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Program-Like Aging and Mitochondria: Instead of Random Damage by Free Radicals

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Abstract As recently suggested, the target of rapamycin (TOR) pathway, rather than molecular damage by free radicals, drives aging and diseases of aging. But may mitochondria nevertheless contribute to aging? Here, I discuss aimless program-like aging (versus altruistic program), conflict between the cell and mitochondria, cell murder (versus cell suicide) and the role of mitochondria in aging. In particular, life-long selection among mitochondria may yield ''selfish'' (malignant) mitochondria resistant to autophagy. And TOR may create an intra-cellular environment that is permissive for such selfish mitochondria. In theory, pharmacologic inhibitors of the TOR pathway may reverse accumulation of defective mitochondria, while also inhibiting the aging process. J. Cell. Biochem. 102: 1389–1399, 2007. © 2007 Wiley-Liss, Inc.

Key words: cell cycle; aging; mitochondria

There are two views on the aging process. First, aging has no purpose (non-programmed) and results from random molecular damage mainly caused by mitochondrial free radicals. Second, aging is an altruistic genetic program that is executed by the same mitochondrial free radicals. Here, we will discuss an alternative view that aging is neither altruistically programmed nor a random damage but a quasi-program, a purposeless continuation of developmental programs. And this quasi-program may exaggerate the conflict between cell and mitochondria.

THE MITOCHONDRIAL CONFLICT

"Cooperation is possible wherever interests coincide. But interests are seldom identical. So cooperation gives rise to potential conflict. When conflict arises within a game of cooperation it arises at the margins, for that is where interests diverge. Even small margins can generate heated disagreement. Thus, wherever we see cooperation we should be prepared also to see conflict [Cronin, 2006]."

Mitochondria are former free-living eubacteria. Ancient invasions by eubacteria more than a billion years ago initiated the evolution of the eukaryotic cell [Dyall et al., 2004]. Mitochondria have their own genomes. Although "interests" of mitochondrial and nuclear genes strongly coincide, they are not identical. In sexual reproduction, mitochondria go to the next generation only via eggs, not via sperm; so they propagate through the female line. On the other hand, mitochondria can reproduce independently from nuclear genes. When cells do not proliferate, mitochondria may "wish" to replicate and to grow further. Thus, "the ancient arrangement with mitochondria was a recipe for conflict [Cronin, 2006]."

Let us consider the unicellular organism *S. cerevisiae*. Yeast undergo replicative aging, meaning that each cell can form a limited number of daughter cells. With each division, the mother cell becomes larger [Zadrag et al., 2006]. In asymmetric division, daughter and mother cells receive an entire number of nuclear genes. Yet, a smaller cell receives less mitochondria (and less mitochondrial genes), whereas the mother cell accumulates mitochondria. A large yeast cell with high mitochondrial DNA (mtDNA) content is destined to die, thus discarding mitochondrial genes. Interestingly,

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multicellular organisms discard mitochondria, selectively destructing male mitochondria in the fertilized oocytes [Sutovsky et al., 1999].

Perhaps, mitochondrial ancestors invaded the ancestral cells [Dyall et al., 2004]. Or maybe ancestral host cells predated on eubacteria, which in turn fought back. Maybe pre-mitochondria and their host cells actually attacked one another [Esteve and Gaju, 1999; Dyall et al., 2004; Kutschera and Niklas, 2005]. In fact, apoptotic processes spread from the cell to mitochondria and back [Blackstone and Green, 1999; Green and Evan, 2002; Ravagnan et al., 2002; van Loo et al., 2002; Jin and El-Deiry, 2005]. For example, cellular proteins Bid and Bax cause mitochondrial membrane depolymerization (the host attacks). In return, mitochondria releases cyctochrome C, activating cellular caspases, as well as apoptosis-inducing factor-1 (AIF-1) and endonuclease G, which degrades nuclear DNA (mitochondrial counter-attack) (Fig. 1). The apoptotic process can be viewed as an "exchange of blows" between the cell and mitochondria (Fig. 1). When cells are induced to undergo apoptosis in the presence of caspase inhibitors, cells may survive while mitochondria disappear from the cells [Tolkovsky et al., 2002].

After a billion years of evolution, almost perfect symbiosis between cells and mitochondria has evolved. The ability of mitochondria to initiate cell killing is now used productively to eliminate unwanted and/or damaged cells for the benefit of the multicellular organism [Blackstone and Green, 1999]. Yet the apoptotic potential of mitochondria can be exploited by "hostile creates." For example, microbes subvert mitochondria-dependent apoptosis to destroy microbe-specific T-lymphocytes [Jendro et al., 2002; Schnaith et al., 2006]. Mitochondria-dependent apoptosis is not necessarily beneficial to the multicellular organism. For example, apoptosis is responsible for damaging effects caused by radiation and anti-cancer drugs in mammals [Komarova et al., 2004; Christophorou et al., 2006; Strom et al., 2006]. Also, acidosis cause mitochondria-dependent apoptosis [Kubasiak et al., 2002]. In fact, yeast undergo mitochondria-dependent apoptosis caused by harsh conditions such as acidosis [Fabrizio et al., 2004; Madeo et al., 2004; Pozniakovsky et al., 2005; Tal et al., 2007]. When conditions are harsh, mitochondria may atavistically attack the yeast cell in a futile attempt to escape. This scenario is not utterly allegorical. Even in human cells, mitochondria are more dynamic than previously considered: mitochondria or mtDNA can move between cells [Spees et al., 2006]. Thus, in theory, mitochondria-induced cell death may be a manifestation of the conflict between the cell and mitochondria.

CELL SUICIDE OR CELL MURDER

It is often assumed that apoptosis is cell suicide. Yet, it may be not only a suicide but also a murder. The mitochondrial deadly potentialcan be manipulated and abused by other organisms. Plants, marine animals, and microorganisms produce poisons that activate mitochondria-dependent apoptotic pathways, causing death

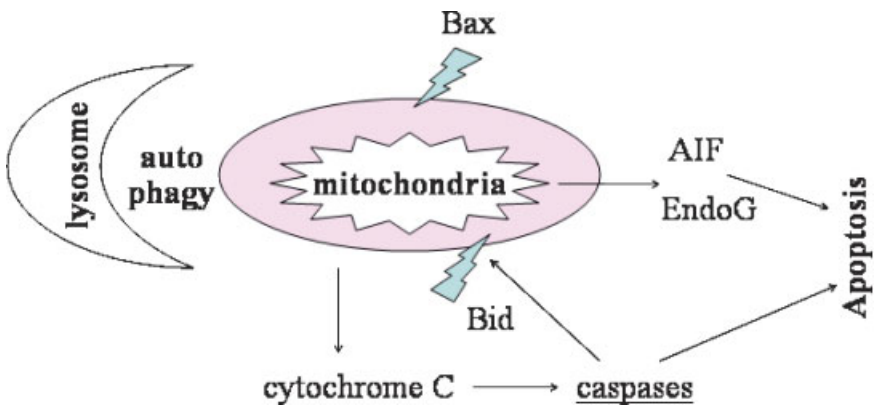

Fig. 1. Mitochondria-dependent cell death. Apoptosis could be depicted as an exchange of blows between mitochondria and the cell. Pro-apoptotic proteins Bid and Bax get inserted in mitochondrial outer membrane. Mitochondria release cytochrome C, which activates caspases, AIF, and endonuclease G. All together cause apoptosis. Caspase-dependent cascade also damage mitochondria. In addition, cellular lysosomes eat mitochondria (autophagy). [Color figure can be viewed in the online issue, which is available at www.interscience.wiley.com.]

