Published in final edited form as:
Mech Ageing Dev. 2008 ; 129(7-8): 467–474. doi:10.1016/j.mad.2008.04.001.

Cellular senescence and organismal aging

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Abstract

Cellular senescence, first observed and defined using *in vitro* cell culture studies, is an irreversible cell cycle arrest which can be triggered by a variety of factors. Emerging evidence suggests that cellular senescence acts as an *in vivo* tumor suppression mechanism by limiting aberrant proliferation. It has also been postulated that cellular senescence can occur independently of cancer and contribute to the physiological processes of normal organismal aging. Recent data have demonstrated the *in vivo* accumulation of senescent cells with advancing age. Some characteristics of senescent cells, such as the ability to modify their extracellular environment, could play a role in aging and age related pathology. In this review, we examine current evidence that links cellular senescence and organismal aging.

Keywords

Cellular senescence; Aging; Telomeres

1. Introduction

Cellular senescence was first described by Hayflick and Moorfield in 1961 who observed that cultures of normal human fibroblasts had a limited replicative potential and eventually became irreversibly arrested (Hayflick and Moorhead, 1961; Campisi and d'Adda di Fagagna, 2007; Sedivy et al., 2007). The majority of senescent cells assume a characteristic flattened and enlarged morphology, and over the years a large number of molecular phenotypes have been described, such as changes in gene expression, protein processing and chromatin organization (Gonos et al., 1998; Shelton et al., 1999; Schwarze et al., 2002; Semov et al., 2002; Narita et al., 2003; Zhang et al., 2003; Yoon et al., 2004; Pascal et al., 2005; Xie et al., 2005; Zhang et al., 2005; Cong et al., 2006; Funayama et al., 2006; Trougakos et al., 2006; Zdanov et al., 2006; Zhang et al., 2007). The growth arrest occurs mostly in G1 phase (Pignolo et al., 1998). Although individual cells arrest rapidly, probably within the duration of a single cell cycle, cultures are typically quite asynchronous with increasing proportions of cells withdrawing into senescence over a period of several weeks (Herbig et al., 2003; Herbig et al., 2004). Senescent cells maintain metabolic activity and can remain viable essentially indefinitely (Matsumura et al., 1979; Pignolo et al., 1994). An important component of this stability in culture may be the capacity of senescent cells to resist apoptosis (Marcotte et al., 2004; Hampel et al., 2005).

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Conceptually, there are two broad categories of replicative cellular senescence. The first is initiated by dysfunctional telomeres or other forms of genotoxic stress eliciting a DNA damage response mediated primarily by the p53 tumor suppressor pathway (d'Adda di Fagagna et al., 2003; Herbig et al., 2004). The second, much less understood response does not involve telomeres or DNA damage, and is characterized by the upregulation of the *CDKN2A* gene (cyclin dependent kinase inhibitor $p16^{INK4a}$). These basic distinctions are however complicated by the fact that p16 can be upregulated by a wide variety of stresses, including some forms of genotoxic damage.

The relationship between elapsed cell divisions and the onset of senescence was clearly apparent from early *in vitro* studies, and led to proposals that senescence may contribute to *in vivo* organismal aging phenotypes (Hayflick, 1985). This view was reinforced by findings that cells explanted from old donors were capable of fewer *in vitro* population doublings than those from young individuals (Martin et al., 1970; Le Guilly et al., 1973; Rheinwald and Green, 1975; Schneider and Mitsui, 1976; Bierman, 1978; Bruce et al., 1986). The gradual attrition of telomeres subsequently provided the molecular mechanism for the cell division clock (Harley et al., 1990; Bodnar et al., 1998). It has been proposed that the term “cellular senescence” be reserved for those phenomena based on an inherent counting mechanism, with other terms, such as “stasis” to be applied to the large variety of stress-induced arrests (Drayton and Peters, 2002; Wright and Shay, 2002). Recent usage has however favored irreversible cell cycle arrest as the defining feature, and “cellular senescence” is now commonly used to encompass states induced by stress and signaling imbalances (Collado et al., 2007).

It is important to note that the intrinsic cell division clock can be significantly affected by extrinsic influences, such as reactive oxygen species, which accelerate the rate of telomere shortening (von Zglinicki, 2002). Although senescent cells display a number of phenotypes that discriminate them from quiescent cells, it has also been suggested that senescence could be considered a form of terminal differentiation (Bayreuther et al., 1988; Seshadri and Campisi, 1990). This view has recently been given new life by observations that downregulation of Wnt signaling may be a factor in triggering the onset of senescence (Ye et al., 2007). Thus, in addition to being influenced by the environment, senescence may also respond to developmental or endocrine cues.

Given that senescence results in the arrest of proliferation, its potential for opposing cancer development was pointed out some time ago (Sager, 1991). This notion was strongly reinforced by the discovery that activation of oncogenes in normal cells could trigger senescence (Serrano et al., 1997). Recent data have indeed implicated cellular senescence as an important *in vivo* tumour suppressor mechanism in a variety of human and mouse tissues (Braig et al., 2005; Chen et al., 2005; Collado et al., 2005; Michaloglou et al., 2005; Courtois-Cox et al., 2006; Cosme-Blanco et al., 2007; Feldser and Greider, 2007; Ventura et al., 2007; Xue et al., 2007). In addition to being on sound experimental footing, the tumor suppressive function of senescence also provides a rational explanation for its evolution. The possible role of senescence in age associated dysfunction is often justified by invoking the concept of antagonistic pleiotropy (Williams, 1957; Rose, 1991; Kirkwood and Austad, 2000; Campisi, 2005), namely, that beneficial traits (such as cancer suppression) under selection in reproductively active individuals may have unselected and unintended effects in more advanced age.

In contrast to tumor suppression, the connections between cellular senescence and the aging of organisms are significantly more tenuous. In this review we focus on these links and their implications. Elucidating these relationships is expected to advance our comprehension of the mechanisms involved in age-related diseases as well as normal aging processes.

2. Telomeres

Telomeres shorten with each round of genome duplication, an unavoidable consequence of the RNA priming mechanism of DNA replication (Olovnikov, 1973). The minimum rate of telomere shortening in human cells (30–50 bp per cell division) is slightly in excess of that predicted by the “end-replication problem”, and probably stems from further exonucleolytic processing of chromosome ends (Sfeir et al., 2005). Telomere attrition can be accelerated by a number of factors, such as oxidative damage (von Zglinicki et al., 1995; von Zglinicki, 2002). Dysfunctional telomeres are sufficient to trigger a full senescence response, and in many cases constitute the primary cause of the observed growth arrest (Bodnar et al., 1998; Wright and Shay, 2002). Even a single dysfunctional telomere may be sufficient to establish senescence in the affected cell (Hemann et al., 2001; Herbig et al., 2006).

Sensitive and accurate methods for measuring single telomere lengths in individual cells, as well as average telomere lengths in populations of cells have been developed, and have produced a wealth of data indicating an inverse correlation between telomere length and age in a number of tissues (Cawthon et al., 2003; Baerlocher et al., 2006; Canela et al., 2007; Kimura et al., 2007). A significant number of important age associated pathologies, such as cardiovascular disease, diabetes and Alzheimer's disease have been correlated with telomere shortening, as have obesity, psychological stress and socio-economic status (Panossian et al., 2003; Epel et al., 2004; Ogami et al., 2004; Valdes et al., 2005; von Zglinicki and Martin-Ruiz, 2005; Cherkas et al., 2006; Adams et al., 2007; Minamino and Komuro, 2008). This literature has grown quite large and engendered some lively debates (Ellison, 2006; Hornsby, 2006; Kuh, 2006; Lansdorp, 2006). The epidemiology of telomere length associations is compromised by wide inter-individual differences in telomere length at birth, as well as variations in telomere attrition rates after birth (Aviv et al., 2006). Although the importance of telomere attrition is well supported by cross-sectional data associating shorter telomeres with age associated pathologies, longitudinal studies will be needed to evaluate the links between telomere attrition and rates of aging (De Meyer et al., 2008).

Dysfunctional telomeres trigger a DNA damage response similar to that elicited by the presence of double strand breaks (DSB), which can be visualized in individual cells by the presence of nuclear foci containing the phosphorylated form of the histone variant H2AX (d'Adda di Fagagna et al., 2004; von Zglinicki and Martin-Ruiz, 2005; Herbig and Sedivy, 2006). Such foci also contain many proteins involved in the recognition, signaling and repair of DNA damage, and when physically localized at telomeres are referred to as telomere dysfunction induced foci (TIF) (d'Adda di Fagagna et al., 2003; Takai et al., 2003; Herbig et al., 2004). The occurrence of DNA damage foci increases with replicative age of cultures as well as organismal aging in both mice and primates. Not all DNA damage foci are telomeric, and the species- and tissue-specific distribution of TIF and non-telomeric foci are only beginning to be investigated (Sedelnikova et al., 2004; Garcia-Cao et al., 2006; Ksiazek et al., 2007; Sedelnikova et al., 2007). An *in vivo* age associated increase in TIF has been demonstrated in primate dermal fibroblasts (Herbig et al., 2006; Jeyapalan et al., 2007).

