https://doi.org/10.53886/gga.e0000240_EN

Moving from an "anti-aging" paradigm toward the concept of "disease-free aging": the role of senolytics in modern medicine

Passando do paradigma "antienvelhecimento" para o conceito de "envelhecimento antidoença": o papel dos senolíticos na medicina moderna

Ivan Aprahamian[a], **Andréia Pain**[a], **Virgílio Garcia Moreira**[b]

[a]Departamento de Clínica Médica, Divisão de Geriatria, Faculdade de Medicina – Jundiaí (SP), Brazil.
[b]Departamento de Clínica Médica, Faculdade de Ciências Médicas, Universidade Estadual do Rio de Janeiro – Rio de Janeiro (RJ), Brazil.

Correspondence data
Ivan Aprahamian – Rua Francisco Telles, 250 – CEP: 13202-550 – Jundiaí (SP). Brazil.
E-mail: ivan.aprahamian@gmail.com

Received on: Aug 31, 2024
Editor decisions on: Sep 12, 2024; Oct 15, 2024.
Accepted on: Nov 1, 2024

Handling Editor: Patrick Alexander Wachholz

How to cite this article: Aprahamian I, Pain A, Moreira VG. Moving from an "anti-aging" paradigm toward the concept of "disease-free aging": the role of senolytics in modern medicine. Geriatr Gerontol Aging. 2024;18:e0000240. https://doi.org/10.53886/gga.e0000240_EN

Abstract
Senescent cells increase with aging and are related to the promotion of inflammation and neoplasia through the senescence-associated secretory phenotype (SASP). These cells are closely related to the biological aging process and, most importantly, to age-associated diseases, such as osteoporosis and osteoarthritis. Senolytics are a new drug class that kills senescent cells by targeting senescent cell anti-apoptotic pathways (SCAPs), which are biological systems that avoid cellular destruction and host innate defenses. Senolytic therapy requires strong evidence in human trials, and dasatinib and quercetin have shown good results in phase II trials with idiopathic pulmonary fibrosis and diabetic renal disease. However, these trials are small and merely represent a proof-of-concept for these drugs. Nevertheless, this evidence calls for an overview of senescent cells and senolytics. We briefly discuss these related topics, summarizing the best evidence for clinical practitioners.
Keywords: aging; biology; senotherapeutics.

Aging is as certain as death. Likewise, aging is a major mortality risk factor due to the consequences of atherosclerosis, dementia, blindness, deafness, osteoporosis, diabetes and arthritis, among other conditions that are typically associated with an adverse pattern of senescence.[1] Beyond being a risk factor for disease and death, aging as a biological process is quite complex and incompletely understood. Although natural cellular senescence is a key player in aging, many other biological processes occur simultaneously and are of utmost importance — such as genomic instability, telomere attrition, epigenetic alterations, loss of proteostasis, deregulated nutrient sensing, mitochondrial dysfunction, cellular senescence, stem cell exhaustion, and altered intercellular communication, which have been described as the hallmarks of aging.[2] All of these pathways can promote cellular senescence (geroconversion). For example, interaction with a virus, such as a coronavirus, can induce cellular senescence in several ways, e.g., extracellular viral DNA, inflammation, or the production of reactive oxygen species.[3] Cellular senescence is characterized by the cessation of replication potential, resistance to apoptosis, altered cellular metabolism, and protein synthesis and secretion.[3] As we age, these cells accumulate in different tissues and develop a SASP, yielding systemic production of active proteins, lipids, nucleotides, and other factors that culminate in inflammation, dysfunctional cells, degenerative tissues, and cancer proliferation.[1,4] Yet, these cells have mechanisms to prevent their own death due to SASP, and these 'senescent' cells increase in number as we age, especially after 60 years of age.[1,4]

Although no specific marker of senescent cells can be described, they have several shared characteristics. They are usually large cells with high concentrations of $p16^{INK4a}$ and $p21^{CIP1}$ (proteins produced by homonymous genes), show evidence of DNA damage (particularly in telomeres) and senescence-associated distention of pericentromeric satellite DNA, and finally, increased beta-galactosidase activity.[5] A fundamental and practical question is the interplay between senescent cells and SASP. Ultimately, SASP stimulates universal hyperfunction,[6] a good illustration of which is metabolic syndrome. Senescent beta cells overproduce insulin,[7] which activates the mammalian-target of rapamycin pathway, especially in hepatocytes and adipocytes. Their hyperfunction results in hyperinsulinemia with hyperglycemia, hyperlipidemia with obesity, and finally hypertension. Thus, eliminating these hyperfunctional senescent cells may prevent certain age-related conditions.[7] However, because not all hyperfunctional cells are bad, it is fundamental to target specific senescent cells. Such therapy does not represent an effort to increase the lifespan but to live better with less multimorbidity, i.e., better quality of life.