of their competitors and predators [Blagosklonny, 2005]. Many of natural poisons such as paclitaxel and doxorubicin are used as anticancer drugs [Blagosklonny, 2005]. Obviously, cancer cells do not commit altruistic suicide in order to benefit the plant that produces paclitaxel for instance. Cells contain apoptotic pathways for other purposes. In mammalian cells, caspases have many non-death functions from survival to proliferation [Newton and Strasser, 2003]. In yeast, caspases may not be intended to cause cell suicide at all. In a simple analogy, an electric toaster may set a home on fire but was not intended for that purpose.

The plant antifungal antibiotic osmotin induces apoptosis in yeast [Narasimhan et al., 2001]. Definitely, this is not an example of altruistic yeast suicide. Similarly, in growth limiting conditions, yeast produces high levels of pheromones, which can induce apoptosis by engaging mitochondria [Pozniakovsky et al., 2005] (this is not much different from mitochondria-dependent apoptosis caused by antibiotic, for instance). After 90–99% of the yeast die, a small apoptosis-resistant subpopulation survives and regrows [Fabrizio et al., 2004]. Importantly, yeast ATP-binding cassette (ABC) proteins pump out pheromones. These ABC-proteins also pump exogenous pheromones and toxins [Jungwirth and Kuchler, 2006]. Therefore, yeast that secrete pheromones may be simultaneously resistant to pheromones, thus killing non-secreting yeast, without killing themselves.

CAN AGING BE ALTRUISTICALLY PROGRAMMED?

To explain the origin of programmed aging, it was suggested that the population can benefits from the death of individuals [Longo et al., 2005]. The strongest argument for "altruistic" programmed aging is that unicellular organisms such as yeast undergo aging and programmed cell death [Skulachev, 2002; Longo et al., 2005; Pozniakovsky et al., 2005; Skulachev and Longo, 2005]. For example, in growth-limiting conditions most yeast ("altruistic yeast") die, while a few apoptosis-resistant ("selfish") yeast survive and regrow [Fabrizio et al., 2004]. In the absence of "altruistic" yeast, the entire population went extinct [Herker et al., 2004]. So populations with "altruistic" members (members who undergo apoptosis) regrow, while populations lacking such members go extinct [Longo et al., 2005]. If all yeast were altruistic, the population would go extinct too. Nevertheless, death of most members can prevent extinction of the population in certain conditions [Skulachev, 2002; Skulachev and Longo, 2005]. However, it is selfish (resistant to apoptosis) yeast that actually survived [Fabrizio et al., 2004]. So even if altruism may prevent the extinction of the population, only selfish yeast will survive. As a related example, somatic cells in the multicellular organism must be utterly altruistic. Only by keeping the organism alive, somatic cells can pass their genes via germ linage. Yet, even somatic cells can "cheat" to acquire unrestricted proliferation and immortality (cancer). Such a selfish cell can kill the organism and terminate itself. Second, yeast aging and yeast death may not be an altruistic suicide but rather a murder. In order to survive crowded conditions, selfish yeast secrete pheromones to activate mitochondria-dependent apoptotic pathways in other yeast. As we discussed, yeast that produce more pheromones could be more resistant to pheromones. Thus, such resistant yeast can kill other cells. Third, mitochondria may initiate atavistic killing of their host cell in harsh conditions. This is consistent with the view that aging as a mitochondria-mediated atavistic program [Skulachev and Longo, 2005]. Therefore, there are several explanations why yeast die. Altruistic motives are difficult to prove, if other explanations are possible.

Still, altruistic aging is an attractive concept because it implies a program, which could be switched off, thus extending our life span. However, it does not specify the program. Instead, it focuses on the execution phase, namely on mitochondria, which produce free radicals or reactive oxygen species (ROS). Remarkably, non-program theories of aging also suggest that aging is simply a decline due to random damage caused by ROS.

COULD FREE RADICALS CAUSE AGING?

There is no compelling evidence that symptoms of aging can be attributed to free radicals. Life span can be extended dramatically without reducing ROS levels indicating that ROS does not limit life span [Seto et al., 1990; Van Remmen et al., 2003; Parker et al., 2004; Andziak et al., 2005, 2006; Buffenstein, 2005;

Corona et al., 2005]. Antioxidants failed numerous clinical trials [Howes, 2006; Bjelakovic et al., 2007]. DNA damage in adult cells is so insignificant that normal mice have been cloned from adult skin cells [Li et al., 2007]. Data that support the ROS theory have alternative explanations, supporting the target of rapamycin (TOR) theory of aging (Blagosklonny, Aging: ROS or TOR, in preparation). Even if ROS-induced damage may eventually kill organism, this does not mean that ROS actually kills it, because the organism may first die from another cause. To be precise, humans and laboratory animals die from uniform age-related diseases (e.g., atherosclerosis, neurodegeneration, cancer, diabetes, hypertension, and organ failure) that are manifestations of the aging process. TOR is involved in age-related diseases [Blagosklonny, 2006a; Tsang et al., 2007]. And TOR inhibits mitophagy (autophagy of mitochondria).

MALIGNANT MITOCHONDRIA AND PERMISSIVE INTRACELLULAR MICROENVIRONMENT

In a lifetime, mutations in mtDNA accumulate in tissues of mammals [Wallace, 2005]. A high proportion of cells in tissues (as diverse as buccal epithelium and heart muscle) contains high proportions of clonal mutant mtDNA expanded from single initial mutant mtDNA molecules [Nekhaeva et al., 2002]. During aging, muscle fibers are taken over by one or only a few types of mtDNA mutants. Mutants accumulate faster in post-mitotic cells than in dividing cells. Cell division can rejuvenate the mitochondrial population, consistent with data that post-mitotic tissues accumulate mitochondrial damage faster than proliferating tissues [Kowald and Kirkwood, 2000]. Why do defective mitochondria accumulate?