Tissues with high cell turnover would be expected to be especially sensitive to telomere shortening, and effects on the regenerative potential of stem cells have been of high interest (Wright and Shay, 2002; Campisi, 2005; Blasco, 2007). Although telomerase is known to be expressed in some stem cells compartments, several important examples of age-associated exhaustion of stem cell pools have also been found (Allsopp et al., 2003a; Allsopp et al., 2003b; Flores et al., 2005; Effros, 2006; Sarin and Artandi, 2007). One example is muscle satellite cells which are vital for the maintenance and repair of skeletal muscle, and satellite cell telomere shortening, replicative senescence, and rescue by telomerase have been reported (Decary et al., 1997; Cudre-Mauroux et al., 2003; Zhu et al., 2007).

A number of human disease states have been linked with excessive or accelerated telomere shortening. Although the phenotypes can be quite varied, all include some forms of age-associated pathology, premature aging and early death. Three syndromes are now known to be caused by mutations in genes that encode the telomerase holoenzyme: dyskeratosis congenita (Vulliamy et al., 2004; Vulliamy and Dokal, 2006), aplastic anemia (Yamaguchi et al., 2005; Drummond et al., 2007), and some cases of idiopathic pulmonary fibrosis (Armanios et al., 2007; Tsakiri et al., 2007). Patients with dyskeratosis congenita have short stature, suffer from hypogonadism and infertility, defects in the skin and hematopoietic system, and typically die of bone marrow failure. Many of the same pathologies also develop in telomerase knockout mice. The most recently discovered syndrome, idiopathic pulmonary fibrosis, is an adult-onset lethal disease of progressive lung scarring followed by respiratory failure.

Human segmental progeroid syndromes have also provided important connections to cellular senescence, and cells from such patients typically display greatly reduced *in vitro* life spans (Brown, 1990; Kudlow et al., 2007). The most thoroughly studied is Werner's syndrome (WS) and Hutchinson-Gilford progeria syndrome (HGPS). WS is an autosomal recessive disorder caused by loss of function mutations in the *WRN* gene encoding a member of the RecQ helicase family involved in DNA repair and transcription (Yu et al., 1996). HGPS is an autosomal dominant disorder caused by a gain of function mutation in the *LMNA* gene encoding lamin A/C, resulting in the expression of a splice variant designated progerin (Eriksson et al., 2003). WS patients display aging phenotypes that include hair graying, pattern baldness, bilateral cataracts, type 2 diabetes, hypogonadism, osteoporosis and atherosclerosis; death occurs at an average age of 47 usually as a result of cancer or coronary disease (Salk, 1982). HGPS patients display aging phenotypes that include extreme short stature, aged facial features, loss of hair, lipodystrophy, scleroderma, osteolysis and atherosclerosis; death occurs at an average age of 12 years as a result of myocardial infarction or stroke (Hennekam, 2006). Although the genetic defects in WS and HGPS are quite different, evidence has been presented for both that the accelerated cellular senescence is associated with increased rates of telomere shortening (Lebel and Leder, 1998; Chang et al., 2004; Multani and Chang, 2007; Crabbe et al., 2007; Huang et al., 2008). The underlying mechanistic causes affecting telomere maintenance are likely to be indirect and are not well understood (Cox and Faragher, 2007).

3. Accumulation of Senescent Cells *In Vivo*

To the extent that senescent cells are considered to confer deleterious effects, including the promotion of organismal aging, it is crucial to distinguish them from the majority of healthy but quiescent cells found in normal tissues. The original definition of irreversible arrest is clearly not feasible in this context, a situation that has led to a concerted search for biomarkers to assess the increasingly numerous molecular phenotypes of senescent cells. Unfortunately, to date no biomarker has been found that is either common to all types of senescent cells, or exclusive of all non-senescent cells. Nevertheless, if used in combination such markers can provide useful evidence for the presence of senescent cells, as well as clues concerning the mechanisms that generated them.

The first and still most widely used marker is the senescence associated β-galactosidase (SA β-Gal) (Dimri et al., 1995; Itahana et al., 2007). One of its limitation is the staining, under some conditions, of non-senescent cells (Yegorov et al., 1998; Severino et al., 2000; Cristofalo, 2005; Yang and Hu, 2005). Other concerns include the possibility of false negative results due to improper processing of samples, and the obscure molecular mechanisms that lead to its upregulation (Lee et al., 2006). More recent markers fall into several broad categories: components of signal transduction pathways known to be involved

in the establishment and maintenance of senescent states (such as the upregulation of cyclin dependent kinase inhibitors p16 or p21), markers of genotoxic stress (such as DNA damage foci and their colocalization with telomeres), appearance of a distinctive type of facultative heterochromatin designated senescence associated heterochromatin foci (SAHF), and the secretion of certain inflammatory cytokines and tissue remodeling factors (Campisi, 2005; Adams, 2007). The visualization of TIF has proven to be an especially robust marker, applicable to multiple species, cell culture as well as tissue sections, but is limited to reporting senescence initiated by telomere dysfunction.

Using such biomarkers senescent cells have been detected *in vivo* in a variety of tissues in a number of different organisms including mouse, primates and humans (Dimri et al., 1995; Krtolica and Campisi, 2002; Satyanarayana et al., 2003; Braig et al., 2005; Chen et al., 2005; Collado et al., 2005; Lazzerini Denchi et al., 2005; Lechel et al., 2005; Michaloglou et al., 2005; Herbig et al., 2006; Janzen et al., 2006; Krishnamurthy et al., 2006; Molofsky et al., 2006; Jeyapalan et al., 2007). Substantial evidence has accumulated that oncogene-induced senescence occurs *in vivo* in response to mutations in *RAS*, *RAF* and *PTEN* genes in mouse as well as human tumors (Braig et al., 2005; Chen et al., 2005; Collado et al., 2005; Michaloglou et al., 2005; Courtois-Cox et al., 2006). These populations of senescent cells were detected in benign, premalignant tumors, corroborating the importance of senescence as a tumor suppressor mechanism. Senescent cells can also be found in normal and tumor tissues following DNA-damaging chemotherapy (Schmitt et al., 2002; te Poele et al., 2002; Roninson, 2003; Roberson et al., 2005). Melanocytic naevi can reside in an organism for many years in a benign state, thus the detection of senescent cells in such naevi suggests that senescent cells could persist *in vivo* for a substantial periods of time (Michaloglou et al., 2005). In other cases, the apparent recognition and clearance of senescent cells by the immune system has been reported (Xue et al., 2007).

In culture, senescent cells accumulate with increasing population doublings until the majority of the culture has reached replicative senescence. *In vivo* studies have also found an age-associated increase in the occurrence of senescent cells in normal tissues (Dimri et al., 1995; Herbig and Sedivy, 2006; Ressler et al., 2006; Jeyapalan et al., 2007). Over 15% of dermal fibroblasts in very old baboons showed senescent phenotypes as determined by the presence of damaged telomeres, activation of ATM kinase and increased expression of p16. (Herbig et al., 2006; Jeyapalan et al., 2007). In contrast, no increase was found in skeletal muscle. Interestingly, the increase in heterochromatinization visualized using the chromatin remodeling protein HIRA as a marker was much more extensive and affected up to 80% of dermal fibroblasts in very old animals (Jeyapalan et al., 2007). The reasons for this discrepancy are not understood. The visualization and quantification of senescent cells in diverse species and tissues remains an area of considerable interest.

4. Senescent cells at sites of age related pathology

The presence of senescence-associated markers at sites of age related pathologies have provided further links between cellular senescence and aging. Telomere length as a function of donor age was found to decrease more rapidly in arterial than in venous endothelial cells, and telomere loss was greater in intimal than in medial cells (Chang and Harley, 1995). Other studies found that age-dependent telomere attrition is faster in the distal than in the proximal segment of the abdominal aorta, providing additional correlation between telomere attrition and hemodynamic stress (Okuda et al., 2000). Evidence for the presence of senescent endothelial cells overlying atherosclerotic plaques and other arterial lesions that could promote atherogenesis were found by several groups (Fenton et al., 2001; Minamino et al., 2002; Matthews et al., 2006). The gene expression changes seen in senescent cells could also be a factor in atherogenesis (Saito and Papaconstantinou, 2001; Vasile et al.,

2001). A recent study correlated telomere-based senescence in vascular smooth muscle cells with human atherosclerosis (Matthews et al., 2006). It has been speculated that the depletion of vascular smooth muscle cells is an important contributing factor to the atherosclerosis seen in HGPS patients (Stehbens et al., 1999).

