Send Orders for Reprints to reprints@benthamscience.ae

356 *Current Genomics,* **2018**, *19,* 356-369

REVIEW ARTICLE

Molecular Insights into Muscle Homeostasis, Atrophy and Wasting

Bianca Maria Scicchitano[1,*], Gabriella Dobrowolny[2], Gigliola Sica[1] and Antonio Musarò[2,3,*]

[1]*Istituto di Istologia e Embriologia, Università Cattolica del Sacro Cuore, Fondazione Policlinico Universitario Agostino Gemelli, Largo Francesco Vito 1-00168, Roma, Italy;* [2]*Center for Life Nano Science@Sapienza, Istituto Italiano di Tecnologia, Rome, Italy;* [3]*DAHFMO-Unit of Histology and Medical Embryology, Sapienza University of Rome, Laboratory Affiliated to Istituto Pasteur Italia - Fondazione Cenci Bolognetti, Rome, Italy*

ARTICLE HISTORY

Received: October 03, 2017
Revised: December 04, 2017
Accepted: December 20, 2017

DOI:
10.2174/1389202919666180101153911

Abstract: Muscle homeostasis is guaranteed by a delicate balance between synthesis and degradation of cell proteins and its alteration leads to muscle wasting and diseases.

In this review, we describe the major anabolic pathways that are involved in muscle growth and homeostasis and the proteolytic systems that are over-activated in muscle pathologies. Modulation of these pathways comprises an attractive target for drug intervention.

Keywords: Muscle homeostasis, Muscle atrophy, Degradation of cell proteins, Muscle wasting, Anabolic pathways, Muscle pathologies.

1. INTRODUCTION

Skeletal muscle represents one of the most plastic tissues of our body. The presence of heterogeneous population of myofibers confers to skeletal muscle a high degree of plasticity necessary to modulate its morpho-functional properties in response to different external stimuli, such as exercise, variation in hormone levels, oxygen and nutrient supply, and activity of motor neurons. However, this important and critical feature of skeletal muscle is impaired in senescent muscle and severely compromised in different diseases, leading to alteration in the functional performance and in the regenerative capacity of skeletal muscle.

Among different signaling, Calcium (Ca^{2+}) controls, through the activation of so-called "toolkit" [1], numerous cellular processes, such as proliferation, cell growth, differentiation, and gene transcription. Defects in signaling "toolkit" can activate proteolytic systems, leading to muscle wasting [2, 3].

2. MUSCLE HOMEOSTASIS: A MATTER OF BALANCE BETWEEN PROTEIN SYNTHESIS AND DEGRADATION

The morpho-functional properties of skeletal muscle are guaranteed by a dynamic turnover between net protein synthesis and degradation and by cell turnover.

*Address correspondence to these authors at the Unit of Histology and Medical Embryology, *Via* A. Scarpa 14, Rome 00161, Italy;
Tel: +390649766956; E-mail: antonio.musaro@uniroma1.it and Institute of Histology and Embryology, Catholic University, Largo Francesco Vito 1, Roma 00168, Italy; Tel/Fax: +390630154463/+39063053261;
E-mail: biancamaria.scicchitano@unicatt.it

The prevalence of protein synthesis leads to muscle growth and hypertrophy, whereas the reduced protein synthesis rates associated with an increase in protein degradation results in muscle atrophy and, in certain pathologic conditions, in muscle wasting. Muscle cell turnover, which is a guarantee by the activity of satellite cells, the muscle stem cells compartment, and by other precursor cells, is also another important homeostatic process of the adult skeletal muscle.

The signaling pathway that regulates protein turnover is mediated by the Insulin-like Growth Factor 1 (IGF1) and by a cascade of intracellular effectors, including Akt, which has the ability to control both protein synthesis, by activating the kinase mTOR, and protein degradation by repressing the transcription factor FoxO [4, 5].

3. IGF-1

IGF-1 exists in different isoforms, due to alternative splicing of the initial transcript. The different isoforms can exert different roles and can act as a systemic growth factor or alternatively in a autocrine/paracrine manner [6-8].

