ARTICLE

Nuclear Apoptosis Contributes to Sarcopenia

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ALWAY, S.E., and P.M. SIU. Nuclear apoptosis contributes to sarcopenia. *Exerc. Sport Sci. Rev.*, Vol. 36, No. 2, pp. 51–57, 2008. *Apoptosis results in DNA fragmentation and, subsequently, destruction of cells containing a single nucleus. Our hypothesis is that multinucleated cells such as muscle fibers can experience apoptotic-induced loss of single nuclei (nuclear apoptosis) without destruction of the entire fiber. The loss of nuclei likely contributes to atrophy and sarcopenia. Furthermore, increased chronic activity attenuates apoptotic signaling, which may reduce sarcopenia.* **Key Words:** atrophy, mitochondria, skeletal myofiber, satellite cells, signaling proteins

INTRODUCTION

Frailty in the healthy elderly has become a widespread problem central to the care of geriatric populations. The major factor contributing to frailty is an age-associated loss of muscle mass and function, which has been termed sarcopenia (10). All elderly people show evidence of sarcopenia and loss of function particularly after the seventh decade of life with approximately 40% decline in muscle mass by the age of 80 years (10). The loss of muscle mass and strength with age (particularly in men older than 60 years) is associated with increased mortality (15).

Improvements in muscle mass are largely dependent upon adding new nuclei (6), and declines in muscle mass occur when nuclei are eliminated from skeletal muscles (1,4,27). Our hypothesis is that multinucleated muscle fibers undergo an aging-induced apoptotic loss of select nuclei (nuclear apoptosis). This is followed by fiber atrophy which in turn leads to sarcopenia. Muscle unloading or the lack of exercise will promote nuclear apoptosis and exacerbate sarcopenia (9,26). Although the loss of nuclei and the decline in muscle mass and strength with age cannot be stopped, increasing chronic activity can attenuate apoptotic signaling (25,30), which may reduce or slow sarcopenia.

Partial restoration of muscle mass and function can be achieved through increased loading and high resistance exercise programs by activating satellite cells (2,3). Satellite cells proliferate to increase the total number of myonuclei, and this in turn leads to increases in muscle fiber size (3,6); however, activated satellite cells are also vulnerable targets for apoptosis (11,27). Therefore, there is a great need to understand the mechanisms regulating nuclear loss and for designing strategies that will reduce nuclear apoptosis and promote an increase in myonuclei in aging.

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MECHANISMS UNDERLYING SARCOPENIA

The mechanisms responsible for sarcopenia are not well understood, but likely have several contributing factors. These include declines in neural function, hormonal deficits, inappropriate signaling caused by inadequate nutrition, and chronic inflammation. Loss of muscle in aging can be exacerbated with reduced activity.

Exercise and overload have been used to compensate for sarcopenia and loss of muscle function in humans, rats, and other animal models of aging; however, the degree of hypertrophy is typically attenuated in senescence as compared with young adults. For example, 30 d of overload results in increases in muscle mass of 44% and 25% in adult and aged quails, respectively (6). Fourteen days of functional overload in the rat plantaris muscle increases muscle weight by 25% in young adult animals but only by 9% in old rats (2,3).

It is clear that compensation to overload requires hypertrophy of the existing muscle fibers, and in some models, addition of new fibers. Hypertrophy of adult skeletal muscle requires increased protein synthesis and accumulation of proteins, and this necessitates increased transcription of muscle genes.

Protein synthesis and accumulation is attenuated with aging in muscles of animals and in humans. This contributes to sarcopenia by reducing the amount of protein available for

normal myofibril turnover or muscle repair. However, it is not known which step(s) in the regulation of muscle protein synthesis fails to respond properly with aging. It is, for example, possible that specific genes are not activated properly in muscles from aged hosts. This would occur if there were transcriptional limitations in aged muscle caused, for example, by lower levels or reduced activity of muscle regulatory factors or increased expression of repressor proteins that interfered directly with DNA regulation of muscle transcription. Alternatively, a lower number of myonuclei and/or satellite cells/muscle precursor cells could account for reduced or inadequate gene regulation.