Links between cellular senescence and another age related disorder, osteoarthritis, have also been reported. Senescent cells were detected in chrondrocyte clusters with mitotic activity in proximity to osteoarthritic lesions in (Price et al., 2002). In normal cartilage chrondocyte turnover is low but is thought to increase during disease progression, which in turn would be expected to promote telomere attrition.

A common disorder in the aging male population is benign prostatic hyperplasia, the continuing growth of the transition zone of the prostate due to both epithelial and stromal cell proliferation. Interestingly, this continued proliferation is thought to be due to the presence of senescent epithelial cells found at these sites (Choi et al., 2000; Castro et al., 2003). These cells secrete the cytokines IL-1α and IL-8 that stimulate stromal growth factor secretion, which in turn correlates with proliferation of the non senescent epithelial cells (Giri and Ittmann, 2000; Castro et al., 2003; Castro et al., 2004). Other cases where senescent cells have been implicated in altering the tissue microenvironment include the skin, where the secretion of growth factors, degradative enzymes and inflammatory cytokines by the aging dermal fibroblasts has been suggested to contribute to the characteristic aged skin morphology (Jenkins, 2002; Hornebeck, 2003; Boukamp, 2005). Moreover, factors secreted by senescent cells have been shown to stimulate the growth and angiogenic activity of premalignant cells (Krtolica et al., 2001; Dilley et al., 2003; Martens et al., 2003; Bavik et al., 2006; Coppe et al., 2006). Thus, the presence of senescent cells may promote the progression of nearby premalignant cells and consequently carcinogenesis in aging tissues (Campisi, 2005).

5. *CDKN2A* (p16) and aging

The cyclin dependent kinase inhibitor p16 has emerged as an important player in aging and age related disease. Biochemically, p16 inactivates CDK4 and CDK6, which maintains pRB in its active, hypophosphorylated form and consequently blocks cell cycle progression in G1 (Sherr and Roberts, 1999). Genetically, p16 is an important and potent tumor suppressor and is frequently inactivated in several human cancers, such as melanoma. Because p16 expression can be upregulated by a wide variety of stresses (Gil and Peters, 2006; Kim and Sharpless, 2006), it may be involved in many (all?) forms of senescence, and has thus received much recent attention as a promising biomarker (Michaloglou et al., 2005; Ressler et al., 2006).

p16 expression is very low or absent in young organisms but increases with advancing age (Zindy et al., 1997). This increase occurs primarily at the transcriptional level, has been detected in a wide variety of tissues, and can be delayed by caloric restriction which potently slows aging (Nielsen et al., 1999; Krishnamurthy et al., 2004). For example, p16 upregulation was found in aging human skin (Ressler et al., 2006). Senescence characterized by p16 upregulation as well as telomere shortening has been observed in human and mouse cardiomyocytes and may contribute to myocardial aging (Kajstura et al., 2000; Chimenti et al., 2003; Torella et al., 2004). A very similar situation has been found in human kidneys (Melk et al., 2000; Chkhotua et al., 2003). Induction of p16 was detected in rejected renal grafts that developed chronic allograft nephropathy, and it was suggested that accelerated senescence may be a contributing factor in graft rejection (Halloran et al., 1999; Chkhotua et al., 2003; Joosten et al., 2003).

The regulation of p16 is complex and only incompletely understood, especially *in vivo* (Gil and Peters, 2006). Three transcriptional regulators, namely the positive effectors ETS1 and JUNB, and the member of the polycomb family of repressors BMI1, have received particular attention (Passegué and Wagner, 2000; Ohtani et al., 2001; Krishnamurthy et al., 2004; Ressler et al., 2006; Yogev et al., 2006; Bracken et al., 2007). The regulation of p16 seems to be of particular importance in the age related decline of stem and progenitor cells. Early studies found that the induction of p16 correlated with the *in vivo* senescence of hematopoietic stem cells (Lewis et al., 2001; Meng et al., 2003; Park et al., 2003). Mice lacking *Bmi1* displayed a striking loss of hematopoietic cells as well as cerebellar neurons, which were correlated with increases in p16 expression and replicative failure of stem cells (Lessard and Sauvageau, 2003; Molofsky et al., 2003; Park et al., 2003). Conversely, an increase in regenerative capacity was found in the bone marrow, pancreatic islets and forebrain of p16 deficient mice (Janzen et al., 2006; Krishnamurthy et al., 2006; Molofsky et al., 2006). These findings reinforce the notion that the age associated upregulation of p16 restricts self renewal and unbalances tissue homeostasis. An interesting new link between cellular senescence and aging was provided by studies of mice deficient in the p53 related gene p63 (Keyes et al., 2005). Affected animals displayed enhanced p16 expression, stem cell defects, widespread senescence, and accelerated aging (Keyes and Mills, 2006).

6. Physiological relevance of senescent cells *in vivo*

Senescent cells are generated in response to a number of stimuli. Telomere attrition and oncogene activation are the best understood triggers, but a variety of stresses, disease or pathological conditions, and environmental and nutritional factors are also likely to play important roles. Although it is now possible to detect and even quantify senescent cells *in vivo* with some confidence, the physiological consequences of the existence of such cells are only beginning to be unravelled. Broadly speaking, we can envision cell autonomous and well as cell non-autonomous effects. Cell autonomous defects would be those arising from the depletion of functional cells from tissues. These would be expected to primarily affect proliferative tissues with high cell turnover, as has indeed been observed in, for example, telomerase deficient mice or telomerase compromised human syndromes. The depletion of stem cell pools would be expected to be especially debilitating because the effects would be amplified in the affected tissue. Cell non-autonomous effects would be mediated by the secretion of a variety of factors by senescent cells leading to effects on neighbouring cells. Such changes in the tissue microenvironment would be expected to affect tissue homeostasis and function, and could lead to tissue degeneration or other pathologies (Parrinello et al., 2005). One potentially very serious consequence could be the stimulation of preneoplastic cells leading to cancer progression (Krtolica et al., 2001; Dilley et al., 2003; Martens et al., 2003; Bavik et al., 2006). Factors secreted by senescent cells (or factors secreted by other cells after stimulation by senescent cells) could also have more systemic effects, for example due to the migration of the stimulated cells or endocrine effects of the secreted factors. To what extent the proinflammatory factors secreted by senescent cells contribute to the tissue or systemic inflammatory states often seen in the elderly is not known.

One of the cardinal phenotypes of senescent cells is their long term survival, and even resistance to apoptosis (Marcotte et al., 2004; Hampel et al., 2005). It has to be noted that this notion stems from cell culture studies, and that the identification of senescent cells *in vivo* has only recently become possible. Hence, the persistence of senescent cells in tissues, an issue of critical importance, is just beginning to be addressed. The secretion of inflammatory cytokines and other proteins by senescent cells would be expected, at least in some cases, to have the potential of identifying such cells to the immune system. Accordingly, the restoration of p53 activity in genetically engineered tumor cells in a mouse transgenic model resulted in the induction of senescence (Ventura et al., 2007; Xue et al.,

2007). Evidence was presented in these studies for the upregulation of inflammatory cytokines, engagement of the innate immune system, infiltration by phagocytic cells, and tumor regression. Interestingly, senescence was induced in liver carcinomas and sarcomas whereas apoptosis was induced in lymphomas, suggesting a balance between senescent and apoptotic responses that is strongly influenced by cell type. The fate of cells that succumb to senescence during natural aging due to influences such as telomere shortening or oxidative stress has not been reported to date.

7. Perspectives

The role of cellular senescence in a variety of age associated pathologies is becoming increasingly accepted. As discussed above, compelling experimental evidence has now linked increased rates of cellular senescence with accelerated aging. The extent to which, if any, cellular senescence contributes to the natural life span of any one species however remains to be established. As a case in point, although quantitative estimates of *in vivo* cellular senescence are just beginning to emerge, the evidence at hand would not support the contention that differential rates of cellular senescence account for the some 50-fold difference in mouse and human life spans. Perhaps cellular senescence will turn out to be one of several (many?) factors that influence or even promote aging. A common (albeit far from experimentally established) contention is that replicative senescence is likely to play major roles in long lived species with considerable renewable tissues. Establishing a causal link between cellular senescence and aging would ideally require an interventive deceleration of *in vivo* cellular senescence rates and a concomitant demonstration of increased maximum life span. Some have voiced the concern that the mouse, with its short lifespan, may be the wrong species for testing the relevance of replicative senescence. However, given that interventive deceleration of *in vivo* cellular senescence will almost certainly require extensive genetic engineering, the mouse is likely to remain the premier model system for some time. Clearly, the road ahead will not be easy, but it will certainly bring some very exciting science.