At molecular level, human and rodent IGF-1 gene contains six exons, separated by five introns. In rodent, Exons 1 and 2 represent leader exons and encode distinct 5′UTRs, as well as different parts of the signal peptide. Exon 3 encodes part of the signal peptide as well as part of the mature IGF-1 peptide. Exon 4 encodes the rest of the mature peptide and part of the amino-terminal region of the E-peptide. Exons 5 and 6 encode two distinct carboxy-terminal E-peptides, namely Ea and Eb, and the 3′UTR.

In humans, similarly to rodent IGF-1 gene, Exons 1 and 2 represent leader exons; however, differently to murine IGF-1

gene, three mRNA variants with alternatively spliced-end have been identified [6]. Splicing of human exon 4 to exon 6 yields an mRNA sequence which encodes the Ea-peptide, shares 91% homology with the mouse Ea-peptide. The second mRNA derives from an exon 4-5 splice variant, which encodes an extension peptide termed Eb-peptide. The third mRNA splice variant, termed as Ec-peptide, contains exon 4, part of exon 5 and exon 6. This human Ec-peptide shares 73% homology with rat Eb-peptide and is considered to be its counterpart [9].

Whether different IGF-1 isoforms exert different biological functions is still an open question.

Gene expression analysis performed in muscle of both human and wild type mice revealed that IGF-1Ea results are more highly expressed as compared to both IGF-1Eb and IGF-1Ec isoforms, while IGF-1Eb appears to be more expressed than IGF-1Ec isoform [6]. IGF-1Ea promotes satellites cell differentiation and provides most of the mature IGF-1 for stimulating protein synthesis. IGF-1Eb isoform can be responsible for satellite cells activation and proliferation and can be induced by physical exercise and stretch [10].

Previous work demonstrated that muscle restriction of IGF-1Ea (mIGF-1) transgene (MLC/mIGF-1) expression sustained muscle hypertrophy and regeneration in young and senescent skeletal muscle [11], enhanced the recruitment of circulating stem cells in injured muscle [12] and counteracted muscle wasting in mdx dystrophic mice [13, 14] and in neuromuscular disease models, such as Amyotrophic Lateral Sclerosis (ALS) [15, 16] Spinal and Bulbar Muscular Atrophy (SBMA) [17] and Spinal Muscular Atrophy [18].

Additional work is necessary to define the role of the different IGF-1 isoforms in the physiopathology of skeletal muscle and to characterize the specific signal transduction pathways that might be activated by each isoform.

4. THE DOWNSTREAM SIGNALING OF IGF-1

Unlike other growth factors, IGF-1, through the activation of different pathways [19] and depending which E-peptide is expressed, can stimulate processes mutually exclusive, such as proliferation, differentiation and hypertrophy both vivo and *in vitro* [10, 11, 20-24].

The Mitogen-Activated Protein (MAP) kinase pathway basically mediates the proliferative response of muscle cells to the activity of IGF-1, whereas the kinase Akt pathway stimulates muscle differentiation and might trigger muscle hypertrophy. The activity of Akt involves the disruption of the tuberous sclerosis complex-1 and -2 proteins (Tsc1 and Tsc2) and the subsequent phosphorylation and activation of mTOR (Fig. **1**). Then, mTOR phosphorylates and activates the 70-kDa ribosomal protein S6 kinase (p70S6K) (Fig. **1**) and inhibits 4E-BP, a negative regulator of the protein initiation factor eIF-4E, increasing translation.

Alternatively, Akt can phosphorylate and inhibit another downstream effector target, namely glycogen synthase kinase 3 beta, (GSK3b) [25], promoting muscle hypertrophy, independently of the mTOR pathway.

