Telomere-Subtelomere-Telomerase System

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Synonyms
Telomere-telomerase system; TST system

Definition
The expression telomere-subtelomere-telomerase system, or shortly TST system, describes the morphologic and functional complex consisting of telomere, subtelomere, and telomerase (which are composed of various distinct molecular parts) and the interactions between them, their parts, and the other cellular components.

Overview
For aging, defined as age-related decreasing fitness (or increasing mortality), there are two opposite explanations. They are mutually incompatible, radically different and with so many diverse assumptions and implications that may deserve the definition of distinct “paradigms” (Libertini 2015a) in the meaning proposed by Kuhn (1962).

The first paradigm, defined as the “non-adaptive or non-programmed aging paradigm,” justifies aging as the cumulative effect of many degenerative factors that natural selection cannot sufficiently oppose (mutation accumulation, antagonistic pleiotropy, disposable soma, and damage accumulation hypotheses [Libertini 2015a, b]).

The second paradigm, defined as the “adaptive or programmed aging paradigm,” explains aging as a physiological phenomenon evolutionarily favoured in terms of supra-individual natural selection (Libertini 2015a). For this paradigm, aging is a form of phenoptosis (Skulachev 1997; Libertini 2012), i.e., “programmed death of an organism” (Skulachev 1999, p. 1418), which is a very common type of phenomena in the biological world, well known as physiological events favored and modeled by natural selection (Finch 1990).

An important and opposite implication of the two paradigms is that: (1) according to the non-adaptive aging paradigm, senescence cannot be caused by specific genetically determined and regulated mechanisms, and (2) on the contrary, according to the adaptive aging paradigm, such mechanisms must exist, and indeed their existence is indispensable in order to consider this paradigm valid (Libertini 2015a).

Although the nonadaptive aging paradigm still represents the group of theories presented as the
correct explanation of aging (Olshansky et al. 2002; Kirkwood and Melov 2011; Kowald and Kirkwood 2016; Fedarko 2018), on the basis of the results of numerous and qualified works, it is possible to describe the mechanisms that appear to determine aging and that are certainly genetically determined and modulated and not the simplistic effect of random accumulation of harmful events (Fossel 2004; Libertini 2015a, b). These mechanisms, which will be briefly described here, have as their core the telomere-subtelomere-telomerase (TST) system. It determines (i) the gradual cell senescence, (ii) the cell senescence, (iii) the slowing down of the cellular turnover, and (iv) the atrophic syndrome of all tissues and organs, phenomena that constitute the substrate of aging (Fossel 2004; Libertini 2009, 2015a, b).

The Machinery of the TST System

After a long period in which it was erroneously believed that cells could duplicate themselves an unlimited number of times, limits in cellular duplication capabilities were shown in 1961 (Hayflick and Moorhead 1961). However, for some time, it was not understood why the cells had these limits. In 1971, the observation that the DNA polymerase enzyme, which allows DNA duplication, fails to duplicate a small part of the terminal region of the molecule (the telomere), suggested the hypothesis that the progressive shortening of the telomere at each duplication could explain the limits in cell duplication (Olovnikov 1971). Subsequently, as germ and stem cells appeared capable of unlimited or very many duplications, respectively, the existence of an enzyme, then called telomerase, capable of restoring the part of the telomere not duplicated was predicted (Olovnikov 1973). This enzyme was isolated and described a few years later (Greider and Blackburn 1985).

It was also demonstrated that the telomere is a repetitive nucleotide sequence, TTAGGG in humans, and other mammals (Moyzis et al. 1988), which was highly conserved during evolution and present in many species that were phylogenetically distant (Blackburn 1991).

Subsequently, valuable information was provided by the study of the yeast (Saccharomyces cerevisiae), a single-celled eukaryotic species. In S. cerevisiae, the telomerase enzyme is always active, and the length of the telomeres remains the same after each duplication (D’Mello and Jazwinski 1991). The yeast reproduces by division of a mother cell that gives rise to a “daughter” cell and another “mother” cell. The cells of daughter lineage may duplicate themselves an unlimited number of times, while those of the mother lineage may reproduce only a limited number of times (25–35 duplications in about 3 days) and show a progressive decline in the ability to sustain stress (Jazwinski 1993). The main difference between the cells of the two lines, apart from a functional decline in the cells of the mother line, was that the mother cells showed, in proportion to the number of duplications, a progressive accumulation of particular substances, extrachromosomal ribosomal DNA circles (ERCs), on the portion of DNA adjacent to the telomere (the subtelomere) (Sinclair and Guarente 1997).