References

Adams J, Martin-Ruiz C, Pearce MS, White M, Parker L, von Zglinicki T. No association between socio-economic status and white blood cell telomere length. Aging Cell. 2007; 6:125–128. [PubMed: 17156082]

Adams PD. Remodeling of chromatin structure in senescent cells and its potential impact on tumor suppression and aging. Gene. 2007; 397:84–93. [PubMed: 17544228]

Allsopp RC, Morin GB, DePinho R, Harley CB, Weissman IL. Telomerase is required to slow telomere shortening and extend replicative lifespan of HSCs during serial transplantation. Blood. 2003a; 102:517–520. [PubMed: 12663456]

Allsopp RC, Morin GB, Horner JW, DePinho R, Harley CB, Weissman IL. Effect of TERT over-expression on the long-term transplantation capacity of hematopoietic stem cells. Nat. Med. 2003b; 9:369–371. [PubMed: 12669037]

Armanios MY, Chen JJ, Cogan JD, Alder JK, Ingersoll RG, Markin C, Lawson WE, Xie M, Vulto I, Phillips J.A.r. Lansdorp PM, Greider CW, Loyd JE. Telomerase mutations in families with idiopathic pulmonary fibrosis. N. Engl. J. Med. 2007; 356:1317–1326. [PubMed: 17392301]

Aviv A, Valdes AM, Spector TD. Human telomere biology: pitfalls of moving from the laboratory to epidemiology. Int. J. Epidemiol. 2006; 35:1424–1429. [PubMed: 16997848]

Baerlocher GM, Vulto I, de Jong G, Lansdorp PM. Flow cytometry and FISH to measure the average length of telomeres (flow FISH). Nat. Protoc. 2006; 1:2365–2376. [PubMed: 17406480]

Bavik C, Coleman I, Dean JP, Knudsen B, Plymate S, Nelson PS. The gene expression program of prostate fibroblast senescence modulates neoplastic epithelial cell proliferation through paracrine mechanisms. Cancer Res. 2006; 66:794–802. [PubMed: 16424011]

Bayreuther K, Rodemann HP, Francz PI, Maier K. Differentiation of fibroblast stem cells. J. Cell Sci. Suppl. 1988; 10:115–130. [PubMed: 3270453]

Bierman EL. The effect of donor age on the in vitro life span of cultured human arterial smooth-muscle cells. In Vitro. 1978; 14:951–955. [PubMed: 730203]

Blasco MA. Telomere length, stem cells and aging. Nat. Chem. Biol. 2007; 3:640–649. [PubMed: 17876321]

Bodnar AG, Ouellette M, Frolkis M, Holt SE, Chiu CP, Morin GB, Harley CB, Shay JW, Lichtsteiner S, Wright WE. Extension of life-span by introduction of telomerase into normal human cells. Science. 1998; 279:349–352. [PubMed: 9454332]

Boukamp P. Skin aging: a role for telomerase and telomere dynamics? Curr. Mol. Med. 2005; 5:171–177. [PubMed: 15974870]

Bracken AP, Kleine-Kohlbrecher D, Dietrich N, Pasini D, Gargiulo G, Beekman C, Theilgaard-Monch K, Minucci S, Porse BT, Marine JC, Hansen KH, Helin K. The Polycomb group proteins bind throughout the INK4A-ARF locus and are disassociated in senescent cells. Genes Dev. 2007; 21:525–530. [PubMed: 17344414]

Braig M, Lee S, Loddenkemper C, Rudolph C, Peters AH, Schlegelberger B, Stein H, Dorken B, Jenuwein T, Schmitt CA. Oncogene-induced senescence as an initial barrier in lymphoma development. Nature. 2005; 436:660–665. [PubMed: 16079837]

Brown WT. Genetic diseases of premature aging as models of senescence. Annu. Rev. Gerontol. Geriatr. 1990; 10:23–42. [PubMed: 2151708]

Bruce SA, Deamond SF, Ts'o PO. In vitro senescence of Syrian hamster mesenchymal cells of fetal to aged adult origin. Inverse relationship between in vivo donor age and in vitro proliferative capacity. Mech. Ageing Dev. 1986; 34:151–173. [PubMed: 3724250]

Campisi J. Senescent cells, tumor suppression, and organismal aging: good citizens, bad neighbors. Cell. 2005; 120:513–522. [PubMed: 15734683]

Campisi J, d'Adda di Fagagna F. Cellular senescence: when bad things happen to good cells. Nat. Rev. Mol. Cell Biol. 2007; 8:729–740. [PubMed: 17667954]

Canela A, Vera E, Klatt P, Blasco MA. High-throughput telomere length quantification by FISH and its application to human population studies. Proc. Natl. Acad. Sci. U.S.A. 2007; 104:5300–5305. [PubMed: 17369361]

Castro P, Giri D, Lamb D, Ittmann M. Cellular senescence in the pathogenesis of benign prostatic hyperplasia. Prostate. 2003; 55:30–38. [PubMed: 12640658]

Castro P, Xia C, Gomez L, Lamb DJ, Ittmann M. Interleukin-8 expression is increased in senescent prostatic epithelial cells and promotes the development of benign prostatic hyperplasia. Prostate. 2004; 60:153–159. [PubMed: 15162381]

Cawthon RM, Smith KR, O'Brien E, Sivatchenko A, Kerber RA. Association between telomere length in blood and mortality in people aged 60 years or older. Lancet. 2003; 361:393–395. [PubMed: 12573379]

Chang E, Harley CB. Telomere length and replicative aging in human vascular tissues. Proc. Natl. Acad. Sci. U.S.A. 1995; 92:11190–11194. [PubMed: 7479963]

Chang S, Multani AS, Cabrera NG, Naylor ML, Laud P, Lombard D, Pathak S, Guarente L, DePinho RA. Essential role of limiting telomeres in the pathogenesis of Werner syndrome. Nat. Genet. 2004; 36:877–882. [PubMed: 15235603]

Chen Z, Trotman LC, Shaffer D, Lin HK, Dotan ZA, Niki M, Koutcher JA, Scher HI, Ludwig T, Gerald W, Cordon-Cardo C, Pandolfi PP. Crucial role of p53-dependent cellular senescence in suppression of Pten-deficient tumorigenesis. Nature. 2005; 436:725–730. [PubMed: 16079851]

Cherkas LF, Aviv A, Valdes AM, Hunkin JL, Gardner JP, Surdulescu GL, Kimura M, Spector TD. The effects of social status on biological aging as measured by white-blood-cell telomere length. Aging Cell. 2006; 5:361–365. [PubMed: 16856882]

Chimenti C, Kajstura J, Torella D, Urbanek K, Heleniak H, Colussi C, Di Meglio F, Nadal-Ginard B, Frustaci A, Leri A, Maseri A, Anversa P. Senescence and death of primitive cells and myocytes lead to premature cardiac aging and heart failure. Circ. Res. 2003; 93:604–613. [PubMed: 12958145]

Chkhotua AB, Gabusi E, Altimari A, D'Errico A, Yakubovich M, Vienken J, Stefoni S, Chieco P, Yussim A, Grigioni WF. Increased expression of p16(INK4a) and p27(Kip1) cyclin-dependent kinase inhibitor genes in aging human kidney and chronic allograft nephropathy. Am. J. Kidney Dis. 2003; 41:1303–1313. [PubMed: 12776284]

Choi J, Shendrik I, Peacocke M, Peehl D, Buttyan R, Ikeguchi EF, Katz AE, Benson MC. Expression of senescence-associated beta-galactosidase in enlarged prostates from men with benign prostatic hyperplasia. Urology. 2000; 56:160–166. [PubMed: 10869659]

Collado M, Blasco MA, Serrano M. Cellular senescence in cancer and aging. Cell. 2007; 130:223–233. [PubMed: 17662938]

Collado M, Gil J, Efeyan A, Guerra C, Schuhmacher AJ, Barradas M, Benguria A, Zaballos A, Flores JM, Barbacid M, Beach D, Serrano M. Tumour biology: senescence in premalignant tumours. Nature. 2005; 436:642. [PubMed: 16079833]

Cong YS, Fan E, Wang E. Simultaneous proteomic profiling of four different growth states of human fibroblasts, using amine-reactive isobaric tagging reagents and tandem mass spectrometry. Mech. Ageing Dev. 2006; 127:332–343. [PubMed: 16434083]

Coppe JP, Kauser K, Campisi J, Beausejour CM. Secretion of vascular endothelial growth factor by primary human fibroblasts at senescence. J. Biol. Chem. 2006; 281:29568–29574. [PubMed: 16880208]

Cosme-Blanco W, Shen MF, Lazar AJ, Pathak S, Lozano G, Multani AS, Chang S. Telomere dysfunction suppresses spontaneous tumorigenesis in vivo by initiating p53-dependent cellular senescence. EMBO R. 2007; 8:497–503.