Mutations in mtDNA may provide selective advantage. Like any self-replicating units, mitochondria undergo natural selection (Fig. 2). Selection among the replicating mitochondria may favor fast replicating variants, even if they are deleterious to the cell in which they reside [Taylor et al., 2002] (in analogy with malignant somatic cells, the term malignant mitochondria may be appropriate). Aging is associated with accumulation of predominantly large defective mitochondria because they are poorly autophagocytosed [Terman et al., 2003; Terman et al., 2004]. From the cell's perspective, aging is

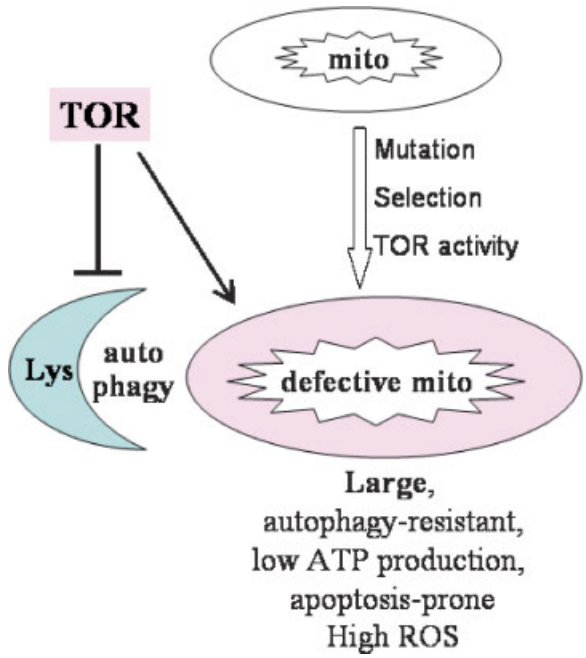

Fig. 2. Selection for ''malignant'' mitochondria. As any self-replicating unit, mitochondria may undergo selection for fitness such as resistance to autophagy. Mutations followed by selection render malignant mitochondria that may be large, resistant to autophagy, easily initiate apoptosis, produce less ATP and more ROS than is produced by normal mitochondria. TOR inhibits autophagy and creates permissive intracellular environment. [Color figure can be viewed in the online issue, which is available at www.interscience.wiley.com.]

associated with defective mitochondria. But from the mitochondrial point of view, mitochondria that do not produce ATP but instead replicate its DNA and resist autophagy are not defective at all. They are the fittest survivors that have out competed normal mitochondria. These are malignant, egoistic mitochondria.

There are cellular mechanisms to prevent accumulation of "malignant" mitochondria. Autophagy plays an important role in the degradation of excess or injured mitochondria [Mijaljica et al., 2007]. In fact, autophagy can be envisioned either as a process of mitochondrial quality control or as an ultimate cellular response triggered when cells are overwhelmed with damaged mitochondria [Priault et al., 2005]. Yet, if autophagy is inhibited that might create a permissive environment for accumulation of large, giant, non-active, malignant mitochondria. Cardiac myocytes, skeletal muscle fibers, and other long-lived post-mitotic cells show age-related clonal expansion of defective mitochondria. Such mitochondria are not prone to autophagy because of (a) mitochondrial enlargement, (b) weakened respiration, and

(c) heavy lipofuscin loading of lysosomes [Terman and Brunk, 2004]. Inhibition of autophagy in cardiac myocytes with 3-methyladenine resulted in accumulation of mitochondria within cells, loss of contractility, and reduced cell survival [Terman et al., 2003]. With advancing age, lipofuscin-loaded lysosomes and defective "giant" mitochondria occupy increasingly larger parts of long-lived post-mitotic cells (e.g., cardiac myocytes, skeletal muscle fibers) [Terman, 2006].

Aging is associated with a decrease in mitochondrial autophagy [Massey et al., 2006]. For example, in aging rats, there is accumulation of DNA-damaged mitochondria, which would otherwise be autophaged selectively [Cavallini et al., 2007]. The antilipolytic agent (3,5-dimethylpyrazole (DMP)) stimulates autophagy and rescued older cells from the accumulation of defective mitochondria [Cavallini et al., 2007].

So why is autophagy insufficient with aging? Does this imply a certain program? Aging looks like a program [Skulachev, 2002; Longo et al., 2005; Prinzinger, 2005; Skulachev and Longo, 2005]. Furthermore, aging is genetically controlled from yeast to mice that would be expected if aging were programmed. Yet, aging cannot be programmed [Williams, 1957; Partridge and Gems, 2002; Kirkwood, 2005a,b].

QUASI-PROGRAM INSTEAD OF ALTRUISTIC PROGRAM

But the program is not the only alternative to non-programmed molecular damage. Another alternative is a quasi-program, an undirected and non-intended continuation of a program. Like a program, it can be switched off. Unlike a program, it has no altruistic or any other purpose.

TOR-DRIVEN QUASI-PROGRAM FOR AGING

In the wild, multicellular organisms die from external causes (infections, predators, starvation, etc.). In the wild, 90% of mice die during their first year of life from infections, cold, starvation, predators. They would not live longer (in the wild), even if they were "immortal" [Williams, 1957; Partridge and Gems, 2002]. There is no need in post-developmental program for aging, because on the average organisms do not live so long anyway. For similar reason, there is no need to switch-off developmental programs. When animals do not die from external causes and live long enough, then a continuation of the developmental program becomes harmful over time [Blagosklonny, 2006a].

Unlike a program, a quasi-program has neither purpose nor altruistic meaning nor any meaning at all. If aging has no adaptive purpose, natural selection will select against the onset of aging as long as aging would not limit longevity of most individuals. In fact, most mice die in the wild before aging occurs. At first glance, Pacific Salmon is an exception. As a famous example, Pacific Salmon die from aging after first reproduction. It was considered as an exceptional program contrasting with "non-programmed" decay in other species [Austad, 2004].

The Pacific Salmon example is used by both advocates and opponents of programmed aging. Advocates consider Salmon as an illustration of programmed aging, whereas opponents consider it as an exception that emphasizes the rule: non-programmed aging in other species. Yet this example was misunderstood because of inaccurate assumption that Pacific Salmon die *invariably* shortly *after* their reproductive episode (spawning). In reality, Pacific Salmon almost invariably die *before* reproduction from accidental death. Each female produces about 2,000 eggs. If each of them would survive until reproduction and die from aging, then there will be $2{,}000 \times 2{,}000 = 4$ million eggs in the next generation, and so on. Actually 99.9% fish must die before reproduction (at different ages and reasons, including of being cooked for us) and only 1 out of 1,000 die after reproduction from aging. Similarly, mice (and most other species) rarely die from aging in the wild. Thus, Pacific Salmon are not exceptional. Like other animals, most Pacific Salmon die from accidental death and a few survivors die from aging. Aging plays a negligible role in the wild not only for mice but also for Pacific Salmon. This quasi-programmed aging is revealed only when accidental death is eliminated (humans, domesticated and laboratory animals).

