Is there a role for placental senescence in the genesis of obstetric complications and fetal growth restriction?

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The placenta ages as pregnancy advances, yet its continued function is required for a successful pregnancy outcome. Placental aging is a physiological phenomenon; however, there are some placentas that show signs of aging earlier than others. Premature placental senescence and aging are implicated in a number of adverse pregnancy outcomes, including fetal growth restriction, preeclampsia, spontaneous preterm birth, and intrauterine fetal death. Here we discuss cellular senescence, a state of terminal proliferation arrest, and how senescence is regulated. We also explore the role of physiological placental senescence and how aberrant placental senescence alters placental function, contributing to the pathophysiology of fetal growth restriction, preeclampsia, spontaneous preterm labor/birth, and unexplained fetal death.

Key words: aging, cellular senescence, cyclin-dependent kinase, DNA damage, fetal death, fetal growth restriction, mammalian target of rapamycin complex, membrane rupture, mitogen-activated protein kinase, oxidative stress, phosphoinositide 3-kinase, placental aging, preeclampsia, preterm birth, preterm labor, reactive oxygen species, senescence-associated heterochromatin foci, small for gestational age, stillbirth, telomere, tumor suppressor protein p53, p16, senescence-associated beta-galactosidase, senescence-associated secretory phenotype

Cellular senescence and aging

A key feature of aging is a progressive loss of function at the cellular, tissue, and organ level, resulting in a reduced adaptability to stress and an increased vulnerability to disease and mortality.[1] In mitotic tissues, the progressive accumulation of senescent cells is thought be one of the causal factors for aging.[2] Thus, the biomarkers of cellular senescence can be used as markers of tissue aging. Such biomarkers of cellular senescence have been summarized in a later section (see *Biomarkers of senescence*).

Senescent cells within tissues contribute to the aging process and disease development by altering normal cellular function, changing the behavior of neighboring cells, degrading structural components such as the extracellular matrix, and accelerating the loss of tissue regeneration capacity by reducing stem and progenitor cells.[2] Elimination of senescent cells can delay aging-associated disorders in mice.[3]

Cellular senescence is a state of irreversible, terminal arrest of cell proliferation (growth), triggered by a plethora of intrinsic and extrinsic stimuli or stressors. These stimuli or stressors include short or dysfunctional telomeres, DNA damage (telomeric or genomic DNA), and DNA damage-response mediators, strong mitogenic signals (eg, overexpression of oncogenic renin-angiotensin system [RAS], a mutant RAS-p21 protein, renin-angiotensin system involves transmitting signals and activating signaling cascades, including mitogen-activated protein kinase [MAPK] and phosphoinositide 3-kinase/mammalian target of rapamycin complex pathways), epigenomic disruption (chromatin disruption), overexpression of certain oncogenes, deteriorating mitochondrial function, and oxidative stress created by reactive oxygen species (ROS) (reviewed in references 4-6) (Figure 1).

The stressors that trigger senescence act by 2 major pathways controlled through stabilization of the tumor suppressor protein p53 and transcriptional inactivation of the cyclin-dependent kinases (CDKs). The suppression of CDKs is produced by transcriptional activation of the CDK inhibitor p21 (also termed $p21^{Cip1}$) in concert with the CDK inhibitor, p16 (also known as $p16^{INK4a}$) and retinoblastoma tumor suppressor protein (pRB) (reviewed in references 7 and 8) (Figure 1).

When activated, p53 inhibits cell proliferation via activation of its transcriptional target p21.[8] Both p21 and p16 maintain the protein pRB in its hypophosphorylated and active state.[8,9] Active pRb suppresses the E2F1 (a member of E2F family of transcription factors, which induce gene transcriptions that are essential for cell proliferation)-dependent expression of genes that regulate progression of the G1/S phase of the cell cycle, and thereby irreversibly blocks cell cycle entry[10]

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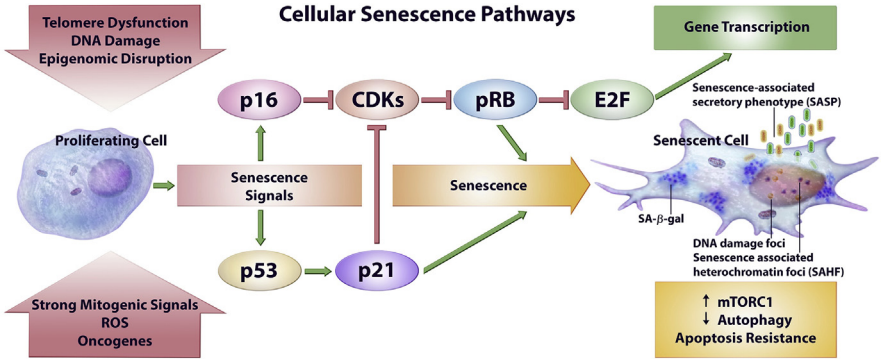
FIGURE 1
An overview of cellular senescence



