Invited Review / Davetli Derleme
doi: 10.18678/dtfd.1500615

Duzce Med J, 2024;26(S1):95-99
Düzce Tıp Fak Derg, 2024;26(S1):95-99

Senescence Model Theories from In Vitro through In Vivo

In Vitro'dan In Vivo'ya Yaşlılık Model Teorileri

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ABSTRACT

The theoretical equivalence of expressing that a cell is aging to its inability to perform the assumed function is not exactly accurate, it involves a gradual decrease in cell aging mechanisms. Factors such as genetics, lifestyle, and environmental effects maintain the biological change of the cell. The concept of cellular senescence was initially introduced by Hayflick and his collaborators in 1961 when they noticed that human diploid fibroblasts cultured in vitro could undergo only a limited number of cell divisions before their ability to proliferate was permanently halted. This phenomenon, known as the 'Hayflick limit', was subsequently linked to the gradual shortening of telomeres with each successive round of cell division. Throughout the aging process, senescent cells collect in different tissues. Their involvement in age-related health issues such as neurodegenerative disorders, heart problems, cancer, kidney-related changes, chronic lung diseases, and osteoarthritis suggests that targeting senescent cells therapeutically could be promising across various health conditions. This review will discuss the available data on which cell types may undergo aging based on biological aging and how these processes may impact age-associated tissue-specific pathologies. Additionally, the markers used to characterize the physiological transition of aging cells from in vitro to in vivo settings will be evaluated. The discussed data may serve as a significant starting point for an expanded definition of the molecular and functional characteristics of aging cells in different organs, thus supporting the development and enhancement of targeting strategies in vivo.
Keywords: Cell senescence; geriatric in vivo models; cellular aging theories; biological aging mechanism.

ÖZ

Bir hücrenin, varsayılan işlevini yerine getiremeyecek kadar yaşlanmasının karşılığı hücrenin biyolojik süreçlerinde her zaman tam bir yetersizliğe yol açmayabilir, hücre yaşlanma mekanizmalarında kademeli bir azalmayı içerir. Hücrenin biyolojik değişimini genetik, yaşam tarzı ve çevresel etkiler gibi faktörler sürdürür. Hücresel yaşlanma kavramı ilk olarak 1961 yılında Hayflick ve çalışma arkadaşları tarafından, in vitro kültüre alınan insan diploid fibroblastlarının çoğalma yetenekleri kalıcı olarak durdurulmadan önce yalnızca sınırlı sayıda hücre bölünmesine maruz kalabileceğini fark ettiklerinde ortaya atıldı. 'Hayflick sınırı' olarak bilinen bu fenomen, daha sonra, birbirini izleyen her hücre bölünmesi turunda telomerlerin kademeli olarak kısalması ile ilişkilendirildi. Yaşlanma süreci boyunca yaşlanan hücreler farklı dokularda toplanmaktadır. Nörodejeneratif ve kardiyovasküler bozukluklar, kanser, böbrekle ilgili değişiklikler, kronik akciğer hastalıkları ve osteoartrit gibi yaşa bağlı sağlık sorunlarına katılımları, yaşlanan hücreleri terapötik olarak hedeflemenin çeşitli sağlık koşullarında umut verici olabileceğini düşündürmektedir. Bu derlemede biyolojik yaşlanmaya bağlı olarak hangi hücre tiplerinin yaşlanabileceğine ve bu süreçlerin yaşla ilişkili dokuya özgü patolojileri nasıl etkileyebileceğine dair mevcut veriler tartışılacaktır. Ek olarak, yaşlanan hücrelerin in vitro ortamdan in vivo ortama fizyolojik geçişini karakterize etmek için kullanılan belirteçler de değerlendirilecektir. Tartışılan veriler, farklı organlarda yaşlanan hücrelerin moleküler ve fonksiyonel özelliklerinin genişletilmiş bir tanımı için önemli bir başlangıç noktası olarak hizmet edebilir, böylece in vivo hedefleme stratejilerinin geliştirilmesini ve artırılmasını destekleyebilir.
Anahtar kelimeler: Hücre yaşlanması; geriatrik in vivo modeller; hücresel yaşlanma teorileri; biyolojik yaşlanma mekanizması.