In the last couple of years, more than 20 biotech companies have been researching and developing compounds against senescent cells, a group of drugs most known as senolytics. Senolytics kill senescent cells by targeting different SCAPs, which consist of biological systems that prevent cellular destruction or are simply conceptualized as a cellular defense (Figure 1).[1,4] There is a research trend to use senolytic fusion proteins against molecules involved in senescent activity or SCAPs.[7] However, one treatment challenge for senolytic therapy is that each senescent cell originates from a different cell type and uses different SCAPs for its defense.[1] Nevertheless, the drugs' selectivity for senescent cells is modest and they can only be restricted to certain cell types, which not only limits their action but causes collateral damage to nonsenescent, healthy cells. Currently, research into several pathways for clearing senescent cells is underway.[4] Most commonly, research groups are using a combination of at least two senolytics. For example, dasatinib (a leukemia drug that targets multiple kinases) and quercetin (a flavonoid from fruit skins) have been used successfully in rodent and human cell cultures (as well as human tissue models) and murine models of accelerated aging.[1,8,9] These agents act through a different pathway, basically against multiple weak spots in signaling that promote the survival of senescent cells found by RNA-interference screening. Dasatinib acts on dependence receptor/Src kinase/tyrosine kinase, while quercetin acts on other anti-apoptotic pathways of senescent cells, such as the Bcl-2 family, p53/p21/serpin, and PI3K/AKT. More recently, it has been found that these two agents reduce physical dysfunction in patients with idiopathic pulmonary fibrosis and reduce several biomarkers related to senescent cell pathways in patients with diabetic kidney disease.[8,9] As senescent cells show a turnover of approximately 30 days, these agents can be used on monthly basis, which is referred by research groups as "hit-and-run" therapy.[1] These short delivery protocols reduce the adverse reactions associated with a number of these drugs and compounds.

Senolytic therapy may have a major impact in the future. These agents have extended the lifespan and improved the health conditions of mice.[1] In murine models corresponding to humans of around 80 years of age, a combination of dasatinib and quercetin increased survival by 36%.[10] Other potential outcomes observed with senolytics were reduced onset of physical dysfunction, a lower incidence of chronic age-associated diseases, such as osteoporosis and cancer, geriatric syndromes such as frailty, and finally, modifying the course of neurodegenerative conditions such as Alzheimer's disease.[1] Most evidence on senolytics comes from experimental studies using murine models and human