It was then observed that in the strains of particular yeast mutants, the tcl1Δ mutants, which exhibited deficient telomerase activity, aside from the fact that both mother and daughter cells showed telomere shortening at each generation, individuals of the daughter line while not showing ERC accumulation as individuals of the wild-type strain, showed a decline in the ability to resist stress and a transcriptome similar to that of older individuals of the mother lineage of the wild strain (Lesur and Campbell 2004).

These facts suggested that the alterations shown by the cells of the mother lineage in the wild strain and also by the daughter cells in tcl1Δ mutant strain were always due to the repression of the subtelomeric region, although determined by two different mechanisms: (i) accumulation of ERCs in the mother cells of wild strains and (ii) shortening of the telomere in the daughter cells of the mutant strain. This also suggested that the decline of cell functions and the replicative senescence observed in multicellular eukaryotic organisms could have a mechanism similar to that explaining the alterations in the daughter cells of tcl1Δ mutants (Libertini 2009).
In fact, it had already been proposed that the telomere was covered by a heterochromatin “hood” and that the progressive shortening of the telomere caused a proportional sliding of the hood with the repression of the adjacent subtelomere: “One model of telomere-gene expression linkage is an altered chromosomal structure (Ferguson et al. 1991), such as a heterochromatin ‘hood’ that covers the telomere and a variable length of the subtelomeric chromosome (Fossel 1996; Villeponteau 1997; Wright et al. 1999). As the telomere shortens, the hood slides further down the chromosome (the heterochromatin hood remains invariant in size and simply moves with the shortening terminus) ... the result is an alteration of transcription from portions of the chromosome immediately adjacent to the telomeric complex, usually causing transcriptional silencing, although the control is doubtless more complex than merely telomere effect through propinquity (Aparicio and Gottschling 1994; Singer et al. 1998; Stevenson and Gottschling 1999). These silenced genes may in turn modulate other, more distant genes (or sets of genes). There is some direct evidence for such modulation in the subtelomere ...” (Fossel 2004, p. 50).

As hinted by Fossel, this model necessarily required a hood with a fixed length, defined in the germ cell of an organism, and that this length remained unchanged in all subsequent duplications (Libertini and Ferrara 2016a). These concepts are illustrated in Fig. 1.

Subtelomere repression has general consequences on cell functions, including extracellular secretions. This phenomenon was first described in yeast in 1990 and was called telomere “position effect” (Gottschling et al. 1990, p. 751). Subsequently, to describe the manifestations of this phenomenon on the cell, the name “gradual cell senescence” was proposed (Libertini 2015b). These concepts are illustrated in Fig. 1.

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As the subtelomere is increasingly repressed by the sliding of the hood caused by telomere shortening, another effect of the gradual cell senescence is a growing probability of activation of a particular program, “cell senescence,” a “fundamental cellular program” (Ben-Porath and Weinberg 2005, p. 962), which has two main effects: (i) blocking of replication capabilities and (ii) manifestations of the gradual cell senescence at the highest level.

A simplistic model could be that cell senescence program is activated when telomere shortening reaches a critical value. However, in synchronized cell cultures, that is, with an equal number of duplications in all cells, the duplication capacity did not stop simultaneously in all the cells but showed a progressive decrease in the whole culture related to the number of duplications, that is to say related to telomere length reduction. The explanation for this phenomenon was proposed by Blackburn (2000).

The telomere is covered by a cap (probably the same hood involved in the explanation of gradual cell senescence [Libertini 2015b]). This cap does not permanently cover the telomere, and therefore the telomere continuously oscillates between two states, “capped” and “uncapped.” The duration of the second state is related to telomere shortening, and in this state, the cell is vulnerable to the passage to replicative senescence, i.e., to the activation of cell senescence program. Even when telomeres have their maximum length and telomerase is active, a small percentage of cells pass to the replicative senescence (Blackburn 2000).

Since the fraction of time in which the telomere is uncapped and vulnerable is proportional to telomere shortening and therefore to the level of subtelomere repression, it has been hypothesized that the percentage of time in which the telomere is uncapped is somehow regulated by subtelomeric repetitive sequences, defined as “r,” which would also have general regulatory action over the functions altered in gradual cell senescence (Libertini and Ferrara 2016a).

The possible existence of “r” sequences is supported by the description of subtelomere structure as an “unusual structure: patchworks of blocks that are duplicated” (Mefford and Trask 2002, p. 91) and “long arrays of tandemly repeated satellite sequences” (Torres et al. 2011, p. 85).