As an unintended continuation of the developmental program, quasi-program is not very precise. This is exactly in agreement with observations that "if genes program aging, they do so only very loosely." This is in sharp contrast to the developmental process, which is so precisely regulated [Kirkwood, 2005b]. In animals

from worm to mice, knockout and mutations in numerous genes can slow down aging and extend life span [Tatar et al., 2003; Vellai et al., 2003; Kapahi et al., 2004]. These genes can be arranged in one single pathway [Blagosklonny, 2006a]: namely, the TOR pathway. TOR links environmental clues and nutrients to cell growth [Beck and Hall, 1999; Schmelzle and Hall, 2000; Crespo et al., 2002; Long et al., 2002; Jia et al., 2004; Kaeberlein et al., 2005; Inoki and Guan, 2006; Powers et al., 2006]. As recently discussed, TOR is involved in cell senescence, organism aging and diseases of aging [Blagosklonny, 2006a,b; Blagosklonny, 2007]. But how can mitochondria be linked to this quasi-program?

TOR AND MITOCHONDRIA

TOR inhibits autophagy and rapamycin (TOR inhibitor) promotes autophagy [Paglin et al., 2005; Kawai et al., 2006; Ravikumar et al., 2006; Williams et al., 2006]. For example, inactivation of TOR (by serum withdrawal) promoted autophagy of mitochondria with deleterious mtDNA mutations but spared their normal counterparts. Autophagy of dysfunctional mitochondria was prevented by IGF-1 [Gu et al., 2004], which activates the TOR pathway. I conclude that by inhibiting autophagy, TOR allows defective mitochondria to accumulate.

Also, the TOR pathway regulates mitochondrial biogenesis. For example, TOR induces hypoxia-inducible factor-1 (HIF-1alpha) [Bernardi et al., 2006]. In turn, this increases production of nitric oxide (NO) [Semenza, 2002]. Chronic increases in NO levels stimulate mitochondrial biogenesis [Nagy et al., 2004] which is associated with aging [Nisoli and Carruba, 2006]. TOR and HIF-1 stimulate iron accumulation [Fukuda et al., 2002; Treins et al., 2002; Lee and Andersen, 2006]. And iron accumulation is associated with cellular senescence [Killilea et al., 2004] and may be involved in neurodegenerative disorders [Zecca et al., 2004; Lee and Andersen, 2006]. Iron inhibits lysosomes [Eaton and Qian, 2002] and thus may inhibit autophagy. In fact, autophagy is inhibited in neurodegenerative diseases [Rubinsztein, 2006]. Also, the TOR pathway regulates mitochondrial activity [Schieke et al., 2006]. In turn, mitochondria regulate the TOR pathway [Desai et al., 2002; Giannattasio et al., 2005; Schieke and Finkel, 2006].

ROS AND TOR

Hydrogen peroxide (H_20_2) results from mitochondrial respiration. Levels of peroxide are regulated by production and degradation. Hydrogen peroxide has important roles as a signaling molecule in the regulation of a variety of biological processes. What is important for our discussion is that hydrogen peroxide activates PI3-K/TOR/S6k pathway [Bae et al., 1999; Huang et al., 2002; Liu et al., 2006]. UV-induced activation of TOR/S6K also involves hydrogen peroxide [Zhang et al., 2001; Huang et al., 2002]. And vice versa the PI3K/mTOR pathway seems to increase production of ROS [Tuñón et al., 2003; Kim et al., 2005] and sensitizes the whole organism to oxidative stress [Patel and Tamanoi, 2006]. Thus, a positive feedback loop between TOR and H_20_2 may amplify TOR-driven aging (Fig. 3).

HOW "MALIGNANT" MITOCHONDRIA CONTRIBUTE TO AGE-RELATED DISEASES

Intriguingly, human stem cells have low mitochondrial numbers [Cho et al., 2006]. In aging cells, in contrast, mitochondrial mass is increased [Lee et al., 2002]. Senescent cells have elongated giant mitochondria [Yoon et al., 2006]. Large mitochondria are difficult to autophage and therefore they accumulate. Large size can be a problem by itself. For example, mitochondria should be small enough

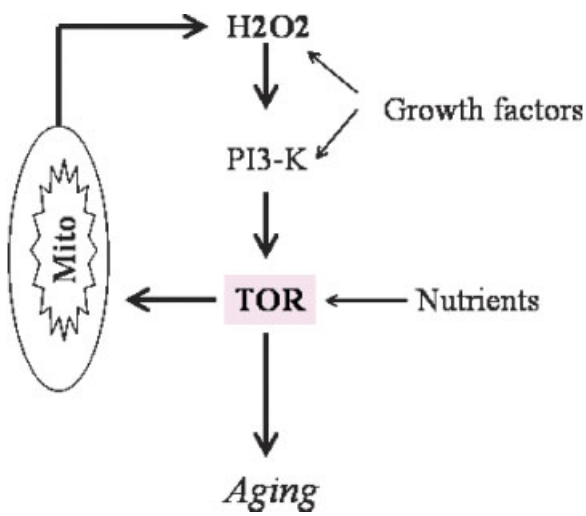

Fig. 3. Positive feedback between ROS and TOR. Via PI3-K peroxide stimulates TOR, which in turn inhibits autophagy of defective mitochondria, potentially increasing peroxide. [Color figure can be viewed in the online issue, which is available at www.interscience.wiley.com.]

to fit in neuronal dendritic and axon terminals [Chan, 2006]. I suggest that enlargement of mitochondria may contribute to synaptic defects in age-related neurodegenerative diseases. Second, increased mitochondrial mass is associated with respiratory defects [Wallace, 2005]. Third, increased mitochondrial mass renders cells sensitive to apoptosis [Petrovas et al., 2006]. Increased susceptibility to apoptosis has been shown in many models of mitochondrial defects [Aure et al., 2006]. Rapamycin enhances clearance of mitochondria and protects cells against pro-apoptotic insults in Drosophila [Ravikumar et al., 2006]. In patients with mtDNA disorders, apoptosis occurred in muscle fibers with mitochondrial proliferation. Thus, large size and propensity to apoptosis can contribute to some age-related diseases. In contrast, ROS may be irrelevant. Thus, in mtDNA mutator mice the amount of ROS produced is normal, despite respiratory chain dysfunction [Trifunovic et al., 2005]. In Drosophila and *C. elegans*, partial disruption of mitochondrial machinery results in life span extension, despite a mild increase in their ROS leak rate [Rea, 2005; Ventura et al., 2006].

Mitochondrial alterations are characteristic of several neurodegenerative human disorders: Parkinson's, Alzheimer's, and Huntington's diseases [Calabrese et al., 2001; Chan, 2006]. Also, mitochondrial dysfunction and mtDNA mutations are associated with age-related hearing loss [Fischel-Ghodsian, 2003; Pickles, 2004; Yamasoba et al., 2007]. Calorie restriction attenuates age-related cochlear degeneration and decreases quantity of mtDNA deletions in the cochlea of mammals [Yamasoba et al., 2007]. Thus, accumulation of defective mitochondria probably contributes to some age-related diseases.