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Received / Geliş Tarihi : 18.04.2024
Accepted / Kabul Tarihi : 20.05.2024
Available Online /
Çevrimiçi Yayın Tarihi : 13.06.2024

INTRODUCTION

The rapid aging or entry into an aging trend of advanced and developing societies, interest in aging, and old age studies have started to increase in recent years. This has led to a growing interest in the disciplines of gerontology and geriatrics, which encompass studies in the field. According to a report by the United Nations (UN) in 2022, approximately 10% of the world's population consists of older adults aged 65 and above. This percentage is expected to reach 16% by the year 2050 (1). Looking at Turkey, it is observed that the proportion of the elderly population has surpassed the world average, reaching 10.2% of the total population (2). This situation can lead to a concentrated focus on popular areas such as social, psychological, health, and care in aging and old age studies, potentially neglecting biogerontology. Biogerontology is a discipline that investigates why and how living organisms age (3). In the light of current knowledge, aging refers to the period from an individual's existence as an organism to death, while old age represents a unique stage of life like childhood and youth, and an older adult refers to a person above a certain age. The chronological age that distinguishes these categories is 60 according to the UN and 65 according to the World Health Organization (WHO). Therefore, it is understood that social conditions and environmental factors change the biological aging process, as the aging process is categorized differently. It is possible to build a society of ageless elderly individuals by optimizing external factors while the biological aging process is in progress (4). Based on all this information, biogerontology seeks answers to the following questions:

- Many aging models have been proposed to explain the aging process, in other words, why do we age?
- What are the biological processes associated with aging?
- Is aging genetically programmed or is it a multivariate random process?
- Are there biomarkers associated with aging?
- Is it possible to slow down or prevent aging? What chronic diseases occur with aging?

Cellular senescence is the result of a series of molecular and cellular changes that cells undergo throughout life (5). These changes can disrupt the function of cells and contribute to a number of diseases associated with aging (Figure 1). Throughout its historical development, many biological theories of aging have been proposed seeking answers to the causes of aging. These theories are discussed under two main headings: evolutionary theories of aging and physiological (mechanistic) theories of aging. While evolutionary theories of aging focus on the ultimate cause, mechanistic theories of aging focus on the convergent or apparent cause. According to stochastic theory, aging occurs as a result of the accumulation of randomly occurring errors in biomolecules. It is suggested that the accumulation of mutations in the genetic material of the cell due to external and internal factors over time and the advanced glycation products formed as a result of the glycation of biomolecules cause aging. According to the hereditary model, it is accepted that aging is a programmed process. The main cause of replicative senescence, defined as replicative cells losing their ability to divide after a certain number of divisions, is telomere shortening (6-9). Numerous processes associated with aging have been identified. These include genomic instability, epigenetic variations, and changes at the transcriptional level, as well as molecular damage, cellular aging and death, inflammation, and metabolic disorders (10,11). Although almost every aging model focuses on a single mechanism related to aging, it does not seem possible to explain it with a single mechanism since aging is a very complex event. This review aims to discuss the available data on which cell types may undergo aging based on biological aging and how these processes may impact age-associated tissue-specific pathologies.

CELLULAR PROCESSES ASSOCIATED WITH AGING

Telomere Shortening

Telomeres are repetitive DNA sequences located at the ends of chromosomes. With each cell division, telomeres shorten. The shortening of telomeres causes cells to lose their ability to divide and their genetic integrity, contributing to the cellular aging process (12).

Genetic Damage

Normally, the DNA molecule is stable and DNA replication is a very conservative process that tries to operate without errors. DNA damage has a significant impact on the aging of cells. Several threats such as ionizing radiation, oxidative stress, chemicals, and other environmental factors can cause DNA damage. With aging, oxidation in mitochondrial DNA (mtDNA) is much more prominent than in nuclear DNA. A range of genetic deteriorations can occur, from point mutation, which is the simplest form of mutation, to chromosome losses and gains. Complex DNA repair mechanisms have evolved to reduce these damages and keep the genome stable. However, these mutations, which are still rare, accumulate over time (age) and the genome becomes unstable. The accumulation of this damage is associated with the aging of cells and the development of aging-related diseases.

Oxidative Stress

Among the types of stress known to stimulate or accelerate cellular aging, oxidative stress is the accumulation of free radicals and other reactive oxygen species in cells. Oxidative stress can lead to a number of cellular damage

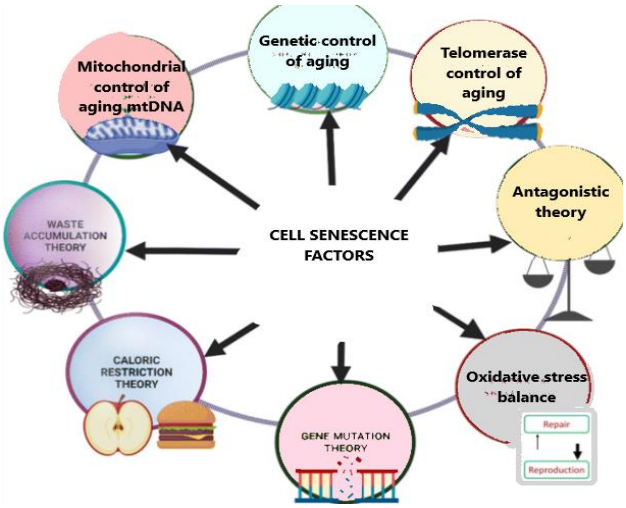

Figure 1. Variety arms of change cell structure

in cells, such as lipid peroxidation, protein damage, and DNA damage. This environment can lead to stress-induced cellular senescence (SIPS), which pushes cells to early aging with the effect of other molecular factors (13,14).