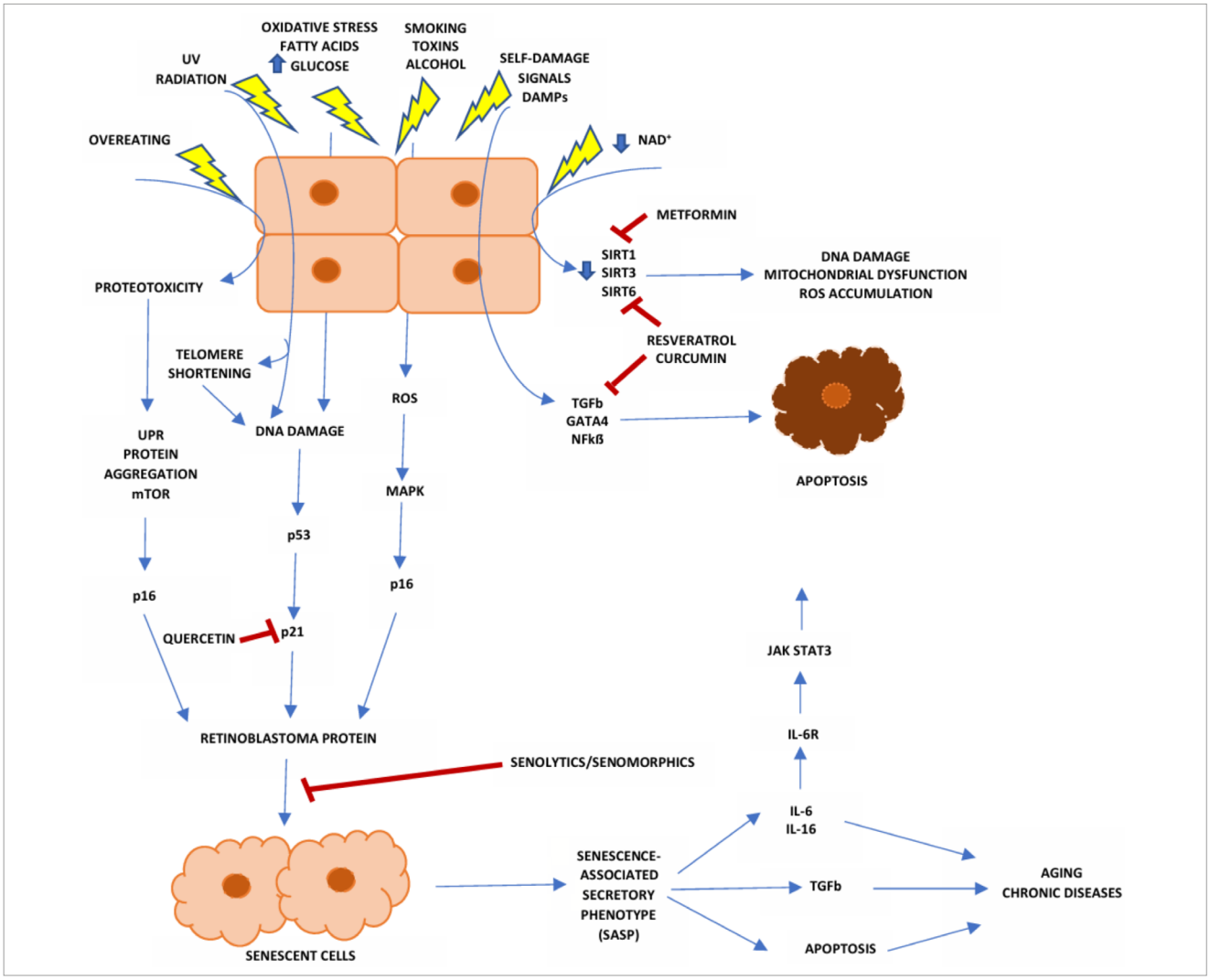

FIGURE 1. An integrated pathway to the generation of senescent cells and the main function for associated ageing and chronic diseases. Note: DAMPs, Damage-associated molecular patterns; GATA4, GATA Binding Protein 4; IL-6, interleukin-6; IL-16, interleukin-16; MAPK, mitogen-activated protein kinase; mTOR, mammalian target of rapamycin; NAD^+, nicotinamide adenine dinucleotide; NF-κB, nuclear factor kappa-light-chain enhancer of activated B cells; ROS, reactive oxygen species; SIRT, Sirtuin; STAT, signal transducer and activator of transcription; TGFß, Transforming growth factor-β.

tissue cell cultures. Moving toward therapeutic counselling in humans is untimely, even considering the safety of most agents like flavonoids (e.g., fisetin and quercetin). Clinical trials are needed that involve multiple age-associated conditions, such as degenerative conditions in the lungs, eyes, brain, heart, bones, and cancer. We hope to see results in the next couple of years. There is an ongoing multicenter trial of fisetin in older nursing facility residents who were rtPCR-positive for SARS-CoV-2.[11] A simple direct translation of research findings from mice to humans is antiscientific and dangerous. However, some senolytics are quite safe, such as flavonoids, although others are antineoplastic drugs such as navitoclax or dasatinib, which involve the risk of neutropenia and thrombocytopenia. Senolytics are not the fountain of youth, but a tool for future precision medicine, especially for individuals at higher risk of age-associated diseases like osteoarthritis or diseases that involve accelerated aging.

In summary, senolytics are real and are knocking at our door. Several ongoing experimental and clinical trials are going beyond SCAP targeting or killing senescent cells. Other pathways for quantitatively reducing senescent cells are being explored, such as immunomodulators or vaccines to accelerate the immune destruction of senescent cells, agents that deliver a toxin to senescent cells, and drugs that block the metabolic pathways of senescent cells or reduce the damage these cells cause by inhibiting the SASP. Previously approved drugs, such as metformin, rapamycin

(sirolimus), and the JAK1/2 inhibitor ruxolitinib partially inhibit the SASP. Non-pharmacological interventions like caloric restriction or exercise can also delay the development of senescent cells. Hence, senolytics should not be dismissed as modern snake oil.[12] The principal reason for using senolytics is to improve our health span and avoid or reduce the burden of age-related diseases like osteoporosis, Alzheimer's, or atherosclerosis. They are not simply anti-aging medicine, but can be viewed as an opportunity of moving from the "anti-aging" therapeutics paradigm toward the concept of "disease-free aging".

However, several questions remain, of which 2 are highly relevant:

1. how to deal with other age-associated pathways like mammalian-target of rapamycin, sirtuins, nicotinamide-adenine dinucleotide (NAD^+), or DNA methylation (Are all of these pathways equally important?), and
2. how to deal with the wide variety of tissue-specific senescent cells.[13-15]

We also don't know enough about long-term safety: most studies are still in the preclinical stages, involving cells and animal models. It will be costly and complex to develop prospective cohorts and randomized clinical trials that assess the safety of these drugs. We are looking forward to an exciting future in senolytic research.