SATELLITE CELLS AND AGING

Satellite cells play a critical role in regeneration, hypertrophy, and postnatal growth in muscles of both young and aging hosts. Satellite cells seem to be equally important to allow muscle growth in old age (6). Aging reduces the ability of muscles to repair or hypertrophy in response to various stimuli. This may be linked to a lower population of satellite cells with advanced age. For example, at birth, satellite cells account for approximately 30% of muscle nuclei, but these drop to less than 5% in the adult muscle. This is likely a direct reflection of satellite cell fusion into new or preexisting myofibers without subsequent renewal of the cell population. Alternatively, some satellite cells could be targeted for elimination. The remaining satellite cells are capable of recruitment and involvement in muscle remodeling in old host animals, although at a lower level than in young adults.

GENERAL CHARACTERISTICS OF APOPTOSIS

Apoptosis is an energy-dependent internally encoded biological destructive process that involves tightly regulated cellular signaling to coordinate the sequential apoptotic cascades. The process of apoptosis is highly conserved from worms to humans. Apoptosis was originally described from investigations in cell populations, which had a single nucleus. In these cells, apoptosis consists of a self-destructive signaling pathway that leads to elimination of the entire cell (Fig. 1A). This is a tightly controlled process involving a sequential progression of induction, execution, and degradation.

Proper regulation of apoptosis is important for correct tissue function. Apoptosis is critical for eliminating damaged, aberrant, or harmful cells, and for regulation of normal embryonic development, tissue turnover, and immunological function. Aberrant regulation of apoptosis contributes to the pathogenesis of severe diseases including viral infections, cancers, neurodegenerative diseases such as Alzheimer and Parkinson diseases, autoimmune diseases including systemic lupus, rheumatoid arthritis, myocardial and cerebral ischemic injuries, loss of pancreatic β cells in diabetes mellitus, toxin-induced liver disease, and acquired immune deficiency syndrome.

Apoptosis has several clearly defined morphological characteristics that include cell shrinkage, cell membrane blebbing, chromatin condensation, internucleosomal degradation of chromosomal DNA, and formation of membrane-bound fragments called apoptotic bodies. Another distinctive characteristic of apoptosis is that it occurs in the absence of any inflammatory response and disturbance to the surrounding cells or tissues. This characteristic of apoptosis permits highly selective dismissal of certain designated individual cells among the whole cell population. This process strongly contrasts with necrosis, which invokes a large cellular immunological response.

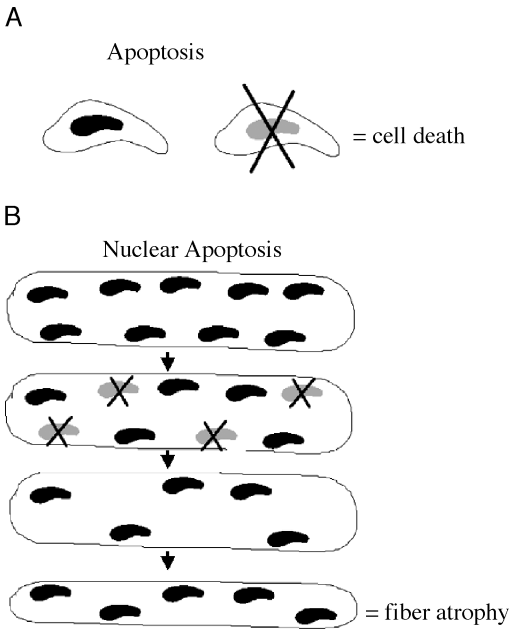

Figure 1. Apoptosis and nuclear apoptosis. A. Apoptosis results in nuclear fragmentation and elimination of the nuclei and death of single cells. B. Our hypothesis is that nuclear apoptotic signaling targets specific myonuclei in a muscle fiber. Elimination of myonuclei results in a decrease in the number of nuclei in the muscle fiber and, therefore, an increase in the cytoplasmic volume per nucleus. The muscle fiber undergoes atrophy to accommodate the fewer available nuclei for transcriptional control in an attempt to reestablish the original cytoplasmic volume per nucleus.