DNA Repair Mechanism Balance

They are segmental early aging (progeria) syndromes that occur as a result of defects in the DNA repair mechanism. It is characterized by defects in DNA helicase and DNA repair mechanisms as a result of mutation in the WRN gene. Lamin A production as a result of LMNA gene mutation. It has been determined that the rarity of the apolipoprotein E ε4 allele triggers cellular aging. It is characterized by disorder. Progeria syndromes shed light on some aspects of normal aging, but they do not include all the features of normal aging (15,16).

Cell Cycle Deregulation

The cell cycle is the process of division and proliferation of cells. Disturbances in the regulation of the cell cycle can prevent or accelerate the controlled growth and division of cells. This can contribute to the cellular aging process and the development of diseases such as cancer (17).

Disruption of Protein Homeostasis (Proteostasis)

Cells maintain protein homeostasis by ensuring the correct folding, processing, and degradation of proteins. With aging, the effectiveness of these mechanisms may decrease and problems such as protein misfolding and accumulation may occur (18,19).

Mitochondrial Dysfunction

Mitochondria are known as energy-producing structures in cells. Mitochondrial dysfunction can cause problems such as oxidative phosphorylation disorder, reactive oxygen species production, and decreased cellular energy production. This can contribute to the aging of cells (20).

Inflammation

Inflammation is increasingly linked to aging and chronic diseases. However, the observed increase in the basal inflammatory response with age causes a sustained low level of inflammation that promotes aging. The decline of the thymus gland, where T cells mature, is a much faster process than aging. Although proliferation decreases in the elderly, the number of T cells generally does not change. Apoptosis occurs with decreasing bcl-2 expression in maturing T cells. B cells also produce fewer antibodies. Senescent cells secrete many cytokines that initiate inflammation. In fact, in advanced ages, there are a small number of senescent cells in organs and tissues, but the inflammation-initiating factors secreted by them affect the functioning of autocrine, paracrine, and endocrine systems, also known as cellular signal transmission mechanisms (21,22).

Apoptosis

Elimination of damaged cells is also known as programmed cell death or apoptosis. The increase in nondividing cells with age has led to the acceptance that it contributes to aging. However, this mechanism can also be seen as an adaptive feature, as it prevents the proliferation of damaged cells and ensures their elimination by the immune system. In this way, it prevents the tissue from being damaged and turning into potentially cancerous cells. However, age-related decline in cell renewal and immune system capacity leads to an increase in the number of senescent cells. This increase contributes to loss of function in tissues and organs and eventually to aging (23,24).

These mechanisms indicate that cellular aging is a complex process and plays an important role in the development of aging-related diseases. Researchers are trying to understand these mechanisms and identify potential therapeutic targets to delay or prevent cellular aging.

Cell senescence and aging mechanisms are the result of a series of complex interactions that biological systems undergo throughout life. Many experimental animal models have been used to understand these mechanisms (25).

IN VIVO (EXPERIMENTAL MODEL) PROCESSES ASSOCIATED WITH AGING

Yeast (*Saccharomyces cerevisiae*)

Yeast is a simple eukaryotic organism and is a fundamental model organism in studies of cell aging. It is especially used to study mechanisms associated with aging, such as telomere length and DNA damage repair. In *S. cerevisiae* yeast, it has been shown that an increase in the copy of the Ras2 gene, mutations in the Ras1 gene, a decrease in the TOR signal, an increase in the AMPK signal, and the sirtuin gene called sir2 prolong lifespan (26,27).

Nematode (*Caenorhabditis elegans*)

Nematodes are a model organism frequently used in genetic and neurological research. In particular, it is a popular option for studying the genetic mechanisms of the aging process and the neurological effects of aging. Decrease in TOR signaling in *C. elegans* worm, It has been determined that an increase in AMPK signaling and the sirtuin gene called SIR-2.1 prolong lifespan (28).

Fruit Fly (*Drosophila melanogaster*)

Fruit flies are another model organism commonly used in aging research. Telomeres are often used to study aging mechanisms such as oxidative stress and metabolic effects. It has been determined that a decrease in the TOR signal in the *D. melanogaster* fruit fly, mutations in the Indy gene, which is important in the Krebs cycle, and the Methuselah gene, which encodes a G protein-related receptor, and the sirtuin gene called sir2, are associated with longevity. There is abundant evidence regarding the importance of diet and environmental temperature in the context of aging. In 1929, it was demonstrated that the lifespan of Drosophila species is inversely proportional to ambient temperature (29).