DECLARATIONS

Conflict of interest
The authors declare no conflicts of interest.

Funding
This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors.

Author's contribution
Ivan Aprahamian: conceptualization, methodology, writing – original draft, writing – review & editing. Andréia Pain: conceptualization, methodology, writing – original draft, writing – review & editing. Virgílio Garcia Moreira: conceptualization, methodology, writing – original draft, writing – review & editing.

Ethical approval and informed consent
Not applicable.

Data availability statement
Not applicable.

Reporting standards guidelines
This paper is a viewpoint, so none of the standard guidelines apply.

REFERENCES

1. Kirkland JL, Tchkonia T. Cellular senescence: a translational perspective. EBioMedicine. 2017;21:21-8. https://doi.org/10.1016/j.ebiom.2017.04.013
2. López-Otín C, Blasco MA, Partridge L, Serrano M, Kroemer G. The hallmarks of aging. Cell. 2013;153(6):1194-217. https://doi.org/10.1016/j.cell.2013.05.039
3. Sargiacomo C, Sotgia F, Lisanti MP. COVID-19 and chronological aging: senolytics and other anti-aging drugs for the treatment or prevention of corona virus infection? Aging (Albany NY). 2020;12(8):6511-7. https://doi.org/10.18632/aging.103001
4. Martel J, Ojcius DM, Wu CY, Peng HH, Voisin L, Perfettini JL, et al. Emerging use of senolytics and senomorphics against aging and chronic diseases. Med Res Rev. 2020;40(6):2114-31. https://doi.org/10.1002/med.21702
5. Lagoumtzi SM, Chondrogianni N. Senolytics and senomorphics: natural and synthetic therapeutics in the treatment of aging and chronic diseases. Free Radic Biol Med. 2021;171:169-90. https://doi.org/10.1016/j.freeradbiomed.2021.05.003
6. van Deursen JM. Senolytic therapies for healthy longevity. Science. 2019;364(6441):636-7. https://doi.org/10.1126/science.aaw1299
7. Helman A, Klochendler A, Azazmeh N, Gabai Y, Horwitz E, Anzi S, et al. p16(Ink4a) – induced senescence of pancreatic beta cells enhances insulin secretion. Nat Med. 2016;22(4):412-20. https://doi.org/10.1038/nm.4054
8. Justice JN, Nambiar AM, Tchkonia T, LeBrasseur NK, Pascual R, Hashmi SK, et al. Senolytics in idiopathic pulmonary fibrosis: results from a first-in-human, open-label, pilot study. EBioMedicine. 2019;40:554-63. https://doi.org/10.1016/j.ebiom.2018.12.052
9. Hickson LJ, Prata LGPL, Bobart SA, Evans TK, Giorgadze N, Hashmi SK, et al. Senolytics decrease senescent cells in humans: preliminary report from a clinical trial of Dasatinib plus Quercetin in individuals with diabetic kidney disease. EBioMedicine. 2019;47:446-56. https://doi.org/10.1016/j.ebiom.2019.08.069

10. Xu M, Pirtskhalava T, Farr JN, Weigand BM, Palmer AK, Weivoda MM, et al. Senolytics improve physical function and increase lifespan in old age. Nat Med. 2018;24(8):1246-56. https://doi.org/10.1038/s41591-018-0092-9

11. Verdoorn BP, Evans TK, Hanson GJ, Zhu Y, Prata LGPL, Pignolo RJ, et al. Fisetin for COVID-19 in skilled nursing facilities: Senolytic trials in the COVID era. J Am Geriatr Soc. 2021;69(11):3023-33. https://doi.org/10:10.1111/jgs.17416

12. Morley JE. Senolytics: the modern snake oil? J Nutr Health Aging. 2019;23(6):490-3. https://doi.org/10.1007/s12603-019-1202-1

13. Grabowska W, Sikora E, Bielak-Zmijewska A. Sirtuins, a promising target in slowing down the ageing process. Biogerontology. 2017;18(4):447-76. https://doi.org/10.1007/s10522-017-9685-9

14. Covarrubias AJ, Perrone R, Grozio A, Verdin E. NAD^+ metabolism and its roles in cellular processes during ageing. Nat Rev Mol Cell Biol. 2021;22(2):119-41. https://doi.org/10.1038/s41580-020-00313-x

15. Wątroba M, Dudek I, Skoda M, Stangret A, Rzodkiewicz P, Szukiewicz D. Sirtuins, epigenetics and longevity. Ageing Res Rev. 2017;40:11-9. https://doi.org/10.1016/j.arr.2017.08.001