NUCLEAR APOPTOSIS IN POSTMITOTIC MYOFIBERS

Apoptosis was initially described as a process that was responsible for elimination of entire cells (Fig. 1A), and this was essential for maintaining the homeostasis of cell growth and death, especially in cells with a high proliferative rate. In the context of single cells, the term apoptosis has a clearly defined process leading to elimination of the nucleus and the cell. However, the better term to describe this same process in multinucleated postmitotic cell populations including cardiomyocytes and skeletal myofibers is "nuclear apoptosis." This is because elimination of a single nucleus can occur without the death of the entire (multinucleated) muscle cell, although this may result in smaller cells (Fig. 1B). We propose that the process of apoptotic loss of myonuclei in skeletal muscle should be best described as nuclear apoptosis. Nuclear apoptosis can occur without inflammation or disturbing adjacent proteins or organelles. Thus, our hypothesis

is that nuclear apoptosis is at least partly responsible for removing nuclei and/or satellite cells under muscle wasting conditions, and nuclear apoptosis both contributes to sarcopenia and limits muscle adaptations to a hypertrophic stimulus.

The concept of nuclear apoptosis (*i.e.*, death of a nucleus without death of the entire cell) is both intriguing and exciting. By definition, nuclear apoptosis involves cell signaling that is so precise that specific individual nuclei can be targeted for elimination in a multinucleated skeletal myofiber without targeting other nuclei. Thus, nuclear apoptosis requires amazingly precise targeting of some nuclei but not others within a single muscle fiber.

Evidence that not all myonuclei in a single myofiber become apoptotic during muscle loss has been observed in experimental denervation and denervation-associated disease (*e.g.*, infantile spinal muscular atrophy). This further supports the hypothesis of nuclear apoptosis in modulating the myofiber volume by controlling the successive myofiber segments. The hypothesis of nuclear apoptosis is consistent with the proposed nuclear domain hypothesis, which explains the phenomenon of cell size remodeling of myofiber by adding or subtracting nuclei because each nucleus controls a specifically defined cytoplasmic area. The skeletal myofiber is a differentiated but highly plastic cell type that adapts to loading and unloading. The nuclear domain hypothesis predicts that a nucleus controls a defined volume of cellular territory in each myofiber. Therefore, addition of extra nuclei (from satellite cells) into the myofiber is required to support the increment of cell size to achieve muscle hypertrophy, and removal of the myonuclei is needed to allow the muscle to atrophy. If fewer nuclei are available, less cytoplasmic area could be supported. Generally, there is a tight relationship between nuclear number and muscle fiber cross-sectional area and volume. Nevertheless, this relationship is not perfect because the nuclear domain increases with age (*i.e.*, fewer nuclei/cytoplasm area). With age, there is a loss of satellite cells, which reduces the muscle's ability to replace nuclei (4,5) that are eliminated (likely by apoptosis). This results in a somewhat transient increase in the nuclear domain with aging, but the excessive domain size triggers fiber atrophy (4), which in turn restores the original nuclear domain size but also contributes to sarcopenia (Fig. 1B).

NUCLEAR APOPTOSIS IN SATELLITE CELLS

Satellite cells are a population of proliferative myoblast cells that are normally located between the basal lamina and plasmalemma of a myofiber and are mitotically quiescent under the nonstimulated conditions. Because myonuclei are postmitotic and incapable of undergoing cell division, muscle satellite cells provide the only known important source for adding new nuclei. Once satellite cells are activated, they proliferate and fuse into the existing myofibers in response to muscle loading. This results in an increase in the number of nuclei in the myofiber, thereby permitting fiber hypertrophy. Inactivation of satellite cell proliferation can prevent or lessen the hypertrophic response to muscle loading. Muscle atrophy caused by chronic disuse/unloading or certain cachexia-causing diseases is associated with the reduction of the existing myonuclei in muscle cell (*i.e.*, decrease in the number of myonuclei) (1,17,27). This is consistent with the nuclear apoptosis hypothesis in skeletal muscle, and it provides an explanation for the reduction of myonuclei number in atrophying myofibers. This idea is substantiated by data showing that some myonuclei undergo apoptosis in many muscle-wasting conditions including muscle disuse/unloading/unweighting (9,14,17,27) and aging-associated sarcopenia (13,14,17).

