, in most malignancies, telomerase gene upregulation or activation of the ALT pathway is thought to be necessary for the establishment of cellular immortality. Telomerase is so frequently increased in tumors and in cancer cell lines as to be considered an appropriate therapeutic target. Currently there are several clinical trials using telomerase inhibitors to treat a variety of cancers, but results have yet to be reported. Telomere shortening would also generate the equivalent of a “mutator phenotype” by increasing spontaneous chromosomal aberrations—from numerical changes to structural abnormalities—and would therefore increase the pool of aberrant cells upon which selection would act.

There are many sources of evidence suggesting that telomere attrition is associated with and likely etiologic of cancer. Patients with dyskeratosis congenita, a rare inherited bone marrow failure disease characterized by telomerase dysfunction,[3. B.P. Alter et al., “Very short telomere length by flow fluorescence in situ hybridization identifies patients with dyskeratosis congenita,” *Blood*, 110:1439-47, 2007.] have a 1,000-fold increase in risk of tongue cancer and about a 100-fold increase in risk of acute myeloid leukemia.[4. B.P. Alter, et al, “Cancer in dyskeratosis congenital,” *Blood*, 113:6549-57, 2009.] In aplastic anemia, patients with the shortest telomeres (absent mutations) are 4- to 5-fold more likely to have their disease undergo malignant transformation to myelodysplasia and leukemia.[5. P. Scheinberg et al., “Association of telomere length of peripheral blood leukocytes with hematopoietic relapse, malignant transformation, and survival in severe aplastic anemia,” *JAMA*, 304:1358-64, 2010.] Telomere-free ends of chromosomes and aneuploidy may be apparent in cultured bone marrow years before progression to leukemia. Furthermore, some acute myeloid leukemia patients without prior bone marrow failure have inherited mutations in *TERT* and *TERC*.[6. R.T. Calado et al., “Constitutional hypomorphic telomerase mutations in patients with acute myeloid leukemia,” *PNAS*, 106:1187-92, 2009.]



Infographic: Telomere Basics, SCOTT LEIGHTON, CMI

More generally, genome-wide analyses have identified single nucleotide polymorphisms in *TERT* as risk factors in many cancers. In a recent report, short leukocyte telomeres were associated with increased risk of all cancers and of cancer fatalities in a large population followed over a decade. Circumstantially, telomere attrition is an accompaniment of aging, itself a major risk factor for cancer. Furthermore, secondary malignancies often occur after chemotherapy and radiation, which would be anticipated to cause marrow stress and telomere shortening. More direct data come from animal experiments. In a knockout mouse model, animals with reduced telomerase activity combined with diminished p53 surveillance of DNA damage developed a variety of epidermal cancers unusual in the rodent but typical of humans.

## Other telomere diseases

In addition to cancer, other diseases have been linked to telomeres. Hematopoietic stem cells express telomerase in response to the enormous daily demand for red blood cells, white blood cells, and platelets. Thus while overexpression of telomerase in other tissues can cause malignant growth, faulty telomere repair in blood stem cells can also result in severe diseases caused by stem cell failure.

One such “telomere disease” is dyskeratosis congenita, an X-linked bone marrow disorder characterized by symptoms such as abnormal nails, a pigmented, net-like rash, a white patch or plaque in the mouth, and aplastic anemia. The disease usually presents in the first decade of life. The liver and lungs can also be affected, as is often observed after a hematopoietic stem-cell transplant is performed to correct the bone marrow disease. The reasons for liver and lung involvement are not clear, but the chemotherapy used for transplant conditioning and the new inflammatory cells in the transplanted bone marrow may accelerate the injury in these organs.

## Telomeres and aging

Telomeres shorten as we age. By analogy to the cellular mitotic clock, telomeres have been postulated as a marker of “genetic age,” and telomere length has been marketed as a simple predictor of longevity. Assays of telomere length have been bundled with recommendations for lifestyle modification and for drug therapy, neither based on appropriate clinical studies. Simple but appealing arguments relating telomeres and aging are currently controversial, likely simplistic, and potentially harmful. Telomere length does indeed reflect a cell’s past proliferative history and future propensity for apoptosis, senescence, and transformation. Cellular aging, however, is not equivalent to organ or organismal aging.