Mice expressing a proof reading-deficient version of the mtDNA polymerase gamma (POLG) accumulate mtDNA mutations and display features of accelerated aging [Kujoth et al., 2005]. Noteworthy, there was no increase in ROS production. Instead there was increased apoptotic rate, particularly in tissues characterized by rapid cellular turnover [Kujoth et al., 2005]. Yet, the levels of mutations in the mutator mice are more than an order of magnitude higher than typical levels in aged humans [Khrapko et al., 2006] and 500 times higher than in mice [Vermulst et al., 2007]. Most of the aging-like features in the mutator mice are not specific to features of normal aging. For example, in mutator mice, aging was associated with weight loss, reduced subcutaneous fat, hair loss, curvature of the spine, osteoporosis, anemia, reduced fertility and heart enlargement. The mitochondrial syndrome is characterized by the widespread apoptosis [Trifunovic et al., 2004]. Yet, this does not resemble regular mammalian aging. Apoptosis is not a significant characteristic of normal aging.

CONCLUSIONS

Mitochondria replicate DNA and acquire mutations. Therefore, they inevitably undergo "Darwinian" selection inside the cell in long-lived organism. "Darwinian" selection among mitochondria yields "malignant" mitochondria, characterized by a giant morphology, resistance to autophagy, and decreased ATP production. Due to increased mass, malignant mitochondria may be inclined to initiate apoptosis. This may be manifested as age-related apoptosis in neurons and auditory cells. Malignant mitochondria can contribute to age-related diseases, when apoptosis is involved (hearing loss). "Malignization" of mitochondria depends on a permissive intracellular environment that is TOR-dependent. TOR inhibits autophagy, thus potentially allowing malignant mitochondria to accumulate and clonally expand.

We can envision two therapeutic approaches. First, malignant mitochondria could be targeted directly by mitochondria-directed agents [Skulachev and Longo, 2005]. The second

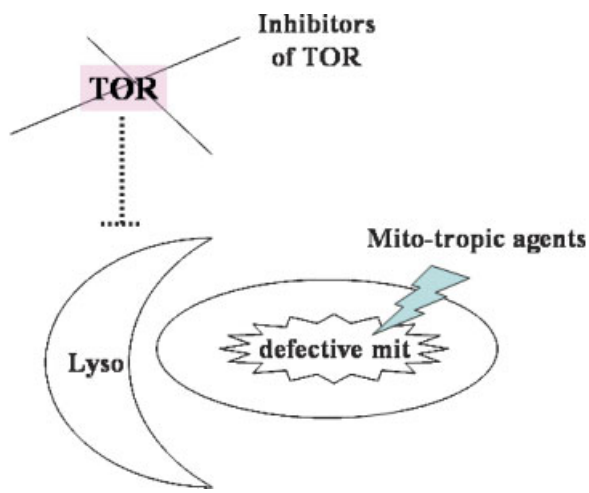

Fig. 4. Therapeutic approach to target both TOR and mitochondria. [Color figure can be viewed in the online issue, which is available at www.interscience.wiley.com.]

approach is to re-activate autophagy by inhibiting TOR. In addition, inhibitors of TOR would stimulate autophagy of aggregation-prone and damaged proteins, inhibit cell hypertrophy, cellular iron and lipid consumption and perhaps protect cells from ROS, if ROS play some role in aging. Two approaches (re-activation of autophagy and direct targeting mitochondria) can be combined (Fig. 4). For example, mitotropic drugs can be combined with inhibitors of the TOR such as rapamycin and metformin.

REFERENCES

Andziak B, O'Connor TP, Buffenstein R. 2005. Antioxidants do not explain the disparate longevity between mice and the longest-living rodent, the naked mole-rat. Mech Ageing Dev 126:1206–1212.

Andziak B, O'Connor TP, Qi W, DeWaal EM, Pierce A, Chaudhuri AR, Van Remmen H, Buffenstein R. 2006. High oxidative damage levels in the longest-living rodent, the naked mole-rat. Aging Cell 5:463–471.

Aure K, Fayet G, Leroy JP, Lacene E, Romero NB, Lombes A. 2006. Apoptosis in mitochondrial myopathies is linked to mitochondrial proliferation. Brain 129:1249–1259.

Austad SN. 2004. Is aging programed? Aging Cell 3:249–251.

Bae GU, Seo DW, Kwon HK, Lee HY, Hong S, Lee ZW, Ha KS, Lee HW, Han JW. 1999. Hydrogen peroxide activates p70(S6k) signaling pathway. J Biol Chem 274:32596–33602.

Beck T, Hall MN. 1999. The TOR signalling pathway controls nuclear localization of nutrient-regulated transcription factors. Nature 402:689–692.

Bernardi R, Guernah I, Jin D, Grisendi S, Alimonti A, Teruya-Feldstein J, Cordon-Cardo C, Simon MC, Rafii S, Pandolfi PP. 2006. PML inhibits HIF-1alpha translation and neoangiogenesis through repression of mTOR. Nature 442:779–785.

Bjelakovic G, Nikolova D, Gluud LL, Simonetti RG, Gluud C. 2007. Mortality in randomized trials of antioxidant supplements for primary and secondary prevention: Systematic review and meta-analysis. JAMA 297:842–857.

Blackstone NW, Green DR. 1999. The evolution of a mechanism of cell suicide. Bioessays 21:84–88.

Blagosklonny MV. 2005. Overcoming limitations of natural anticancer drugs by combining with designed agents. Trends Pharm Sci 26:77–81.

Blagosklonny MV. 2006a. Aging and immortality: Quasi-programmed senescence and its pharmacologic inhibition. Cell Cycle 5:2087–2102.

Blagosklonny MV. 2006b. Cell senescence: Hypertrophic arrest beyond restriction point. J Cell Physiol 209:592–597.

Blagosklonny MV. 2007. An anti-aging drug today: From senescence-promoting genes to anti-aging pill. Drug Disc Today 12:218–224.

Buffenstein R. 2005. The naked mole-rat: A new long-living model for human aging research. J Gerontol A Biol Sci Med Sci 60:1369–1377.

Calabrese V, Scapagnini G, Stella AMG, Bates TE, Clark JB. 2001. Mitochondrial involvement in brain function and dysfunction: Relevance to aging, neurodegenerative disorders and longevity. Neurochemical Res 26:739–764.

Cavallini G, Donati A, Taddei M, Bergamini E. 2007. Evidence for selective mitochondrial autophagy and failure in aging. Autophagy 3:26–27.

Chan DC. 2006. Mitochondria: Dynamic organelles in disease, aging, and development. Cell 125:1241–1252.

Cho YM, Kwon S, Pak YK, Seol HW, Choi YM, Park dJ, Park KS, Lee HK. 2006. Dynamic changes in mitochondrial biogenesis and antioxidant enzymes during the spontaneous differentiation of human embryonic stem cells. Biochem Biophys Res Commun 348:1472–1478.

Christophorou MA, Ringshausen I, Finch AJ, Swigart LB, Evan GI. 2006. The pathological response to DNA damage does not contribute to p53-mediated tumour suppression. Nature 443:214–217.