These concepts are illustrated in Fig. 2.
Absence of a Relationship Between Initial Telomere Length and Longevity

It is well known that in the comparison between individuals of the same species, there is an inverse correlation between age of the individual and cell duplication capacities (Martin et al. 1970; Libertini 2009). The reduction of these capacities was anticipated by Olovnikov (1971, 1973) as related to telomere shortening. From this, a

**Telomere-Subtelomere-Telomerase System, Fig. 1** (A) Yeast, normal stock, daughter lineage; (B) yeast, normal stock, old individuals of mother lineage; (C) yeast, *tcl1A* mutants, daughter lineage; (D) multicellular eukaryotes, cells of the germinal line; (E) multicellular eukaryotes, somatic cells. In A and D, telomeres are not shortened, and subtelomeric DNA is not repressed. In B, subtelomeric DNA is repressed by ERC accumulation. In C and E, telomeres are shortened, and the subtelomeric DNA is repressed by the proteinic hood. (Figure from Libertini 2015a, modified and redrawn)
possible relation, in the comparison among species, between telomere length in germ line cells and longevity has been hypothesized (Gorbunova et al. 2008). However, this expected relation is denied by strong contrary evidence (Gorbunova et al. 2008; Libertini and Ferrara 2016a), and this could induce to doubt the hypothesis that aging alterations are determined by telomere shortening.

Similarly, it could be hypothesized that in the comparison among species, there is a correlation between longevity and a greater activity of telomerase, but also this possible relation is contradicted by evidence (Gorbunova et al. 2008).

The topic was discussed elsewhere, highlighting that the aforesaid two correlations falsified by the evidence do not have a theoretical foundation (Libertini and Ferrara 2016a).

In the germ cell, in a phase definable as “reset” phase, the hood is modeled on the length of the telomere, and its size does not change in all subsequent duplications. As far as longevity is concerned, the absolute “telomere length is irrelevant” (Fossel 2004, p. 36), provided that the telomere length is not less than a critical value (Fossel 2004). In subsequent duplications, the telomere is shortened, and the hood slides on the subtelomeric region. The critical element is the proportion of subtelomere that is inhibited as a consequence of the relative shortening of the telomere. Therefore, if, other things being equal, in case 1 we have a long telomere but a short subtelomere, while in case 2, we have a short telomere but a long subtelomere, it is easy to predict that after an equal number of duplications, we will have a degree of subtelomere repression that is greater in case 1. A similar concept is shown in Fig. 3, where the case of two mutant strains of mice is considered, with telomeres long the first 20 kb and the second 10 kb (or also the case of a donor animal and a cloned animal), which showed equal longevity and aging rates (Fossel 2004). In this case, we have telomeres of different lengths but equal subtelomeres: an identical sliding of the hood causes in the cells of the two strains an identical repression of the subtelomere and therefore identical manifestations related to aging.

Similarly, a lower/higher telomerase activity could be balanced by a lower/greater length of the subtelomere (Libertini and Ferrara 2016a).

The Effect of the TST System on the Whole Organism

The effects of the TST system, which vary according to how it is regulated in each species, are extremely important for the whole body.

In mammals, even in the absence of diseases or traumatic lesions, most of the cells are subjected to continuous turnover. In fact, cells die of various types of programmed cell death (PCD), e.g.
Keratinization of epidermis and hair cells.

- Detachment of cells of mucous membrane from the lining of intestines or other body cavities.
- Osteocytes phagocytized by osteoclasts.
- Transformation of erythroblasts in erythrocytes with subsequent removal by macrophages.
- Apoptosis, an ordinate process of self-destruction for the first time described in the study of normal epatocytes (Kerr et al. 1972), well documented for many tissues and organs (e.g., biliary epithelial cells, gliocytes, kidneys, pancreatic β-cells, liver, thyroid, type II alveolar epithelial cells, cartilage, prostate, adipocytes, bone, skeletal muscle [Libertini 2009]).

The cells eliminated by PCD are replaced by duplication of appropriate stem cells. Cell turnover rates vary greatly depending on cell types (Richardson et al. 2014). While some cell types are renewed in a few days (e.g., in the intestinal epithelium, “cells are replaced every 3–6 days” [Albers et al. 2013, p. 705]), for others, the renewal takes place over a period of years (e.g., “bone has a turnover time of about 10 years in humans” [Albers et al. 2013, p. 705], and “the heart is replaced approximately every 4.5 years” [Anversa et al. 2006, p. 1457]).

Cell turnover is slackened and finally stopped by cell duplication limits determined by the TST system. The decline in cell turnover causes a
reduction in the number of functional cells and their replacement with nonspecific cells. This determines a decline in the function of tissues and organs increased by the dysfunction of cells in gradual cell senescence and in cell senescence (which involves the gradual cell senescence at the highest level). These cells also show alterations of extracellular secretion that cause dysfunctions in other cells that otherwise would be perfectly functional (Libertini and Ferrara 2016a).

