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Perspective

How Does Proliferative Homeostasis Change With Age? What Causes It and How Does It Contribute to Aging?

Judith Campisi^{1,2} and John Sedivy³

¹Life Sciences Division, Lawrence Berkeley National Laboratory, Berkeley, California.

²Buck Institute for Age Research, Novato, California.

³Department of Molecular and Cell Biology and Biochemistry, Brown University, Providence, Rhode Island.

The notion that there might be a cellular basis for aging stems from research that began several decades ago and was proposed to explain the loss of proliferative homeostasis, which is a hallmark of complex animals. Recent years have seen growing support for the idea that two cell fates—apoptosis and cellular senescence, both now well-established tumor suppressor mechanisms—may be important drivers of aging phenotypes and age-related disease. However, there remain many unanswered questions, some quite basic, about how these processes change with age and how they might contribute to aging. It is now clear that failures in apoptosis or senescence can result in hyperproliferative diseases such as cancer. Less is known about whether and how increased apoptosis or senescence can cause tissue degeneration and aging. In addition, there is now a growing recognition that cellular senescence can have cell-nonautonomous effects within tissues. New molecular tools and model organisms, some already on the horizon, will need to be developed to better understand the roles of apoptosis and cellular senescence in age-associated changes in proliferative homeostasis.

Key Words: Proliferative homeostasis—Aging.

AN OLD HYPOTHESIS—STILL ALIVE!

Nearly five decades ago, the finding that normal human somatic cells cannot proliferate indefinitely in culture (1) spawned the idea that some aging phenotypes might be due to intrinsic cellular processes. Currently, two such processes, or cell fates, are thought to contribute to the decline in tissue homeostasis that is a hallmark of mammalian aging: cell death or apoptosis, and cellular senescence or irreversible loss of cell division potential. Both are now well established as crucial mechanisms for eliminating or preventing the proliferation of damaged or dysfunctional cells, primarily for the purpose of suppressing cancer (2,3). Whether these processes play important roles in other mammalian aging phenotypes is more tenuous. Nonetheless, there is growing evidence to support this idea, and so it remains an important hypothesis in need of critical testing.

This session focused on the role of apoptosis and cellular senescence in the decline in proliferative homeostasis that occurs during mammalian aging. Aging mammalian tissues can experience two contrasting defects in proliferative homeostasis: hypoproliferation, which compromises tissue regeneration and repair, and hyperproliferation, which can lead to lethal cancers. Additionally, both hypo- and hyperproliferation can disrupt normal tissue architecture and hence tissue function. If apoptosis and cellular senescence prove important for age-related declines in tissue

function, as well as aging-associated pathology, they may be examples of evolutionary antagonistic pleiotropy (4).

It is now clear that proliferative homeostasis declines during mammalian aging, but many questions remain regarding the roles played by apoptosis and senescence in this decline, whether and how apoptotic and senescence responses change with age, and in which tissues and contexts these cell fates are beneficial and in which they are detrimental. The answers to these questions will be important not only for understanding why and how mammalian organisms age but also for developing rational strategies for mitigating or postponing aging in a variety of aging phenotypes. We summarize in the following what is known, and more importantly unknown, about the role of apoptosis and senescence in the impaired function of aging tissues and identify important areas for further study.

ARE THERE AGE-RELATED CHANGES IN APOPTOSIS AND DO THEY CONTRIBUTE TO AGING?

A number of studies have examined the incidence of apoptotic cells in rodent and human tissues, and both increases and declines have been reported. In general, increased apoptosis is associated with tissue degeneration, particularly in the nervous system (5), whereas decreased apoptosis is associated with cancer (3). However, these

generalities obscure important complexities, which need to be addressed by further research.

First, only a few mammalian tissues have been carefully examined thus far, and the nature of age-related changes in apoptosis may be highly cell type specific or tissue specific. It will therefore be important to determine in which cell types, in which direction, and to what extent, the aging of major organ systems is accompanied by changes in apoptosis.

Second, distinctions need to be made between basal and stress- or damage-induced rates of apoptosis. Which of these changes with age; in which types of cells, tissues, and organs; and in which direction? And, in the case of stress- or damage-induced apoptosis, does the age-related change depend on the nature of the stress or damage?

Third, and perhaps most importantly, the impact of age-related changes in apoptosis needs to be critically assessed by selectively modifying apoptotic responses *in vivo*. Genetic or pharmacological interventions would be the most likely avenues. Such studies will be crucial for advancing the field from correlations to causality. Two important questions are as follows: (a) do the age-related changes in apoptosis cause aging phenotypes (ie, are they deleterious?) or do they compensate for the loss of tissue homeostasis (ie, are they adaptive?) and (b) does the answer to this question depend on the tissue? For example, an age-related decline in genotoxin-induced apoptosis has been documented in the rodent liver (6). Does this decline help preserve liver function in the face of age-associated loss of liver regenerative capacity (7)? Or does this decline exacerbate age-related deterioration, as suggested by the effects of caspase-2 deficiency in the livers of genetically manipulated mice (8)? Or does the decline contribute to the increased incidence of cancer that is a common feature of aging in renewable tissues (9)?

ARE THERE AGE-RELATED CHANGES IN CELLULAR SENESCENCE AND DO THEY CONTRIBUTE TO AGING?

Several studies have examined aging in mammalian tissues for the presence of senescent cells. In general, the number of senescent cells increases with age in renewable tissues (10). However, again, only a few tissues have been examined, and estimates of how many senescent cells are present in aged tissues vary widely. Moreover, there is no single marker that universally or unequivocally identifies senescent cells. Thus, further studies are warranted in which multiple tissues are examined for the presence of senescent cells, using multiple senescence markers.