Corona M, Hughes KA, Weaver DB, Robinson GE. 2005. Gene expression patterns associated with queen honey bee longevity. Mech Ageing Dev 126:1230–1238.

Crespo JL, Powers T, Fowler B, Hall MN. 2002. The TOR-controlled transcription activators GLN3, RTG1, and RTG3 are regulated in response to intracellular levels of glutamine. Proc Natl Acad Sci U S A 99:6784–6789.

Cronin H. 2006. The battle of the sexes reviseted. In: Grafen A, Ridley M, editors. Richard Dawkins: How a scientist changed the way we think. NY: Oxford Univ. Press.

Desai BN, Myers BR, Schreiber SL. 2002. FKBP12-rapamycin-associated protein associates with mitochondria and senses osmotic stress via mitochondrial dysfunction. PNAS 99:4319–4324.

Dyall SD, Brown MT, Johnson PJ. 2004. Ancient invasions: From endosymbionts to organelles. Science 304:253–257.

Eaton JW, Qian M. 2002. Molecular bases of cellular iron toxicity. Free Radic Biol Med 32:833–840.

Esteve I, Gaju N. 1999. Bacterial symbioses. Predation and mutually beneficial associations. Int Microbiol 2:81–86.

Fabrizio P, Battistella L, Vardavas R, Gattazzo C, Liou LL, Diaspro A, Dossen JW, Gralla EB, Longo VD. 2004. Superoxide is a mediator of an altruistic aging program in *Saccharomyces cerevisiae*. J Cell Biol 166:1055–1067.

Fischel-Ghodsian N. 2003. Mitochondrial deafness. Ear Hear 24:303–313.

Fukuda R, Hirota K, Fan F, Jung YD, Ellis LM, Semenza GL. 2002. Insulin-like growth factor 1 induces hypoxia-inducible factor 1-mediated vascular endothelial growth factor expression, which is dependent on MAP kinase and phosphatidylinositol 3-kinase signaling in colon cancer cells. J Biol Chem 277:38205–38211.

Giannattasio S, Liu Z, Thornton J, Butow RA. 2005. Retrograde response to mitochondrial dysfunction is separable from TOR1/2 regulation of retrograde gene expression. J Biol Chem 280:42528–42535.

Green DR, Evan GI. 2002. A matter of life and death. Cancer Cell 1:19–30.

Gu Y, Wang C, Cohen A. 2004. Effect of IGF-1 on the balance between autophagy of dysfunctional mitochondria and apoptosis. FEBS Lett 577:357–360.

Herker E, Jungwirth H, Lehmann KA, Maldener C, Frohlich KU, Wissing S, Buttner S, Fehr M, Sigrist S, Madeo F. 2004. Chronological aging leads to apoptosis in yeast. J Cell Biol 164:501–507.

Howes RM. 2006. The free radical fantasy: A panoply of paradoxes. Ann N Y Acad Sci 1067:22–26.

Huang C, Li J, Ke Q, Leonard SS, Jiang BH, Zhong XS, Costa M, Castranova V, Shi X. 2002. Ultraviolet-induced phosphorylation of p70(S6K) at Thr(389) and Thr(421)/Ser(424) involves hydrogen peroxide and mammalian target of rapamycin but not Akt and atypical protein kinase C. Cancer Res 62:5689–5697.

Inoki K, Guan KL. 2006. Complexity of the TOR signaling network. Trends Cell Biol 16:206–212.

Jendro MC, Kohler L, Kuipers JG, Zeidler H. 2002. Microbe-induced T cell apoptosis: Subversion of the host defense system? FEMS Microbiol Lett 207:121–126.

Jia K, Chen D, Riddle DL. 2004. The TOR pathway interacts with the insulin signaling pathway to regulate *C. elegans* larval development, metabolism and life span. Development 131:3897–3906.

Jin Z, El-Deiry WS. 2005. Overview of cell death signaling pathways. Cancer Biol Ther 4.

Jungwirth H, Kuchler K. 2006. Yeast ABC transporters—A tale of sex, stress, drugs and aging. FEBS Lett 580:1131–1138.

Kaeberlein M, Powers RW, III, Steffen KK, Westman EA, Hu D, Dang N, Kerr EO, Kirkland KT, Fields S, Kennedy BK. 2005. Regulation of yeast replicative life span by TOR and Sch9 in response to nutrients. Science 310:1193–1196.

Kapahi P, Zid BM, Harper T, Koslover D, Sapin V, Benzer S. 2004. Regulation of lifespan in Drosophila by modulation of genes in the TOR signaling pathway. Curr Biol 14:885–890.

Kawai A, Takano S, Nakamura N, Ohkuma S. 2006. Quantitative monitoring of autophagic degradation. Biochem Biophys Res Commun 351:71–77.

Khrapko K, Kraytsberg Y, de Grey AD, Vijg J, Schon EA. 2006. Does premature aging of the mtDNA mutator mouse prove that mtDNA mutations are involved in natural aging? Aging Cell 5:279–282.

Killilea DW, Wong SL, Cahaya HS, Atamna H, Ames BN. 2004. Iron accumulation during cellular senescence. Ann N Y Acad Sci 1019:365–367.

Kim JH, Chu SC, Gramlich JL, Pride YB, Babendreier E, Chauhan D, Salgia R, Podar K, Griffin JD, Sattler M. 2005. Activation of the PI3K/mTOR pathway by BCR-ABL contributes to increased production of reactive oxygen species. Blood 105:1717–1723.

Kirkwood TB. 2005a. Time of our lives. What controls the length of life? EMBO Rep 6:S4–8.

Kirkwood TB. 2005b. Understanding the odd science of aging. Cell 120:437–447.

Komarova EA, Kondratov RV, Wang K, Christov K, Golovkina TV, Goldblum JR, Gudkov AV. 2004. Dual effect of p53 on radiation sensitivity in vivo: p53 promotes hematopoietic injury, but protects from gastro-intestinal syndrome in mice. Oncogene 23:3265–3271.

Kowald A, Kirkwood TB. 2000. Accumulation of defective mitochondria through delayed degradation of damaged organelles and its possible role in the ageing of post-mitotic and dividing cells. J Theor Biol 202:145–160.

Kubasiak LA, Hernandez OM, Bishopric NH, Webster KA. 2002. Hypoxia and acidosis activate cardiac myocyte death through the Bcl-2 family protein BN IP3. Proc Natl Acad Sci U S A 99:12825–12830.

Kujoth GC, Hiona A, Pugh TD, Someya S, Panzer K, Wohlgemuth SE, Hofer T, Seo AY, Sullivan R, Jobling WA, Morrow JD, Van Remmen H, Sedivy JM, Yamasoba T, Tanokura M, Weindruch R, Leeuwenburgh C, Prolla TA. 2005. Mitochondrial DNA mutations, oxidative stress, and apoptosis in mammalian aging. Science 309:481–484.