Courtois-Cox S, Genther Williams SM, Reczek EE, Johnson BW, McGillicuddy LT, Johannessen CM, Hollstein PE, MacCollin M, Cichowski K. A negative feedback signaling network underlies oncogene-induced senescence. Cancer Cell. 2006; 10:459–472. [PubMed: 17157787]

Cox LS, Faragher RG. From old organisms to new molecules: integrative biology and therapeutic targets in accelerated human ageing. Cell. Mol. Life Sci. 2007; 64:2620–2641. [PubMed: 17660942]

Crabbe L, Jauch A, Naeger CM, Holtgreve-Grez H, Karlseder J. Telomere dysfunction as a cause of genomic instability in Werner syndrome. Proc. Natl. Acad. Sci. U.S.A. 2007; 104:2205–2210. [PubMed: 17284601]

Cristofalo VJ. SA beta Gal staining: biomarker or delusion. Exp. Gerontol. 2005; 40:836–838. [PubMed: 16181758]

Cudre-Mauroux C, Occhiodoro T, Konig S, Salmon P, Bernheim L, Trono D. Lentivector-mediated transfer of Bmi-1 and telomerase in muscle satellite cells yields a duchenne myoblast cell line with long-term genotypic and phenotypic stability. Hum. Gene Ther. 2003; 14:1525–1533. [PubMed: 14577914]

d'Adda di Fagagna F, Reaper PM, Clay-Farrace L, Fiegler H, Carr P, Von Zglinicki T, Saretzki G, Carter NP, Jackson SP. A DNA damage checkpoint response in telomere-initiated senescence. Nature. 2003; 426:194–198. [PubMed: 14608368]

d'Adda di Fagagna F, Teo SH, Jackson SP. Functional links between telomeres and proteins of the DNA-damage response. Genes Dev. 2004; 18:1781–1799. [PubMed: 15289453]

De Meyer T, Rietzschel ER, De Buyzere ML, Van Criekinge W, Bekaert S. Studying telomeres in a longitudinal population based study. Front. Biosci. 2008; 13:2960–2970. [PubMed: 17981769]

Decary S, Mouly V, Hamida CB, Sautet A, Barbet JP, Butler-Browne GS. Replicative potential and telomere length in human skeletal muscle: implications for satellite cell-mediated gene therapy. Hum. Gene Ther. 1997; 8:1429–1438. [PubMed: 9287143]

Dilley TK, Bowden GT, Chen QM. Novel mechanisms of sublethal oxidant toxicity: induction of premature senescence in human fibroblasts confers tumor promoter activity. Exp. Cell Res. 2003; 290:38–48. [PubMed: 14516786]

Dimri GP, Lee X, Basile G, Acosta M, Scott G, Roskelley C, Medrano EE, Linskens M, Rubelj I, Pereira-Smith O, et al. A biomarker that identifies senescent human cells in culture and in aging skin in vivo. Proc. Natl. Acad. Sci. U.S.A. 1995; 92:9363–9367. [PubMed: 7568133]

Drayton S, Peters G. Immortalisation and transformation revisited. Curr. Opin. Genet. Dev. 2002; 12:98–104. [PubMed: 11790562]

Drummond MW, Balabanov S, Holyoake TL, Brummendorf TH. Telomere biology in normal and leukemic hematopoietic stem cells. Stem Cells. 2007; 25:1853–1861. [PubMed: 17510216]

Effros RB. Telomerase induction in T cells: A cure for aging and disease? Exp. Gerontol. 2006 Epub ahead of print.

Ellison PT. How old are you now? Aging Cell. 2006; 6:581. [PubMed: 17081155]

Epel ES, Blackburn EH, Lin J, Dhabhar FS, Adler NE, Morrow JD, Cawthon RM. Accelerated telomere shortening in response to life stress. Proc. Natl. Acad. Sci. U.S.A. 2004; 101:17312–17315. [PubMed: 15574496]

Eriksson M, Brown WT, Gordon LB, Glynn MW, Singer J, Scott L, Erdos MR, Robbins CM, Moses TY, Berglund P, Dutra A, Pak E, Durkin S, Csoka AB, Boehnke M, Glover TW, Collins FS. Recurrent de novo point mutations in lamin A cause Hutchinson-Gilford progeria syndrome. Nature. 2003; 423:293–298. [PubMed: 12714972]

Feldser DM, Greider CW. Short telomeres limit tumor progression in vivo by inducing senescence. Cancer Cell. 2007; 11:461–469. [PubMed: 17433785]

Fenton M, Barker S, Kurz DJ, Erusalimsky JD. Cellular senescence after single and repeated balloon catheter denudations of rabbit carotid arteries. Arterioscler. Thromb. Vasc. Biol. 2001; 21:220–226. [PubMed: 11156856]

Flores I, Cayuela ML, Blasco MA. Effects of telomerase and telomere length on epidermal stem cell behavior. Science. 2005; 309

Funayama R, Saito M, Tanobe H, Ishikawa F. Loss of linker histone H1 in cellular senescence. J. Cell Biol. 2006; 175:869–880. [PubMed: 17158953]

Garcia-Cao I, Garcia-Cao M, Tomas-Loba A, Martin-Caballero J, Flores JM, Klatt P, Blasco MA, Serrano M. Increased p53 activity does not accelerate telomere-driven ageing. EMBO R. 2006; 7:546–552.

Gil J, Peters G. Regulation of the INK4b-ARF-INK4a tumour suppressor locus: all for one or one for all. Nat. Rev. Mol. Cell Biol. 2006; 7:667–677. [PubMed: 16921403]

Giri D, Ittmann M. Interleukin-1alpha is a paracrine inducer of FGF7, a key epithelial growth factor in benign prostatic hyperplasia. Am. J. Pathol. 2000; 157:249–255. [PubMed: 10880394]

Gonos ES, Derventzi A, Kveiborg M, Agiostratidou G, Kassem M, Clark BF, Jat PS, Rattan SI. Cloning and identification of genes that associate with mammalian replicative senescence. Exp. Cell Res. 1998; 240:66–74. [PubMed: 9570922]

Halloran PF, Melk A, Barth C. Rethinking chronic allograft nephropathy: the concept of accelerated senescence. J. Am. Soc. Nephrol. 1999; 10:167–181. [PubMed: 9890324]

Hampel B, Wagner M, Teis D, Zwerschke W, Huber LA, Jansen-Durr P. Apoptosis resistance of senescent human fibroblasts is correlated with the absence of nuclear IGFBP-3. Aging Cell. 2005; 4:325–330. [PubMed: 16300485]

Harley CB, Futcher AB, Greider CW. Telomeres shorten during ageing of human fibroblasts. Nature. 1990; 345:458–460. [PubMed: 2342578]

Hayflick L. Theories of biological aging. Exp. Gerontol. 1985; 20:145–159. [PubMed: 3905424]

Hayflick L, Moorhead PS. The serial cultivation of human diploid cell strains. Exp. Cell Res. 1961; 25:585–621.

Hemann MT, Strong MA, Hao LY, Greider CW. The shortest telomere, not average telomere length, is critical for cell viability and chromosome stability. Cell. 2001; 107:67–77. [PubMed: 11595186]

Hennekam RC. Hutchinson-Gilford progeria syndrome: review of the phenotype. Am. J. Med. Genet. 2006; 140A:2603–2624. [PubMed: 16838330]

Herbig U, Ferreira M, Condel L, Carey D, Sedivy JM. Cellular senescence in aging primates. Science. 2006; 311:1257. [PubMed: 16456035]

Herbig U, Jobling WA, Chen BP, Chen DJ, Sedivy JM. Telomere shortening triggers senescence of human cells through a pathway involving ATM, p53, and p21(CIP1), but not p16(INK4a). Mol. Cell. 2004; 14:501–513. [PubMed: 15149599]

Herbig U, Sedivy JM. Regulation of growth arrest in senescence: telomere damage is not the end of the story. Mech. Ageing Dev. 2006; 127:16–24. [PubMed: 16229875]

Herbig U, Wei W, Dutriaux A, Jobling WA, Sedivy JM. Real-time imaging of transcriptional activation in live cells reveals rapid up-regulation of the cyclin-dependent kinase inhibitor gene CDKN1A in replicative cellular senescence. Aging Cell. 2003; 2:295–304. [PubMed: 14677632]

Hornebeck W. Down-regulation of tissue inhibitor of matrix metalloprotease-1 (TIMP-1) in aged human skin contributes to matrix degradation and impaired cell growth and survival. Pathol. Biol. 2003; 51:569–573. [PubMed: 14622947]

Hornsby PJ. Short telomeres: cause or consequence of aging? Aging Cell. 2006; 6:577–578.