The depletion of the number and the decline in the proliferative potential of muscle satellite cells contribute to the impairment of muscle regenerative capacity and reduction of contractile function in muscular degenerative situations (*e.g.*, denervation, aging, Duchenne muscular dystrophy, and unloading-induced muscle atrophy). This loss of satellite cells has been attributed to aging-induced susceptibility to nuclear apoptosis, and this is consistent with the age-associated loss of mass and decline in muscle function.

SELECTIVE LOSS OF RECENTLY ACTIVATED SATELLITE CELLS WITH UNLOADING AND AGING

Satellite cells isolated from old rats have a greater response to proapoptotic agents by increasing nuclear apoptosis and caspases when compared with young rats (11). Furthermore, the age-related decrement in muscle recovery after stretch-shortening–induced injury in rodents has also been shown to be associated with the increase in satellite cell apoptosis (12). We have recently examined the susceptibility of satellite cells to nuclear apoptosis in aging muscles during unloading (27). In this study, one wing of young and aged Japanese quails were loaded for 14 d to induce hypertrophy, then the weight was removed for 7 or 14 d to induce muscle atrophy. The contralateral wing served as the intra-animal control for these studies. A time-released bromodeoxyuridine (BrdU) pellet was implanted subcutaneously with wing weighting to identify activated satellite cells/muscle precursor cells throughout the experimental period. The BrdU-positive nuclei were found in all unloaded muscles from both age groups, but the number of BrdU-positive nuclei relative to the total nuclei decreased after 14 d of unloading as compared with 7 d of unloading. The number of apoptotic nuclei as determined by an index of DNA fragmentation by TdT-mediated dUTP nick end labeling (TUNEL) was higher after 7 d of unloading in both young and aged muscles and after 14 d of unloading in aged muscles. Immunofluorescent staining revealed that almost all of the TUNEL-positive nuclei were also BrdU immunopositive, suggesting that activated satellite cell nuclei (both fused and unfused) underwent nuclear apoptosis during unloading. There were significant correlations among apoptotic signaling proteins B cell leukemia/lymphoma-2 (Bcl-2), BCL-2 associate-X protein (Bax), apoptosis-inducing factor (AIF), and TUNEL index. Our data are consistent with the hypothesis that nuclear apoptosis regulates, at least in part, unloading-induced muscle atrophy and loss of activated

satellite cell nuclei in previously loaded muscles. Moreover, these data suggest that aging increases satellite cell susceptibility to nuclear apoptosis during unloading after hypertrophy in skeletal myocytes (27).

Our hypothesis for the involvement of nuclear apoptosis and its importance in sarcopenia is summarized in Figure 2. We propose that aging results in targeted nuclear apoptosis in skeletal muscles, and the loss of nuclei, in turn, increases the cytoplasm volume per myonucleus. Fibers undergo atrophy to reduce the fiber cytoplasm and reestablish the original cytoplasm fiber volume per nucleus (4). Additional targeted nuclear apoptosis occurs as part of aging, and this continues to perpetuate fiber atrophy resulting in sarcopenia. Overload and resistance types of exercise will provide a stimulus to induce proliferation of satellite cells of aged muscles (albeit at a lower extent than in muscles of young animals), thereby decreasing the cytoplasm volume per nucleus. The fibers hypertrophy to reestablish the original cytoplasm volume per nucleus in each fiber, and this hypertrophic adaptation will partly offset sarcopenia (although sarcopenia cannot be completely halted). If the resistance exercise/loading is discontinued, the most recently activated satellite cells are the first ones that are targeted for nuclear apoptosis (27). The elimination of nuclei increases the muscle fiber cytoplasm volume per nucleus, and the fiber reestablishes its original cytoplasm volume per nucleus by inducing atrophy (4).