Kutschera U, Niklas KJ. 2005. Endosymbiosis, cell evolution, and speciation. Theory Biosci 124:1–24.

Lee DW, Andersen JK. 2006. Role of HIF-1 in iron regulation: Potential therapeutic strategy for neurodegenerative disorders. Curr Mol Med 6:883–893.

Lee HC, Yin PH, Chi CW, Wei YH. 2002. Increase in mitochondrial mass in human fibroblasts under oxidative stress and during replicative cell senescence. J Biomed Sci 9:517–526.

Li J, Greco V, Guasch G, Fuchs E, Mombaerts P. 2007. Mice cloned from skin cells. Proc Natl Acad Sci U S A 104:2738–2743.

Liu LZ, Hu XW, Xia C, He J, Zhou Q, Shi X, Fang J, Jiang BH. 2006. Reactive oxygen species regulate epidermal growth factor-induced vascular endothelial growth factor and hypoxia-inducible factor-1alpha expression through activation of AKT and P70S6K1 in human ovarian cancer cells. Free Radic Biol Med 41:1521–1533.

Long X, Spycher C, Han ZS, Rose AM, Muller F, Avruch J. 2002. TOR deficiency in *C. elegans* causes developmental arrest and intestinal atrophy by inhibition of mRNA translation. Curr Biol 12:1448–1461.

Longo VD, Mitteldorf J, Skulachev VP. 2005. Programmed and altruistic ageing. Nat Rev Genet 6:66–872.

Madeo F, Herker E, Wissing S, Jungwirth H, Eisenberg T, Frohlich KU. 2004. Apoptosis in yeast. Curr Opin Microbiol 7:655–660.

Massey AC, Kiffin R, Cuervo AM. 2006. Autophagic defects in aging: Looking for an "emergency exit"? Cell Cycle 5:1292–1296.

Mijaljica D, Prescott M, Devenish RJ. 2007. Different fates of mitochondria: Alternative ways for degradation? Autophagy 3:417–421.

Nagy G, Barcza M, Gonchoroff N, Phillips PE, Perl A. 2004. Nitric oxide-dependent mitochondrial biogenesis generates Ca2+ signaling profile of lupus T cells. J Immunol 173:3676–3683.

Narasimhan ML, Damsz B, Coca MA, Ibeas JI, Yun DJ, Pardo JM, Hasegawa PM, Bressan RA. 2001. A plant defense response effector induces microbial apoptosis. Mol Cell 8:921–930.

Nekhaeva E, Bodyak ND, Kraytsberg Y, McGrath SB, Van Orsouw NJ, Pluzhnikov A, Wei JY, Vijg J, Khrapko K. 2002. Clonally expanded mtDNA point mutations are abundant in individual cells of human tissues. Proc Natl Acad Sci U S A 99:5521–5526.

Newton K, Strasser A. 2003. Caspases signal not only apoptosis but also antigen-induced activation in cells of the immune system. Genes Dev 17:819–825.

Nisoli E, Carruba MO. 2006. Nitric oxide and mitochondrial biogenesis. J Cell Sci 119:2855–2862.

Paglin S, Lee NY, Nakar C, Fitzgerald M, Plotkin J, Deuel B, Hackett N, McMahill M, Sphicas E, Lampen N, Yahalom J. 2005. Rapamycin-sensitive pathway regulates mitochondrial membrane potential, autophagy, and survival in irradiated MCF-7 cells. Cancer Res 65:11061–11070.

Parker JD, Parker KM, Sohal BH, Sohal RS, Keller L. 2004. Decreased expression of Cu-Zn superoxide dismutase 1 in ants with extreme lifespan. Proc Natl Acad Sci U S A 101:3486–3489.

Partridge L, Gems D. 2002. Mechanisms of ageing: Public or private? Nat Rev Genet 3:165–175.

Patel PH, Tamanoi F. 2006. Increased Rheb-TOR signaling enhances sensitivity of the whole organism to oxidative stress. J Cell Sci 119:4285–4292.

Petrovas C, Mueller YM, Dimitriou ID, Altork SR, Banerjee A, Sklar PA, Mounzer KC, Altman JD, Katsikis PD. 2006. Increased mitochondrial mass characterizes the survival defect of HIV-specific CD8+ T cells. Blood 109:2505–2513.

Pickles JO. 2004. Mutation in mitochondrial DNA as a cause of presbyacusis. Audiol Neurootol 9:23–33.

Powers RW, III, Kaeberlein M, Caldwell SD, Kennedy BK, Fields S. 2006. Extension of chronological life span in yeast by decreased TOR pathway signaling. Genes Dev 20:174–184.

Pozniakovsky A, Knorre DA, Markova OV, Hyman AA, Skulachev VP, Severin FF. 2005. Role of mitochondria in the pheromone- and amiodarone-induced programmed death of yeast. J Cell Biol 168:257–269.

Priault M, Salin B, Schaeffer J, Vallette FM, di Rago JP, Martinou JC. 2005. Impairing the bioenergetic status and the biogenesis of mitochondria triggers mitophagy in yeast. Cell Death Differ 12:1613–1621.

Prinzinger R. 2005. Programmed ageing: The theory of maximal metabolic scope. EMBO Rep 6:S14–19.

Ravagnan L, Roumier T, Kroemer G. 2002. Mitochondria, the killer organelles and their weapons. J Cell Physiol 192:131–137.

Ravikumar B, Berger Z, Vacher C, O'Kane CJ, Rubinsztein DC. 2006. Rapamycin pre-treatment protects against apoptosis. Hum Mol Genet 15:1209–1216.

Rea SL. 2005. Metabolism in the *Caenorhabditis elegans* Mit mutants. Exp Gerontol 40:841–849.

Rubinsztein DC. 2006. The roles of intracellular protein-degradation pathways in neurodegeneration. Nature 443:780–786.

Schieke SM, Finkel T. 2006. Mitochondrial signaling, TOR, and life span. Biol Chem 387:1357–1361.

Schieke SM, Phillips D, McCoy JPJ, Aponte AM, Shen RF, Balaban RS, Finkel T. 2006. The mammalian target of rapamycin (mTOR) pathway regulates mitochondrial oxygen consumption and oxidative capacity. J Biol Chem 281:27643–27652.

Schmelzle T, Hall MN. 2000. TOR, a central controller of cell growth. Cell 103:253–262.

Schnaith A, Kashkar H, Leggio S, Addicks K, Kronke M, Krut O. 2006. Staphylococcus aureus subvert autophagy for induction of caspase-independent host cell death. J Biol Chem

Semenza GL. 2002. HIF-1 and tumor progression: Pathophysiology and therapeutics. Trends Mol Med 8:S62–S67.

Seto NO, Hayashi S, Tener GM. 1990. Overexpression of Cu–Zn superoxide dismutase in Drosophila does not affect life-span. Proc Natl Acad Sci U S A 87:4270–4274.

Skulachev VP. 2002. Programmed death phenomena: From organelle to organism. Ann N Y Acad Sci 959:214–237.