Huang S, Risques RA, Martin GM, Rabinovitch PS, Oshima J. Accelerated telomere shortening and replicative senescence in human fibroblasts overexpressing mutant and wild-type lamin A. Exp. Cell Res. 2008; 314:82–91. [PubMed: 17870066]

Itahana K, Campisi J, Dimri GP. Methods to detect biomarkers of cellular senescence: the senescence-associated beta-galactosidase assay. Methods Mol. Biol. 2007; 371:21–31. [PubMed: 17634571]

Janzen V, Forkert R, Fleming HE, Saito Y, Waring MT, Dombkowski DM, Cheng T, DePinho RA, Sharpless NE, Scadden DT. Stem-cell ageing modified by the cyclin-dependent kinase inhibitor p16INK4a. Nature. 2006; 443:421–426. [PubMed: 16957735]

Jenkins G. Molecular mechanisms of skin ageing. Mech. Ageing Dev. 2002; 123:801–810. [PubMed: 11869737]

Jeyapalan JC, Ferreira M, Sedivy JM, Herbig U. Accumulation of senescent cells in mitotic tissue of aging primates. Mech. Ageing Dev. 2007; 128:36–44. [PubMed: 17116315]

Joosten SA, van Ham V, Nolan CE, Borrias MC, Jardine AG, Shiels PG, van Kooten C, Paul LC. Telomere shortening and cellular senescence in a model of chronic renal allograft rejection. Am. J. Pathol. 2003; 162:1305–1312. [PubMed: 12651622]

Kajstura J, Pertoldi B, Leri A, Beltrami CA, Deptala A, Darzynkiewicz Z, Anversa P. Telomere shortening is an in vivo marker of myocyte replication and aging. Am. J. Pathol. 2000; 156:813–819. [PubMed: 10702397]

Keyes WM, Mills AA. p63: a new link between senescence and aging. Cell Cycle. 2006; 5:260–265. [PubMed: 16434880]

Keyes WM, Wu Y, Vogel H, Guo X, Lowe SW, Mills AA. p63 deficiency activates a program of cellular senescence and leads to accelerated aging. Genes Dev. 2005; 19:1986–1999. [PubMed: 16107615]

Kim WY, Sharpless NE. The regulation of INK4/ARF in cancer and aging. Cell. 2006; 127:265–275. [PubMed: 17055429]

Kimura M, Barbieri M, Gardner JP, Skurnick J, Cao X, van Riel N, Rizzo MR, Paolisso G, Aviv A. Leukocytes of exceptionally old persons display ultra-short telomeres. Am. J. Physiol. Regul. Integr. Comp. Physiol. 2007 Epub ahead of print.

Kirkwood TB, Austad SN. Why do we age? Nature. 2000; 408:233–238. [PubMed: 11089980]

Krishnamurthy J, Ramsey MR, Ligon KL, Torrice C, Koh A, Bonner-Weir S, Sharpless NE. p16INK4a induces an age-dependent decline in islet regenerative potential. Nature. 2006; 443:453–457. [PubMed: 16957737]

Krishnamurthy J, Torrice C, Ramsey MR, Kovalev GI, Al-Regaiey K, Su L, Sharpless NE. Ink4a/Arf expression is a biomarker of aging. J. Clin. Invest. 2004; 114:1299–1307. [PubMed: 15520862]

Krtolica A, Campisi J. Cancer and aging: a model for the cancer promoting effects of the aging stroma. Int. J. Biochem. Cell Biol. 2002; 34:1401–1414. [PubMed: 12200035]

Krtolica A, Parrinello S, Lockett S, Desprez PY, Campisi J. Senescent fibroblasts promote epithelial cell growth and tumorigenesis: a link between cancer and aging. Proc. Natl. Acad. Sci. U.S.A. 2001; 98:12072–12077. [PubMed: 11593017]

Ksiazek K, Passos JF, Olijslagers S, Saretzki G, Martin-Ruiz C, von Zglinicki T. Premature senescence of mesothelial cells is associated with non-telomeric DNA damage. Biochem. Biophys. Res. Commun. 2007; 362:707–711. [PubMed: 17720141]

Kudlow BA, Kennedy BK, Monnat RJJ. Werner and Hutchinson-Gilford progeria syndromes: mechanistic basis of human progeroid diseases. Nat. Rev. Mol. Cell Biol. 2007; 8:394–404. [PubMed: 17450177]

Kuh D. A life course perspective on telomere length and social inequalities in aging. Aging Cell. 2006; 6:579–580. [PubMed: 17081153]

Lansdorp PM. Stress, social rank and leukocyte telomere length. Aging Cell. 2006; 6:583–584. [PubMed: 17081156]

Lazzerini Denchi E, Attwooll C, Pasini D, Helin K. Deregulated E2F activity induces hyperplasia and senescence-like features in the mouse pituitary gland. Mol. Cell Biol. 2005; 25:2660–2672. [PubMed: 15767672]

Le Guilly Y, Simon M, Lenoir P, Bourel M. Long-term culture of human adult liver cells: morphological changes related to in vitro senescence and effect of donor's age on growth potential. Gerontologia. 1973; 19:303–313. [PubMed: 4788965]

Lebel M, Leder P. A deletion within the murine Werner syndrome helicase induces sensitivity to inhibitors of topoisomerase and loss of cellular proliferative capacity. Proc. Natl. Acad. Sci. U.S.A. 1998; 95:13097–13102. [PubMed: 9789047]

Lechel A, Satyanarayana A, Ju Z, Plentz RR, Schaetzlein S, Rudolph C, Wilkens L, Wiemann SU, Saretzki G, Malek NP, Manns MP, Buer J, Rudolph KL. The cellular level of telomere dysfunction determines induction of senescence or apoptosis in vivo. EMBO R. 2005; 6:275–281.

Lee BY, Han JA, Im JS, Morrone A, Johung K, Goodwin EC, Kleijer WJ, DiMaio D, Hwang ES. Senescence-associated β-galactosidase is lysosomal β-galactosidase. Aging Cell. 2006; 5:187–195. [PubMed: 16626397]

Lessard J, Sauvageau G. Bmi-1 determines the proliferative capacity of normal and leukaemic stem cells. Nature. 2003; 423:255–260. [PubMed: 12714970]

Lewis JL, Chinswangwatanakul W, Zheng B, Marley SB, Nguyen DX, Cross NC, Banerji L, Glassford J, Thomas NS, Goldman JM, Lam EW, Gordon MY. The influence of INK4 proteins on growth and self-renewal kinetics of hematopoietic progenitor cells. Blood. 2001; 97:2604–2610. [PubMed: 11313248]

Marcotte R, Lacelle C, Wang E. Senescent fibroblasts resist apoptosis by downregulating caspase-3. Mech. Ageing Dev. 2004; 125:777–783. [PubMed: 15541772]

Martens JW, Sieuwerts AM, Bolt-deVries J, Bosma PT, Swiggers SJ, Klijn JG, Foekens JA. Aging of stromal-derived human breast fibroblasts might contribute to breast cancer progression. Thromb. Haemost. 2003; 89:393–404. [PubMed: 12574821]

Martin GM, Sprague CA, Epstein CJ. Replicative life-span of cultivated human cells. Effects of donor's age, tissue, and genotype. Lab. Invest. 1970; 23:86–92. [PubMed: 5431223]

Matsumura T, Zerrudo Z, Hayflick L. Senescent human diploid cells in culture: survival, DNA synthesis and morphology. J. Gerontol. 1979; 34:328–334. [PubMed: 429765]

Matthews C, Gorenne I, Scott S, Figg N, Kirkpatrick P, Ritchie A, Goddard M, Bennett M. Vascular smooth muscle cells undergo telomere-based senescence in human atherosclerosis: effects of telomerase and oxidative stress. Circ. Res. 2006; 99:156–164. [PubMed: 16794190]

Melk A, Ramassar V, Helms LM, Moore R, Rayner D, Solez K, Halloran PF. Telomere shortening in kidneys with age. J. Am. Soc. Nephrol. 2000; 11:444–453. [PubMed: 10703668]

Meng A, Wang Y, Van Zant G, Zhou D. Ionizing radiation and busulfan induce premature senescence in murine bone marrow hematopoietic cells. Cancer Res. 2003; 63:5414–5419. [PubMed: 14500376]

Michaloglou C, Vredeveld LC, Soengas MS, Denoyelle C, Kuilman T, van der Horst CM, Majoor DM, Shay JW, Mooi WJ, Peeper DS. BRAFE600-associated senescence-like cell cycle arrest of human naevi. Nature. 2005; 436:720–724. [PubMed: 16079850]

Minamino T, Komuro I. Role of telomeres in vascular senescence. Front. Biosci. 2008; 13:2971–2979. [PubMed: 17981770]

Minamino T, Miyauchi H, Yoshida T, Ishida Y, Yoshida H, Komuro I. Endothelial cell senescence in human atherosclerosis: role of telomere in endothelial dysfunction. Circulation. 2002; 105:1541–1544. [PubMed: 11927518]