APOPTOSIS SIGNALING IN AGING SKELETAL MUSCLE

There are three general classes of apoptotic signaling pathways: mitochondria-, death receptor– and calcium-mediated pathways. These pathways seem to be well conserved across cell types including skeletal muscle cells. Although the exact physiologic function of nuclear apoptosis in skeletal myofibers is not fully understood, the data support a role for nuclear apoptosis in mature skeletal myofibers under various muscle wasting conditions including hindlimb unweighting, muscle disuse/unloading, muscle denervation, and sarcopenia (18,20,27–29).

Several lines of evidence have suggested that aging increases proapoptotic signaling in skeletal muscle. For example, basal levels of apoptotic proteins, inhibitor of

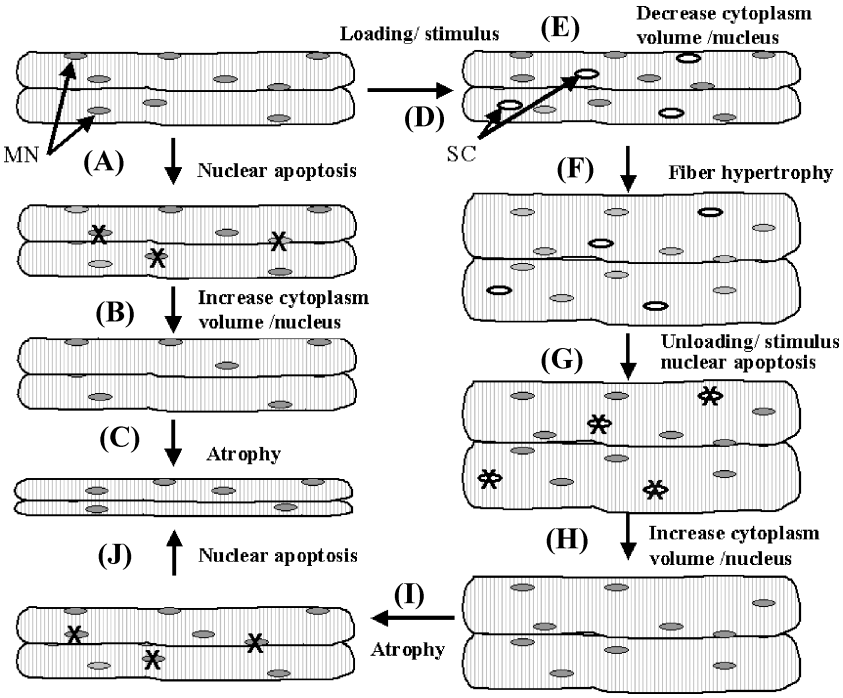

Figure 2. Involvement of nuclear apoptosis in sarcopenia during loading and unloading (A–C). Our hypothesis is that apoptotic signaling targets specific MN in multinucleated muscle fiber cells (nuclei targeted for apoptosis are indicated by an ''X'') without inflammation or disturbing adjacent proteins or organelles in aging (A). Elimination of nuclei results in a decrease in the total number of nuclei available to sustain the cytoplasmic volume, and this increases the cytoplasmic volume per nucleus (B). The muscle fiber undergoes atrophy to accommodate the fewer available nuclei for transcriptional control. This is an attempt to reestablish the original cytoplasmic volume per nucleus (C). If a sufficient number of nuclei are targeted for death in the multinucleated muscle fiber, the entire cell will be eliminated (D–F). Sarcopenia can be partially offset by exercise and muscle loading (*e.g.*, by resistance exercise), such that loading of the original muscle fibers results in a stimulus that (D) results in a proliferation of SC, which increases the number of nuclei and reduces the fiber cytoplasm volume per nucleus (E). However, proliferation of satellite cells is largely attenuated in aging compared with young muscles. The decreased cytoplasmic volume–to–nucleus ratio provides a stimulus to induce fiber hypertrophy in an attempt to reestablish the original cytoplasmic volume per nucleus ratio (F). As aged muscle fibers have fewer total nuclei than young muscles, aged muscle fibers cannot hypertrophy to the same absolute size as in young muscle (G–J). If the hypertrophied muscle is now unloaded (*e.g.*, stop exercise training, prolonged bed rest, etc.), nuclei are targeted for apoptosis, but nuclear apoptosis occurs preferentially (at least initially) in the most recently activated satellite cells (G). This reduces the total number of nuclei and increases cytoplasmic volume per nucleus ratio (H). Fibers atrophy to reestablish the original cytoplasmic volume per nucleus ratio, but fiber atrophy is more pronounced in aging (I). Nuclear apoptosis continues to target nuclei (J) as a result of an aging-induced apoptotic signaling environment, and this further exacerbates muscle fiber atrophy (C), which compounds muscle loss associated with sarcopenia. MN indicates myonuclei; SC, satellite cells.