Skulachev VP, Longo VD. 2005. Aging as a mitochondria-mediated atavistic program: Can aging be switched off? Ann N Y Acad Sci 1057:145–164.

Spees JL, Olson SD, Whitney MJ, Prockop DJ. 2006. Mitochondrial transfer between cells can rescue aerobic respiration. Proc Natl Acad Sci U S A 103:1283–1288.

Strom E, Sathe S, Komarov PG, Chernova OB, Pavlovska I, Shyshynova I, Bosykh DA, Burdelya LG, Macklis RM, Skaliter R, Komarova EA, Gudkov AV. 2006. Small-molecule inhibitor of p53 binding to mitochondria protects mice from gamma radiation. Nat Chem Biol 2: 474–479.

Sutovsky P, Moreno RD, Ramalho-Santos J, Dominko T, Simerly C, Schatten G. 1999. Ubiquitin tag for sperm mitochondria. Nature 402:371–372.

Tal R, Winter G, Ecker N, Klionsky DJ, Abeliovich H. 2007. Aup1p, a yeast mitochondrial protein phosphatase homolog, is required for efficient stationary phase mitophagy and cell survival. J Biol Chem 282:5617–5624.

Tatar M, Bartke A, Antebi A. 2003. The endocrine regulation of aging by insulin-like signals. Science 299:1346–1351.

Taylor DR, Zeyl C, Cooke E. 2002. Conflicting levels of selection in the accumulation of mitochondrial defects in *Saccharomyces cerevisiae*. Proc Natl Acad Sci U S A 99:3690–3694.

Terman A. 2006. Catabolic insufficiency and aging. Ann N Y Acad Sci 1067:27–36.

Terman A, Brunk UT. 2004. Myocyte aging and mitochondrial turnover. EXP Gerontology 39:701–705.

Terman A, Dalen H, Eaton JW, Neuzil J, Brunk UT. 2003. Mitochondrial recycling and aging of cardiac myocytes: The role of autophagocytosis. Exp Gerontol 38:863–876.

Terman A, Dalen H, Eaton JW, Neuzil J, Brunk UT. 2004. Aging of cardiac myocytes in culture: Oxidative stress, lipofuscin accumulation, and mitochondrial turnover. Ann N Y Acad Sci 1019:70–77.

Tolkovsky AM, Xue L, Fletcher GC, Borutaite V. 2002. Mitochondrial disappearance from cells: A clue to the role of autophagy in programmed cell death and disease. Biochimie 84:233–240.

Treins C, Giorgetti-Peraldi S, Murdaca J, Semenza GL, Van Obberghen E. 2002. Insulin stimulates hypoxia-inducible factor 1 through a phosphatidylinositol 3-kinase/target of rapamycin-dependent signaling pathway. J Biol Chem 277:27975–27981.

Trifunovic A, Wredenberg A, Falkenberg M, Spelbrink JN, Rovio AT, Bruder CE, Bohlooly-Y M, Gidlof S, Oldfors A, Wibom R, Tornell J, Jacobs HT, Larsson NG. 2004. Premature ageing in mice expressing defective mitochondrial DNA polymerase. Nature 429:417–423.

Trifunovic A, Hansson A, Wredenberg A, Rovio AT, Dufour E, Khvorostov I, Spelbrink JN, Wibom R, Jacobs HT, Larsson NG. 2005. Somatic mtDNA mutations cause aging phenotypes without affecting reactive oxygen species production. Proc Natl Acad Sci U S A 102: 17993–17998.

Tsang CK, Qi H, Liu LF, Zheng XFS. 2007. Targeting mammalian target of rapamycin (mTOR) for health and diseases. Drug Disc Today 12:112–124.

Tuñón MJ, Sánchez-Campos S, Gutiérrez B, Culebras JM, González-Gallego J. 2003. Effects of FK506 and rapamycin on generation of reactive oxygen species, nitric oxide

production and nuclear factor kappa B activation in rat hepatocytes. Biochem Pharmacol 66:439–445.

van Loo G, Saelens X, van Gurp M, MacFarlane M, Martin SJ, Vandenabeele P. 2002. The role of mitochondrial factors in apoptosis: A Russian roulette with more than one bullet. Cell Death Differ 9:1031–1042.

Van Remmen H, Ikeno Y, Hamilton M, Pahlavani M, Wolf N, Thorpe SR, Alderson NL, Baynes JW, Epstein CJ, Huang TT, Nelson J, Strong R, Richardson A. 2003. Life-long reduction in MnSOD activity results in increased DNA damage and higher incidence of cancer but does not accelerate aging. Physiol Genomics 16:29–37.

Vellai T, Takacs-Vellai K, Zhang Y, Kovacs AL, Orosz L, Muller F. 2003. Genetics: Influence of TOR kinase on lifespan in *C. elegans*. Nature 426:620.

Ventura N, Rea SL, Testi R. 2006. Long-lived *C. elegans* Mitochondrial mutants as a model for human mitochondrial-associated diseases. Exp Gerontol 41:974–991.

Vermulst M, Bielas JH, Kujoth GC, Ladiges WC, Rabinovitch PS, Prolla TA, Loeb LA. 2007. Mitochondrial point mutations do not limit the natural lifespan of mice. Nat Genet 39:540–543.

Wallace DC. 2005. A mitochondrial paradigm of metabolic and degenerative diseases, aging, and cancer: A dawn for evolutionary medicine. Annu Rev Genet 39:359–407.

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

Williams A, Jahreiss L, Sarkar S, Saiki S, Menzies FM, Ravikumar B, Rubinsztein DC. 2006. Aggregate-prone proteins are cleared from the cytosol by autophagy: Therapeutic implications. Curr Top Dev Biol 76:89–101.

Yamasoba T, Someya S, Yamada C, Weindruch R, Prolla TA, Tanokura M. 2007. Role of mitochondrial dysfunction and mitochondrial DNA mutations in age-related hearing loss. Hear Res 226:185–193.

Yoon YS, Yoon DS, Lim IK, Yoon SH, Chung HY, Rojo M, Malka F, Jou MJ, Martinou JC, Yoon G. 2006. Formation of elongated giant mitochondria in DFO-induced cellular senescence: Involvement of enhanced fusion process through modulation of Fis1. J Cell Physiol 209:468–480.

Zadrag R, Kwolek-Mirek M, Bartosz G, Bilinski T. 2006. Relationship between the replicative age and cell volume in *Saccharomyces cerevisiae*. Acta Biochim Pol 53:747–751.

Zecca L, Youdim MB, Riederer P, Connor JR, Crichton RR. 2004. Iron, brain ageing and neurodegenerative disorders. Nat Rev Neurosci 5:863–873.

Zhang Y, Dong Z, Nomura M, Zhong S, Chen N, Bode AM, Dong Z. 2001. Signal transduction pathways involved in phosphorylation and activation of p70S6K following exposure to UVA irradiation. J Biol Chem 276:20913–20923.