Molofsky AV, Pardal R, Iwashita T, Park IK, Clarke MF, Morrison SJ. Bmi-1 dependence distinguishes neural stem cell self-renewal from progenitor proliferation. Nature. 2003; 425:962–967. [PubMed: 14574365]

Molofsky AV, Slutsky SG, Joseph NM, He S, Pardal R, Krishnamurthy J, Sharpless NE, Morrison SJ. Increasing p16INK4a expression decreases forebrain progenitors and neurogenesis during ageing. Nature. 2006; 443:448–452. [PubMed: 16957738]

Multani AS, Chang S. WRN at telomeres: implications for aging and cancer. J. Cell Sci. 2007; 120:713–721. [PubMed: 17314245]

Narita M, Nunez S, Heard E, Lin AW, Hearn SA, Spector DL, Hannon GJ, Lowe SW. Rb-mediated heterochromatin formation and silencing of E2F target genes during cellular senescence. Cell. 2003; 113:703–716. [PubMed: 12809602]

Nielsen GP, Stemmer-Rachamimov AO, Shaw J, Roy JE, Koh J, Louis DN. Immunohistochemical survey of p16INK4A expression in normal human adult and infant tissues. Lab. Invest. 1999; 79:1137–1143. [PubMed: 10496532]

Ogami M, Ikura Y, Ohsawa M, Matsuo T, Kayo S, Yoshimi N, Hai E, Shirai N, Ehara S, Komatsu R, Naruko T, Ueda M. Telomere shortening in human coronary artery diseases. Arterioscler. Thromb. Vasc. Biol. 2004; 24:546–550. [PubMed: 14726417]

Ohtani N, Zebedee Z, Huot TJ, Stinson JA, Sugimoto M, Ohashi Y, Sharrocks AD, Peters G, Hara E. Opposing effects of Ets and Id proteins on p16INK4a expression during cellular senescence. Nature. 2001; 409:1067–1070. [PubMed: 11234019]

Okuda K, Khan MY, Skurnick J, Kimura M, Aviv H, Aviv A. Telomere attrition of the human abdominal aorta: relationships with age and atherosclerosis. Atherosclerosis. 2000; 152:391–398. [PubMed: 10998467]

Olovnikov AM. A theory of marginotomy. The incomplete copying of template margin in enzymic synthesis of polynucleotides and biological significance of the phenomenon. J. Theor. Biol. 1973; 41:181–190. [PubMed: 4754905]

Panossian LA, Porter VR, Valenzuela HF, Zhu X, Reback E, Masterman D, Cummings JL, Effros RB. Telomere shortening in T cells correlates with Alzheimer's disease status. Neurobiol. Aging. 2003; 24:77–84. [PubMed: 12493553]

Park IK, Qian D, Kiel M, Becker MW, Pihalja M, Weissman IL, Morrison SJ, Clarke MF. Bmi-1 is required for maintenance of adult self-renewing haematopoietic stem cells. Nature. 2003; 423:302–305. [PubMed: 12714971]

Parrinello S, Coppe JP, Krtolica A, Campisi J. Stromal-epithelial interactions in aging and cancer: senescent fibroblasts alter epithelial cell differentiation. J. Cell Sci. 2005; 118:485–496. [PubMed: 15657080]

Pascal T, Debacq-Chainiaux F, Chretien A, Bastin C, Dabee AF, Bertholet V, Remacle J, Toussaint O. Comparison of replicative senescence and stress-induced premature senescence combining differential display and low-density DNA arrays. FEBS Lett. 2005; 579:3651–3659. [PubMed: 15963989]

Passegué E, Wagner EF. JunB suppresses cell proliferation by transcriptional activation of p16(INK4a) expression. EMBO J. 2000; 19:2969–2979. [PubMed: 10856241]

Pignolo RJ, Martin BG, Horton JH, Kalbach AN, Cristofalo VJ. The pathway of cell senescence: WI-38 cells arrest in late G1 and are unable to traverse the cell cycle from a true G0 state. Exp. Gerontol. 1998; 33:67–80. [PubMed: 9467718]

Pignolo RJ, Rotenberg MO, Cristofalo VJ. Alterations in contact and density-dependent arrest state in senescent WI-38 cells. In Vitro Cell Dev. Biol. Anim. 1994; 30A:471–476. [PubMed: 7952516]

Price JS, Waters JG, Darrah C, Pennington C, Edwards DR, Donell ST, Clark IM. The role of chondrocyte senescence in osteoarthritis. Aging Cell. 2002; 1:57–65. [PubMed: 12882354]

Ressler S, Bartkova J, Niederegger H, Bartek J, Scharffetter-Kochanek K, Jansen-Durr P, Wlaschek M. p16INK4A is a robust in vivo biomarker of cellular aging in human skin. Aging Cell. 2006; 5:379–389. [PubMed: 16911562]

Rheinwald JG, Green H. Serial cultivation of strains of human epidermal keratinocytes: the formation of keratinizing colonies from single cells. Cell. 1975; 6:331–343. [PubMed: 1052771]

Roberson RS, Kussick SJ, Vallieres E, Chen SY, Wu DY. Escape from therapy-induced accelerated cellular senescence in p53-null lung cancer cells and in human lung cancers. Cancer Res. 2005; 65:2795–2803. [PubMed: 15805280]

Roninson IB. Tumor cell senescence in cancer treatment. Cancer Res. 2003; 63:2705–2715. [PubMed: 12782571]

Rose, MR. The Evolutionary Biology of Aging. Oxford University Press; Oxford: 1991.

Sager R. Senescence as a mode of tumor suppression. Environ. Health Perspect. 1991; 93:59–62. [PubMed: 1663451]

Saito H, Papaconstantinou J. Age-associated differences in cardiovascular inflammatory gene induction during endotoxic stress. J. Biol. Chem. 2001; 276:29307–29312. [PubMed: 11382779]

Salk D. Can we learn about aging from a study of Werner's syndrome? J. Am. Geriatr. Soc. 1982; 30:334–339. [PubMed: 7077011]

Sarin KY, Artandi SE. Aging, graying and loss of melanocyte stem cells. Stem Cell Rev. 2007; 3:212–217. [PubMed: 17917134]

Satyanarayana A, Wiemann SU, Buer J, Lauber J, Dittmar KE, Wustefeld T, Blasco MA, Manns MP, Rudolph KL. Telomere shortening impairs organ regeneration by inhibiting cell cycle re-entry of a subpopulation of cells. EMBO J. 2003; 22:4003–4013. [PubMed: 12881434]

Schmitt CA, Fridman JS, Yang M, Lee S, Baranov E, Hoffman RM, Lowe SW. A senescence program controlled by p53 and p16INK4a contributes to the outcome of cancer therapy. Cell. 2002; 109:335–346. [PubMed: 12015983]

Schneider EL, Mitsui Y. The relationship between in vitro cellular aging and in vivo human age. Proc. Natl. Acad. Sci. U.S.A. 1976; 73:3584–3588. [PubMed: 1068470]

Schwarze SR, DePrimo SE, Grabert LM, Fu VX, Brooks JD, Jarrard DF. Novel pathways associated with bypassing cellular senescence in human prostate epithelial cells. J. Biol. Chem. 2002; 277:14877–14883. [PubMed: 11836256]

Sedelnikova OA, Horikawa I, Redon C, Nakamura A, Zimonjic DB, Popescu NC, Bonner WM. Delayed kinetics of DNA double-strand break processing in normal and pathological aging. Aging Cell. 2007 Epub ahead of print.

Sedelnikova OA, Horikawa I, Zimonjic DB, Popescu NC, Bonner WM, Barrett JC. Senescing human cells and ageing mice accumulate DNA lesions with unrepairable double-strand breaks. Nat. Cell Biol. 2004; 6:168–170. [PubMed: 14755273]

Sedivy, JM.; Munoz-Najar, UM.; Jeyapalan, JC.; Campisi, J. Cellular senescence: a link between tumor suppression and organismal aging?. In: Guarente, L.; Partridge, L., editors. The Molecular Biology of Aging. Cold Spring Harbor Laboratory Press; Cold Spring Harbor, NY: 2007.