The senescence response is more complicated than the apoptotic response. First, senescent cells increase in number with age, suggesting they accumulate or persist *in vivo*. This is in contrast to apoptotic cells, which rapidly disappear from tissues. Second, because organ size does not in general increase with age, it is unlikely that senescent cells are replaced by proliferation-competent cells. Third, senescent cells upregulate many genes encoding secreted factors,

which have the potential to disrupt the integrity and function of normal tissues, and promote malignant phenotypes in neighboring cells (11). This finding raises the possibility that senescent cells affect tissues by both cell-autonomous and cell-nonautonomous mechanisms, as discussed in the following.

Mouse models in which the senescence response has been eliminated by genetic manipulation, as well as limited human clinical data, support the idea that the senescence response is crucial for preventing cancer (2,12,13). But is this response really antagonistically pleiotropic? That is, to what extent, if any, do senescent cells contribute to aging phenotypes? Support for the idea that senescent cells can drive aging phenotypes comes from cell culture studies, which indicate that factors secreted by senescent cells can disrupt normal tissue architecture and create local inflammation, an important component of many age-related pathologies (11). Support also comes from genetically modified mouse models in which defective DNA repair or inappropriately heightened tumor suppressor activity causes excessive cellular senescence—in many cases, these mice show multiple signs of premature aging (14–16, see also article by Campisi & Vijg, [17]). These findings are promising but suffer from the fact that senescence is generally induced from early ages in many cells of most tissues. However, senescent cells might have different effects depending on the tissue or age of the animal. Thus, better animal models are needed in which senescence can be induced with spatiotemporal control and in only a limited number of cells in a given tissue. The goal of these models would be to more accurately reflect what appears to occur during natural aging. More importantly, animal models are needed in which it is possible to eliminate senescent cells or blunt their cell-nonautonomous effects. Only through these new approaches will it be possible to critically evaluate whether senescent cells contribute to aging phenotypes and diseases.

HOW OR WHY DO APOPTOTIC AND SENESCENT CELLS ARISE DURING AGING?

There are several important unanswered questions concerning the proximal causes for the changes in apoptosis and cellular senescence that occur during aging. Do changes in apoptosis rates result from increased stochastic damage, intrinsic alterations in specific cells or tissues (eg, genomic mutations or epigenetic drift), or systemic changes that affect many cell types and tissues (eg, an altered hormonal milieu)? For both apoptosis and senescence, what types of damage or stresses trigger these cell fates, and is the damage, stress, or cell fate response cell type specific or tissue specific? And in the case of cellular senescence, are all senescent states, regardless of the inducer, similar (eg, do all inducers result in a similar secretory profile)? Answers to these questions will likely require carefully conceived approaches that take advantage of both cell culture and animal models.

ARE THE EFFECTS OF SENESCENT CELLS AUTONOMOUS OR NON-AUTONOMOUS?

Senescent cells might promote aging phenotypes because their growth impairment limits the number of cells that can participate in tissue repair and regeneration. Alternatively, senescent cells might drive aging phenotypes nonautonomously by virtue of the inflammatory cytokines, growth factors, and matrix-degrading proteases they secrete. This secretory phenotype has the potential to alter local tissue microenvironments or even the systemic milieu. Which of these attributes is more important for the proaging effects of senescent cells, or are they both important? Evidence has been reported that senescent cells can be eliminated by the immune system, at least under some circumstances (18,19). If so, do senescent cells accumulate with age due to an increased frequency of generation, a decline in immune surveillance, or both? Is it possible that some senescent cells are intrinsically resistant to immune recognition or clearance? Answers to these questions will require a better understanding of how the secretory phenotype is induced and regulated, and the mechanisms by which senescent cells are cleared in vivo. An important aspect of this research should be the development of strategies for improving the elimination of senescent cells and suppressing the secretory phenotype.

ARE THE EFFECTS OF SENESCENT CELLS ALWAYS DETRIMENTAL?

Just as apoptosis might be either beneficial or detrimental depending on the physiological context, the possibility that the senescence-associated secretory phenotype might have beneficial effects should be considered. In many respects, this phenotype resembles a wounding or short-term inflammatory response. One possibility, then, is that the senescence secretory phenotype might have evolved to signal tissue damage, alerting neighboring cells to the damage, stimulating tissue remodeling, and ultimately stimulating the immune system to terminate the tissue damage response. Recently, support for this idea was obtained from an animal model of acute liver damage, where the ability of cells to undergo senescence was important for limiting the extent of fibrosis (19). It will be important to determine whether senescent cells protect against fibrosis—and other sequelae of stress or damage—in other tissues and physiological contexts.

STEM CELL FATES AND AGING

Little is known about how age-related damage and stress affect the fate of adult stem or progenitor cells. Whether such cells are poised to undergo apoptosis, senescence, or both is likely to be tissue and context specific, but at present, our knowledge of the extent to which different tissues are replenished by stem or progenitor cells is still in its infancy. Likewise, very little is known about the cellular and molecular components of stem cell niches, how the niche changes dur-

ing aging, and whether senescent stem or support cells alters the niche. These gaps in our knowledge are more thoroughly explored in the separate session devoted to stem cells and aging (see also article by Sharpless & Schatten, this [20]).

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CORRESPONDENCE

Address correspondence to Judith Campisi, PhD, Life Sciences Division, Lawrence Berkeley National Laboratory, 1 Cyclotron Road, Berkeley, CA 94720. Email: jcampisi@buckinstitute.org.

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