There are several considerations in relating telomere biology to aging. First, physiologically there is overlap between the shortest telomere length of young children and the longest telomeres of the elderly. Most telomere shortening occurs early in life, in association with growth, and when the rate of disease in general is low. The paradigmatic telomere syndrome of dyskeratosis congenita is not at all typical of progerias, inherited syndromes in which patients appear old and suffer diseases of aging such as premature atherosclerosis or dementia. Furthermore, the organ damage of dyskeratosis congenita is not very similar to normal aging of marrow, lungs, and liver. The marrow becomes mildly hypocellular in older individuals, but stem cell numbers may actually increase, and blood counts remain stable; and neither the liver nor lungs normally become fibrotic with advanced age, as they often do in dyskeratosis congenita patients. Although in adults, relatively short leukocyte telomeres have been associated with cardiovascular events—a common morbidity of the aging population—the clinical correlations have not been consistent, and may be related to overall reactive oxygen species exposure.

Studies in humans have attempted to relate telomere length to life span. In the provocative initial publication from the University of Utah in 2003, individuals around 60 years of age who had the longest telomeres lived longer than did subjects with the shortest telomeres, but the main cause of death in the latter group was, inexplicably, infectious disease; the persons with shorter telomeres did not have a higher rate of cancer deaths.[8. R.M. Cawthon et al., “Association between telomere length in blood and mortality in people aged 60 years or older,” *Lancet*, 361:393-95, 2003.] Moreover, these findings have not been confirmed in other studies of older subjects. In another study evaluating a different population, telomere length failed to predict survival, but interestingly it correlated with years of healthy life.[9. O.T. Njajou et al., “Association between telomere

length, specific causes of death, and years of healthy life in health, aging, and body composition, a population-based cohort study,” *J Gerontol A Biol Sci Med Sci*, 64:860-64, 2009.] In a Danish study of people aged 73 to 101 years, telomeres correlated with life expectancy in a simple univariate analysis, but only before the researchers corrected for age, suggesting that the correlation was driven simply by the fact that younger subjects had longer telomeres. And a Dutch study of 78-year-old men found that while telomere lengths eroded with age, they failed to correlate with mortality.[10. J.M. Houben et al., “Telomere length and mortality in elderly men: the Zutphen Elderly Study,” *J Gerontol A Biol Sci Med Sci*, 66:38-44, 2011.] These discrepancies may have several causes. In some analyses, telomere lengths may have been studied as a surrogate marker of age. In addition, retrospective studies may identify “positive” associations that are random and cannot be reproduced in follow-up investigations.

## Implications for medicine

Telomeres and their repair are important in the growing field of regenerative medicine. Dolly the sheep had chromosomes with shorter telomeres probably because she was cloned from an adult mammary gland cell. This may have contributed to Dolly’s illnesses, especially her progressive lung disease. Embryonic stem cells, however, express telomerase and are able to maintain their telomere lengths despite numerous cell divisions. More recently, reprogramming mature adult skin cells to the pluripotent state has been achieved with introduction of just a few defined nuclear factors. During the process of reverting cells to a more immature and pluripotent state, the reprogrammed cells’ telomeres are highly elongated. In the first steps of reprogramming and likely in the early stages of embryogenesis, cells can elongate, and thus “rejuvenate,” their telomeres. Since telomere shortening limits cell proliferation, mechanisms that can elongate telomeres are highly desirable for effective regenerative medicine.

## Conclusions

Telomeres and telomere repair are basic molecular processes in cells possessing linear DNA chromosomes. Accelerated telomere attrition due to genetic defects in telomerase and in the shelterin protein genes is etiologic in several human diseases not previously considered related in the clinic. These include aplastic anemia, pulmonary fibrosis, and hepatic cirrhosis. The telomeropathies, especially in their milder and more chronic forms, may not be rare and almost certainly are often unrecognized by physicians. The importance of telomere repair in tissues under regenerative stress is of special interest, particularly in the reactive responses of fibrogenesis in the liver and the lungs. The maintenance of adequate telomere lengths also may be important in embryonic and adult stem cells to enable proliferation while preventing chromosome instability, thus avoiding potential malignant transformation. Also of interest is the connection linking telomere attrition and chronic inflammation to cancer and other diseases. (See “[An Aspirin for Your Cancer](#),” *The Scientist*, April 2011.)

There is still much to be learned about how telomerase gene mutations cause disease, why they only affect certain organs, and how telomeres can be targeted for therapies. Both the genetic regulation of telomerase expression and the effect of an organism’s environment on telomere attrition are poorly understood. Drugs or hormones that might modulate telomerase expression and maintain or elongate telomeres would be appealing in the treatment of the telomeropathies and in conditions in which telomere attrition has known medical consequences. Whether telomere shortening mediates human aging—and conversely, whether telomere elongation may reverse aging or prevent age-related diseases—are still controversial issues.

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