differentiation 2 (Id2) and Bax, are greater in muscles of aged rats as compared with young adult rodents (2,17,26,28). We do not know if Id2 directly affects the proapoptotic Bax protein levels or if it regulates apoptosis independently of Bax. Song *et al.* (30) reported a significant increase in Bax levels and a decrease in the antiapoptotic Bcl-2 in the gastrocnemius and soleus muscles of old sedentary rats. The Bax/Bcl-2 ratio may be very important in predicting the muscles most likely to undergo aging-associated muscle loss because an increased Bax/Bcl-2 ratio is found in aging muscles, and this is associated with increased levels of caspase 3 and enhanced DNA fragmentation.

Some studies have shown that cytochrome *c* levels in nonmitochondrial protein fractions are greater in muscles of old animals (22). This is important because mitochondria can release cytochrome *c*, which acts in a proapoptotic role by binding 2′-deoxyadenosine 5′-triphosphate (dATP) and apoptosis protease activating factor-1 (Apaf-1), forming an apoptosome that cleaves caspase 9. Aging-associated increases in proapoptotic Apaf-1 (7,8,26) and proapoptotic cleaved caspase 9 levels (7,26) have been reported in the gastrocnemius muscle, and this coincided with increased DNA fragmentation in gastrocnemius muscles of old rats (7,26). Although Chung and Ng (7) and Dirks and Leeuwenburgh (8) did not find greater basal levels of cytochrome *c* in muscles of old rodents, Siu and colleagues (26) found increased levels of cytochrome *c* in the gastrocnemius muscle of aged rats. The discrepancy in the reports of apoptotic signaling may be caused in part by age-related adaptations in antiapoptotic proteins in an attempt to reduce apoptotic involvement in skeletal muscle.

Models of muscle wasting provide a means to examine the effects of aging on apoptotic signaling, and these provide greater insight into potential mechanisms for sarcopenia. The general age-associated differences in apoptotic signaling in response to muscle disuse, reduced loading, or denervation are shown in Figure 3. Using a wing unloading model to induce muscle atrophy in birds, we have shown a significant reduction in Bcl-2 in both young and old birds after 7 d of unloading (27). In contrast, hind limb unloading in rats seemed to result in a compensatory increase in Bcl-2 in muscles of both young and old animals presumably in an attempt to reduce the consequences of the suspension-induced increase in Bax protein (17,26). Unloading increases proapoptotic Bax levels and cytochrome *c* release in muscles of both young and old animals, but there is a greater increase in muscles from aged rodents (17,26). Interestingly, hind limb unloading in rats increases Apaf1 and Id2 in aged, but not young, animals (17,28). Hind limb unweighting increases the proapoptotic proteins AIF, Endo G, and mitochondrial second mitochondria–derived activator of caspase/direct IAP protein with low pI (Smac/DIABLO) in muscles of old, but not young, animals (9,14,17). Although unloading does not change AIF in young birds, it increases nuclear AIF in muscles of old birds (27), and also increases AIF and Smac/DIABLO in muscles of old rats (26). Caspase 9 and caspase 3 increase in muscles of both young and old animals in some, but not all, models of muscle wasting. Aging increases the release of the proapoptotic protein Endo G and under conditions of unloading in aging muscles (14).