It should be noted that this functional decline also affects perennial cells, i.e., not subject to cell turnover, which therefore would not seem to have to suffer from the aforementioned phenomena. However, as these cells depend for their functionality and vitality from other cells that are subject to turnover, the phenomena of gradual cell senescence, cell senescence, and decline in cell turnover affecting their satellite cells cause dysfunction and death in perennial cells (Libertini and Ferrara 2016b). These two types of decline, the first caused by direct impairment of the functional cells of a tissue and the second caused by impairment of the perennial cells as a result of that of their satellite trophic cells (which anyway may have other important functions), may be distinguished with the terms “direct” and “indirect” aging.

This decline in the functionality of tissues and organs determines a general fitness decline, that is to say a lower ability to overcome difficulties caused by external factors and, in the most advanced stages, the sure death of the organism (Libertini 2009).

In order to describe this set of phenomena, to which it is necessary to add the greater vulnerability to cancer caused by telomere dysfunction (DePinho 2000; Meena et al. 2015; Bernal and Tusell 2018), the definition “atrophic syndrome” has been proposed (Libertini 2009, p. 95). A general scheme of these alterations is shown in Fig. 4 and Table 1.

Telomere-Subtelomere-Telomerase System, Fig. 4  A scheme of aging phenomenon
It should be noted that cell turnover is not a feature of all animals (e.g., the adult stage of the worm *Caenorhabditis elegans* does not show cell turnover and has a fixed number of cells) (Finch 1990).

### The TST System and the Limits in Cell Turnover as a General Defense Against Cancer

TST system, cell turnover, and its progressive decline appear as a very sophisticated machinery, certainly determined and regulated by genes, which reduce the fitness of the organism and, ultimately, favors the death of the individual. These phenomena appear incompatible with the thesis of nonadaptive aging, while on the contrary, they are perfectly compatible with the opposite paradigm of adaptive aging, for which they are indispensable to allow its validity (Libertini 2015a, b).

As these phenomena invalidate the paradigm of nonadaptive aging, the proponents of this thesis have somehow tried to justify their existence by attributing to cell senescence and to the limits imposed on cell turnover the aim, favored by natural selection, of a general defense against cancer (Campisi 2000).
This explanation is contradicted by many facts, which, for the sake of brevity, cannot be explained in detail here. However, we will mention some of them, e.g., (i) existence of animals without any age-related increase in mortality (animals with “negligible senescence” [Finch 1990, p. 206]) and with no increase in cancer mortality in the older ages (as it is implicitly demonstrated by the invariability of mortality rates at any age); (ii) gradual cell senescence, i.e., a mechanism that reduces cell functionality and consequently the fitness of the organism and does not make sense as a defense against cancer; (iii) telomere shortening that determines DNA instability and so increases the chances of cancer (DePinho 2000); and (iv) in mice, the selective elimination of senescent cells (p16Ink4a+ cells), i.e., with functions altered by cell senescence, contrasts various age-dependent manifestations, delays the progression of neoplastic diseases, and increased lifespan, and this is against the possibility that cell senescence might be a defense against cancer (Libertini and Ferrara 2016a).

Consequently, the hypothesis that the TST and the consequent phenomena might be a general defense against cancer appears completely untenable because falsified by evidence (Libertini 2009; Mitteldorf 2013; Libertini and Ferrara 2016a).

However, this thesis remains common in the scientific community and supported by authoritative researchers (Campisi and Robert 2014), perhaps because its rejection would also imply the rejection of the nonadaptive aging paradigm and the transition to the adaptive aging paradigm: “The hypothesis that telomerase is restricted to achieve a net increase in lifespan via cancer prevention is certainly false. Were it not for the unthinkability of the alternative – programmed death – the theory would be dead in the water” (Mitteldorf 2013, p. 1058). In fact, a paradigm shift is always a difficult and of slow gestation event (Kuhn 1962).

**Conclusion**

Gradual cell senescence, cell senescence, and limits in cell duplication capacity appear to have their main mechanisms and regulations in the TST system. As these phenomena are the likely determinants of aging manifestations, the study of TST system is essential to understand aging mechanisms. About the widespread idea that aging is due to the accumulation of oxidized substances or even of altered mitochondria, in a sort of modern repetition of old theories that explain aging on the basis of “wear and tear” phenomena, it is important to highlight an authoritative opinion: “Cells do not senesce because of wear and tear, but because they permit wear and tear to occur because of an altered pattern of gene expression” (Fossel 2004, p. 53) and that this altered gene expression is under the control of telomere, telomerase, and subtelomere (Fossel 2004), i.e., the STS system.

**Cross-References**

- Cell Senescence
- Effects of Telomerase Activation
- Gradual Cell Senescence
- Subtelomere
- Telomerase
- Telomeres
- Timeline of Aging Research

**References**


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