Telomere-dependent replicative senescence and stress-induced premature senescence act through the modulation of proteins p53 and Rb. Senescence stimuli, such as DNA damage, strong mitogenic signals, overexpression of oncogenes, epigenomic disruption, telomere dysfunction, and ROS engage in cell signaling cascades that cause activation of 1 or both of the pathways that regulate cell senescence, the p53-p21 and p16-pRB pathways. Activation of p53 induces the expression of a CDK inhibitor, p21. Senescence stimuli, which involve the p16-pRB pathway upregulate the expression of another CDK inhibitor, p16. Both p21 and p16 suppress the phosphorylation and inactivation of pRB, and hereby maintain its hypophosphorylated and active state. Active pRB halts cell cycle progression by inhibiting gene transcription via downregulating transcription factor E2F. Senescent cells remain metabolically active, despite their terminal growth arrest, and secrete proinflammatory cytokines, chemokines, growth factors, and proteases, collectively termed the senescence-associated secretory phenotype.

CDK, cyclin-dependent kinase; *pRB*, retinoblastoma tumor suppressor protein; *Rb*, retinoblastoma; *ROS*, reactive oxygen species.

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(see Panel 1 for cell cycle). Silencing of E2F target genes is mediated by pRB-dependent reorganization of chromatin into distinct heterochromatin structures that accumulate in the nucleus of senescent cells termed senescence-associated heterochromatin foci (SAHF).[9]

Interestingly, a senescent cell can re-enter the cell cycle following inhibition of p53 if the cell senescence occurred because of activation of the p53-p21 pathway; however, cells that senesce solely via the p16-pRB pathway are unable to resume proliferation, even after the inhibition of p53, pRB, or p16.[11]

Causes of cellular senescence. A critically short telomere is thought to be one factor initiating cellular senescence. Telomeres are highly conserved repetitive DNA regions, consist of tandem arrays of the hexanucleotide sequence TTAGGG in the human, which is typically 10−15 kb long,[12] located at the end of linear chromosomes, and are essential for chromosomal stability and cell survival.[13,14]

Telomeres protect DNA ends from double-strand breaks, end-to-end fusion, and degradation by forming a protective cap with a guanine-rich single-stranded telomere overhang and telomere-binding protein complexes.[15,16] Because of an inability to replicate telomeric DNA at the ends of chromosomes (known as the end-replication problem of eukaryote DNA), telomeres are progressively shortened every time a cell divides.[2] When telomeres reach a critical minimum length, their protective structure is distorted, resulting in the exposure of DNA ends and a DNA damage response, which lead to the activation of the cellular senescence pathway.[5,14,17,18] This phenomenon is commonly known as replicative senescence. Telomere shortening is also accelerated as a consequence of environmental stressors and insults, such as hyperglycemia, hypoxia, and oxidative stress,[19-22] which lead to the oxidation of the guanosine residues. Telomere length is regulated by the enzyme telomerase, which is a specific reverse transcriptase capable of

PANEL 1 CELL CYCLE

The cell cycle or cell-proliferation cycle is a series of events that take place in a mitotic cell to produce 2 daughter cells. In eukaryotic cells, the stages of the cell cycle are divided into 2 major phases: interphase and the M phase.

Interphase: During the interphase the cell grows in size and makes a copy of the cell's DNA (called DNA replication) to prepare for the cell division. The interphase is comprised of 3 stages: G1, S, and G2.

- **G_1.** In the first gap phase, the cell increases in size, copies organelles, and makes the molecular building blocks it will need in later steps. The G_1 checkpoint control mechanism ensures that everything is ready for DNA synthesis.
- **S phase.** DNA synthesis occurs during this phase. It also duplicates a microtubule-organizing structure called the centrosome. The centrosomes help separate DNA during M phase.
- **G_2.** The cell continues to grow in the second gap phase and synthesizes proteins and organelles. During this phase microtubules begin to reorganize to form a spindle. The G_2 checkpoint control mechanism ensures that everything is ready to enter the M phase and divide.

M phase: During the M phase, cell growth stops and cellular energy is focused on the orderly division into 2 daughter cells. At this stage the cell separates its DNA into 2 sets and divides its cytoplasm, forming 2 new cells.

G_1, gap 1; *G_2*, gap 2; *M*, mitotic; *S*, synthesis phase.

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