Denervation-induced atrophy is also relevant to the study of sarcopenia because some of the loss of skeletal muscle is thought to be caused by a loss of motor units and therefore loss of innervation. Interestingly, denervated rodent muscle has a decrease in antiapoptotic Bcl-2, and an increase in proapoptotic Bax, mitochondrial release of cytochrome *c*,

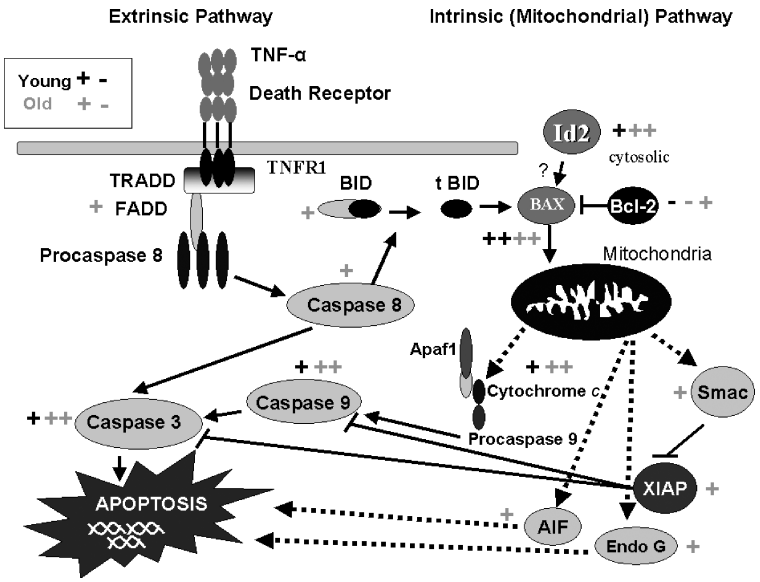

Figure 3. Apoptotic signaling in aging muscles. This figure summarizes the extrinsic death domain pathway and intrinsic pathway for mitochondrial apoptosis in aging muscles under conditions of muscle loss (*e.g.*, unloading, denervation). The increase (+) and the decrease (-) in the direction of the change in apoptotic signaling in response to muscle wasting insults for proteins in these pathways are indicated in muscles from young adult (black) and old (gray) animals. Double positive (++) or double negative (−−) signs indicate a large increase or decrease in protein levels, respectively. Proapoptotic pathways are shown by lines and arrows. A line with a blunt end indicates an antiapoptotic influence. Dotted lines show examples of proapoptotic proteins that are released from the mitochondria in response to muscle wasting, which can either directly (*e.g.*, AIF) or indirectly (*e.g.*, cytochrome *c*) induce nuclear apoptosis in muscle cells. TRADD, tumor necrosis factor receptor 1 associated death domain.

Smac/DIABLO, and AIF, and an up-regulation of caspase 3 and caspase 9 mRNA, active protein fragment and protease enzymatic activity, and a reduction of the antiapoptotic protein X-linked inhibitor of apoptosis protein (XIAP) (21,23). Eliminating Bax offsets much of the apoptosis-associated muscle loss in denervation (23).

Together, these data suggest that compared with muscles of young animals, aging muscles have a proapoptotic environment, which is exacerbated when the muscles receive some atrophy-inducing signal. There does seem to be an attempt by muscles of old animals to offset the higher levels of apoptotic signaling associated with aging and amplified by muscle wasting. For example, antiapoptotic XIAP functions to inhibit proapoptotic caspase 9. XIAP is higher in both control and unloaded muscles of old rats compared with young animals (29). Interestingly, aging muscles have increased levels of XIAP during unloading compared with muscles of young animals (29). This may be an unsuccessful attempt by aging muscles to reduce proapoptotic signaling during muscle disuse because countering the increase in this antiapoptotic protein is an aging-induced increase in proapoptotic Smac/DIABLO during muscle unloading, which acts to inhibit XIAP.