It has been shown in many studies conducted in yeast, worms, fruit flies, and rodents that calorie restriction at a level that does not cause malnutrition extends lifespan. As a result of calorie restriction, many changes have been observed in metabolism, cellular level, genetic structure, and neuroendocrine system. When metabolism slows down, ROU production, which is thought to play an important role in aging, also decreases (30).

Mammalian Cell Cultures

Mammalian models such as mice and mouse cell cultures are widely used in research on human aging. These models have biochemical and genetic properties similar to human cells and are used specifically to study mechanisms such as cellular aging, DNA damage repair, and apoptosis (31-33).

Aging Mouse Models

Some mouse strains show physiological and pathological characteristics similar to human aging when exposed to the

natural aging process (Figure 2). These mice are valuable models used to study aging mechanisms and aging-related diseases (34). These experimental animal models provide fundamental information on cell senescence and aging mechanisms and may help identify potential therapeutic targets for understanding and treating aging-related diseases. However, these models as well as research in humans are vital for a full understanding of human aging.
Aging mouse models are laboratory mice used to understand human aging and aging-related diseases. These mice show physiological and pathological characteristics similar to human aging when exposed to the natural aging process. More commonly used aging mouse models to understand aging-associated phenotypes and the mechanisms underlying the aging process will be described (35).

Senescence Accelerated Mouse (SAM)

SAM mice are a mouse model bred at Shizuoka University in Japan and are frequently used in aging research. SAM mice have a genetic predisposition to accelerate the normal aging process and display a number of aging-associated phenotypes. SAM mice constitute an important model for studying molecular and cellular mechanisms in the aging process. SAM mice are divided into two subtypes, senescence-accelerated prone mouse (SAMP) and senescence-accelerated resistant mouse (SAMR), depending on their genetic diversity. SAMP mice are more prone to signs of aging and diseases associated with aging, while SAMR mice are more resistant. Levels of CD4 and CD8 memory T cells and naïve T cells have been used to give good estimates of life expectancy of middle-aged mice. Differences between these two subtypes are examined to understand the genetic factors underlying the aging process and the mechanisms of aging-related pathologies (36,37).

Akkerman Health Aging Mice (*Akkermansia muciniphila*)

A bacterium called *Akkermansia muciniphila* plays an important role in the gut microbiota and has been associated with the aging process. Aging mouse models have been used to study the effects of this bacterium on mice. The effects of *Akkermansia muciniphila* on the aging process are studied to investigate issues such as changes in gut health and metabolic disorders associated with aging (38).

Transgenic Mouse

In a transgenic mouse model, without implementing calorie restriction, researchers achieved a prolonged median lifespan of 20% by consistently lowering body temperature by an average of 0.5-0.6°C through modulation of the hypothalamic 'central thermostat' over an extended period (39).

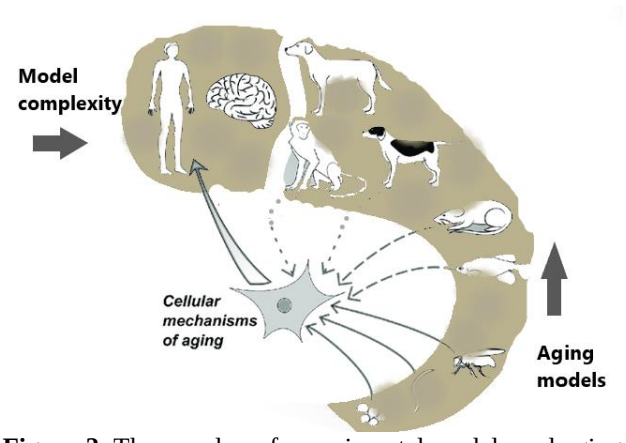

Figure 2. The paradox of experimental models and aging biology

CONCLUSION

Aging is a natural process and a natural part of life. However, it is a major risk factor for many diseases and can limit life expectancy. Interventions in model organisms have prevented or postponed the emergence of many chronic diseases as well as prolonging life. These interventions offer a promising approach to reducing aging-related health problems. Although our knowledge of aging and senescent cell types depicted in in vivo models and their roles in aging has increased significantly, the experimental models used to determine the molecular relationships of senescent cells in human tissues must be diversified and a comprehensive analysis of their roles in the aging phase is needed.

Ethics Committee Approval: Since our study was a review, ethics committee approval was not required.

Conflict of Interest: None declared by the authors.

Financial Disclosure: None declared by the authors.

Acknowledgments: None declared by the authors.

Author Contributions: Idea/Concept: MA; Design: MA; Data Collection/Processing: MA; Analysis/Interpretation: MA; Literature Review: MA; Drafting/Writing: MA; Critical Review: MA.

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