Semov A, Marcotte R, Semova N, Ye X, Wang E. Microarray analysis of E-box binding-related gene expression in young and replicatively senescent human fibroblasts. Anal. Biochem. 2002; 302:38–51. [PubMed: 11846374]

Serrano M, Lin AW, McCurrach ME, Beach D, Lowe SW. Oncogenic ras provokes premature cell senescence associated with accumulation of p53 and p16INK4a. Cell. 1997; 88:593–602. [PubMed: 9054499]

Seshadri T, Campisi J. Repression of c-fos transcription and an altered genetic program in senescent human fibroblasts. Science. 1990; 247:205–209. [PubMed: 2104680]

Severino J, Allen RG, Balin S, Balin A, Cristofalo VJ. Is beta-galactosidase staining a marker of senescence in vitro and in vivo? Exp. Cell Res. 2000; 257:162–171. [PubMed: 10854064]

Sfeir AJ, Chai W, Shay JW, Wright WE. Telomere end processing: the terminal nucleotides of human chromosomes. Mol. Cell. 2005; 18:131–138. [PubMed: 15808515]

Shelton DN, Chang E, Whittier PS, Choi D, Funk WD. Microarray analysis of replicative senescence. Curr. Biol. 1999; 9:939–945. [PubMed: 10508581]

Sherr CJ, Roberts JM. CDK inhibitors: positive and negative regulators of G1-phase progression. Genes Dev. 1999; 13:1501–1512. [PubMed: 10385618]

Stehbens WE, Wakefield SJ, Gilbert-Barness E, Olson RE, Ackerman J. Histological and ultrastructural features of atherosclerosis in progeria. Cardiovasc. Pathol. 1999; 8:29–39. [PubMed: 10722246]

Takai H, Smogorzewska A, de Lange T. DNA damage foci at dysfunctional telomeres. Curr. Biol. 2003; 13:1549–1556. [PubMed: 12956959]

te Poele RH, Okorokov AL, Jardine L, Cummings J, Joel SP. DNA damage is able to induce senescence in tumor cells in vitro and in vivo. Cancer Res. 2002; 62:1876–1883. [PubMed: 11912168]

Torella D, Rota M, Nurzynska D, Musso E, Monsen A, Shiraishi I, Zias E, Walsh K, Rosenzweig A, Sussman MA, Urbanek K, Nadal-Ginard B, Kajstura J, Anversa P, Leri A. Cardiac stem cell and

myocyte aging, heart failure, and insulin-like growth factor-1 overexpression. Circ. Res. 2004; 94:514–524. [PubMed: 14726476]

Trougakos IP, Saridaki A, Panayotou G, Gonos ES. Identification of differentially expressed proteins in senescent human embryonic fibroblasts. Mech. Ageing Dev. 2006; 127:88–92. [PubMed: 16213575]

Tsakiri KD, Cronkhite JT, Kuan PJ, Xing C, Raghu G, Weissler JC, Rosenblatt RL, Shay JW, Garcia CK. Adult-onset pulmonary fibrosis caused by mutations in telomerase. Proc. Natl. Acad. Sci. U.S.A. 2007; 104:7552–7557. [PubMed: 17460043]

Valdes AM, Andrew T, Gardner JP, Kimura M, Oelsner E, Cherkas LF, Aviv A, Spector TD. Obesity, cigarette smoking, and telomere length in women. Lancet. 2005; 366:662–664. [PubMed: 16112303]

Vasile E, Tomita Y, Brown LF, Kocher O, Dvorak HF. Differential expression of thymosin beta-10 by early passage and senescent vascular endothelium is modulated by VPF/VEGF: evidence for senescent endothelial cells in vivo at sites of atherosclerosis. FASEB J. 2001; 15:458–466. [PubMed: 11156961]

Ventura A, Kirsch DG, McLaughlin ME, Tuveson DA, Grimm J, Lintault L, Newman J, Reczek EE, Weissleder R, Jacks T. Restoration of p53 function leads to tumour regression in vivo. Nature. 2007; 445:661–665. [PubMed: 17251932]

von Zglinicki T. Oxidative stress shortens telomeres. Trends Biochem. Sci. 2002; 27:339–344. [PubMed: 12114022]

von Zglinicki T, Martin-Ruiz CM. Telomeres as biomarkers for ageing and age-related diseases. Curr. Mol. Med. 2005; 5:197–203. [PubMed: 15974873]

von Zglinicki T, Saretzki G, Docke W, Lotze C. Mild hyperoxia shortens telomeres and inhibits proliferation of fibroblasts: a model for senescence? Exp. Cell Res. 1995; 220:186–193. [PubMed: 7664835]

Vulliamy T, Dokal I. Dyskeratosis congenita. Semin. Hematol. 2006; 43:157–166. [PubMed: 16822458]

Vulliamy T, Marrone A, Szydlo R, Walne A, Mason PJ, Dokal I. Disease anticipation is associated with progressive telomere shortening in families with dyskeratosis congenita due to mutations in TERC. Nat Genet. 2004; 36:447–449. [PubMed: 15098033]

Williams GC. Pleiotrophy, natural selection and the evolution of senescence. Evolution. 1957; 11:398–411.

Wright WE, Shay JW. Historical claims and current interpretations of replicative aging. Nat. Biotechnol. 2002; 20:682–688. [PubMed: 12089552]

Xie L, Tsaprailis G, Chen QM. Proteomic identification of insulin-like growth factor-binding protein-6 induced by sublethal H2O2 stress from human diploid fibroblasts. Mol. Cell. Proteomics. 2005; 4:1273–1283. [PubMed: 15958393]

Xue W, Zender L, Miething C, Dickins RA, Hernando E, Krizhanovsky V, Cordon-Cardo C, Lowe SW. Senescence and tumour clearance is triggered by p53 restoration in murine liver carcinomas. Nature. 2007; 445:656–660. [PubMed: 17251933]

Yamaguchi H, Calado RT, Ly H, Kajigaya S, Baerlocher GM, Chanock SJ, Lansdorp PM, Young NS. Mutations in TERT, the gene for telomerase reverse transcriptase, in aplastic anemia. N. Engl. J. Med. 2005; 352:1413–1424. [PubMed: 15814878]

Yang NC, Hu ML. The limitations and validities of senescence associated-beta-galactosidase activity as an aging marker for human foreskin fibroblast Hs68 cells. Exp. Gerontol. 2005; 40:813–819. [PubMed: 16154306]

Ye X, Zerlanko B, Kennedy A, Banumathy G, Zhang R, Adams PD. Downregulation of Wnt signaling is a trigger for formation of facultative heterochromatin and onset of cell senescence in primary human cells. Mol. Cell. 2007; 27:183–196. [PubMed: 17643369]

Yegorov YE, Akimov SS, Hass R, Zelenin AV, Prudovsky IA. Endogenous beta-galactosidase activity in continuously nonproliferating cells. Exp. Cell Res. 1998; 243:207–211. [PubMed: 9716464]

Yogev O, Anzi S, Inoue K, E S. Induction of transcriptionally active Jun proteins regulates drug-induced senescence. J. Biol. Chem. 2006; 281:34475–34483. [PubMed: 16966326]

Yoon IK, Kim HK, Kim YK, Song IH, Kim W, Kim S, Baek SH, Kim JH, Kim JR. Exploration of replicative senescence-associated genes in human dermal fibroblasts by cDNA microarray technology. Exp. Gerontol. 2004; 39:1369–1378. [PubMed: 15489060]

Yu CE, Oshima J, Fu YH, Wijsman EM, Hisama F, Alisch R, Matthews S, Nakura J, Miki T, Ouais S, Martin GM, Mulligan J, Schellenberg GD. Positional cloning of the Werner's syndrome gene. Science. 1996; 272:258–262. [PubMed: 8602509]

Zdanov S, Debacq-Chainiaux F, Remacle J, Toussaint O. Identification of p38MAPK-dependent genes with changed transcript abundance in H2O2-induced premature senescence of IMR-90 hTERT human fibroblasts. FEBS Lett. 2006; 580:6455–6463. [PubMed: 17101135]

Zhang H, Pan KH, Cohen SN. Senescence-specific gene expression fingerprints reveal cell-type-dependent physical clustering of up-regulated chromosomal loci. Proc. Natl. Acad. Sci. U.S.A. 2003; 100:3251–3256. [PubMed: 12626749]

Zhang R, Chen W, Adams PD. Molecular dissection of formation of senescence-associated heterochromatin foci. Mol. Cell. Biol. 2007; 27:2343–2358. [PubMed: 17242207]

Zhang R, Poustovoitov MV, Ye X, Santos HA, Chen W, Daganzo SM, Erzberger JP, Serebriiskii IG, Canutescu AA, Dunbrack RL, Pehrson JR, Berger JM, Kaufman PD, Adams PD. Formation of MacroH2A-containing senescence-associated heterochromatin foci and senescence driven by ASF1a and HIRA. Dev. Cell. 2005; 8:19–30. [PubMed: 15621527]

Zhu CH, Mouly V, Cooper RN, Mamchaoui K, Bigot A, Shay JW, Di Santo JP, Butler-Browne GS, Wright WE. Cellular senescence in human myoblasts is overcome by human telomerase reverse transcriptase and cyclin-dependent kinase 4: consequences in aging muscle and therapeutic strategies for muscular dystrophies. Aging Cell. 2007; 6:515–523. [PubMed: 17559502]

Zindy F, Quelle DE, Roussel MF, Sherr CJ. Expression of the p16INK4a tumor suppressor versus other INK4 family members during mouse development and aging. Oncogene. 1997; 15:203–211. [PubMed: 9244355]