Although more work has been done on the mitochondrial-dependent apoptotic signaling pathways in aging, we have recently shown that the death receptor pathway is also activated in muscles of aging rodents, and there is cross-talk between the death receptor and intrinsic (mitochondrial) apoptotic signaling pathways in aging muscles (16). Tumor necrosis factor-α (TNF-α) and FasL activate the death receptor apoptotic pathway, which subsequently activates caspase 8. Muscles from aged rats have higher type I TNF receptor and Fas-associated death domain protein (FADD) messenger RNA (mRNA), protein contents for FADD, BCL-2 Interacting Domain (BID), FLICE-inhibitory protein, and enzymatic activities of caspase 8 and caspase 3 than muscles from young adult rats. These data demonstrate that proapoptotic signaling downstream of the TNF receptor is active in aged muscles (see Fig. 3).

EXERCISE AND LOADING INDUCED DECREASES IN APOPTOTIC SIGNALING

Exercise and overload may partially offset apoptotic signaling in skeletal muscle. Song *et al.* (30) have shown that 12 wk of treadmill exercise training significantly reduced Bax expression in both gastrocnemius and soleus muscles of senescent animals and increased the expression of Bcl-2 in the same muscles. These changes were accompanied by a reduction in DNA fragmentation (30). These findings are consistent with data from Siu *et al.* (24) who used endurance training for 5 d weekly for 8 wk in young rats. Bax mRNA content was reduced, and Bcl-2 protein content was increased in the soleus muscle of the exercised animals compared with the sedentary group. Furthermore, aerobic exercise was also able to increase the myocytes' expression of the antiapoptotic protein, apoptosis repressor with caspase recruitment domain and XIAP, which can bind to caspase 3 and inhibit its proteolytic activity. Our laboratory has also shown an elevation of XIAP content in the patagialis muscle of old quails after 14 d of stretch-induced overload (29). Furthermore, we have found a decreased Bax expression in slow-tonic anterior latissimus dorsi muscles of young quails and an increased Bcl-2 content in both young and aged birds after 21 d of stretch-induced muscle loading, concomitant with a greater expression of heat shock protein 27 (HSP27) and HSP72 only in the young group (22). Taken together, these findings suggest that exercise and muscle overload even at old age is able to at least partially offset muscle loss via an attenuation of the proapoptotic signaling potential. This provides the possibility that exercise may delay the onset and/or attenuate the rate of progression of sarcopenia by reducing the proapoptotic environment of old muscles (3,19,22,24,25,29).

CONCLUSIONS AND FUTURE PERSPECTIVES

The data suggest that proapoptotic signaling is increased in muscles of old animals. This apoptotic environment includes increased signaling for both intrinsic and extrinsic pathways. The aged muscle consists of an environment that is not conducive for nuclear survival, and it is likely that some nuclei that are activated as part of a loading/exercise paradigm may undergo selective apoptosis (nuclear apoptosis). This would result in fewer new nuclei that are added to the muscle in response to a hypertrophic stimulus, and therefore less overall hypertrophy could be obtained because this process is nuclei dependent.

Periods of unloading exacerbate apoptotic signaling in aging muscles. However, there seems to be an attempt in aging muscles to reduce the effect of increased proapoptotic proteins by increasing the level of antiapoptotic proteins during periods of muscle disuse. Adaptations to exercise or overload provides an environment that is less apoptotic. This would be expected to promote satellite cell survival so they are able to contribute to muscle hypertrophy and repair. In this way, muscles of old animals could adapt to increased loading (*e.g.*, hypertrophy). We hypothesize that muscles in young animals obtain greater hypertrophy with the same stimulus as compared with muscles of old animals in part because the basal levels of proapoptotic proteins is low in muscles of young animals as compared with old animals.

Understanding the manner in which selective nuclei are targeted for death while others are not will provide important new information for future studies. Furthermore, understanding why recently activated satellite cells seem to be most vulnerable and preferential targets for apoptotic death during muscle inactivity will provide insight into new strategies for understanding muscle remodeling in aging. Identifying the upstream-regulatory pathways leading to nuclear apoptosis may lead to novel preventive or therapeutic regimens to protect satellite cells so that they can contribute to hypertrophic growth and/or reduce or delay sarcopenic muscle loss. When implemented, the appropriate interventions should offset the apoptotic aging-induced net loss of muscle nuclei, reduce and/or slow sarcopenia, and improve muscle function in the elderly.

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