In particular, mTOR exerts its role forming two functionally distinct multiprotein signaling complexes, known as mTOR Complex 1 (mTORC1) and 2 (mTORC2). At structural level, mTORC1 is formed by three components: mTOR, regulatory protein associated with mTOR (Raptor), and mammalian lethal with Sec13 protein 8 (mLST8) [26-28] (Fig. **1**). Like mTORC1, mTORC2 contains mTOR and mLST8, but differently to mTORC1, it contains Rictor (rapamycin insensitive companion of mTOR), [29, 30] and other regulatory proteins, such as DEPTOR [31], the regulatory subunits mSin1 [32-34] and Protor1/2 [35-37] (Fig. **1**). At functional level, mTORC1 is inhibited by rapamycin, whereas mTORC2 is insensitive to the activity of rapamycin.

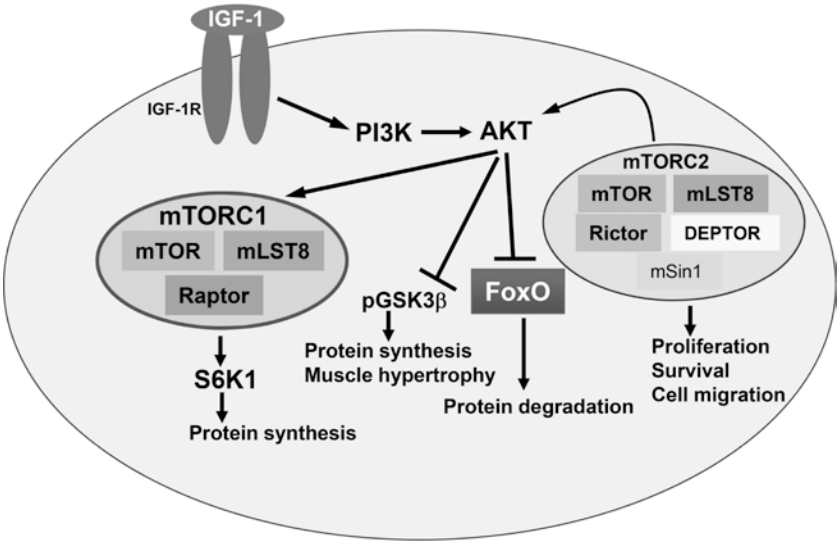

Fig. (1). A schematic representation of the effects of the Insulin-like Growth Factor 1 (IGF-1) on muscle growth and homeostasis. IGF-1 stimulates mTORC1 activity and inhibits FoXO expression through the PI3K-Akt pathway, promoting protein synthesis and counteracting protein degradation. IGF-1 induces also mTORC2 activity, along with other signal pathways, to stimulate proliferation, survival, and cell migration.

mTORC1 plays a central role in regulating several anabolic processes including protein and lipid synthesis, and ribosome biogenesis while also suppressing catabolic pathways [38]. mTORC1 directly phosphorylates S6K1 that in turn activates several substrates that promote mRNA translation initiation [39-42] (Fig. **1**). Conversely to mTORC1, which regulates cell growth and metabolism, mTORC2 controls proliferation and survival, inducing the phosphorylation and activation of Akt [43], which phosphorylates and inhibits several substrates, including the FoxO1/3a transcription factors, the metabolic regulator GSK3b, and the mTORC1 inhibitor TSC2 (Fig. **1**). Moreover, mTORC2 phosphorylates several members of the PKC family [29, 43-46], regulating cytoskeletal remodeling and cell migration.

Additional pathways that are modulated by IGF-1 are ROCK2, p90RSK and the components of the kinase cascade p42/p44 MAPK, which act in concert to promote muscle differentiation and maturation [47-50].

5. THE PROTEOLYTIC SYSTEMS OF MUSCLE WASTING

A general feature associated with several pathologic conditions such as both genetic diseases and acquired disorders (*i.e.* muscle disuse, cachexia, heart failure, sarcopenia) is the lose of adaptability of skeletal muscle leading to wasting, a process in which the delicate balance between anabolic and catabolic process is impaired [51, 52]. Nevertheless, the mechanisms underlying muscle atrophy and wasting associated with different conditions might be different. For example, PI3K/Akt/mTOR pathway is not modulated in human sarcopenic sedentary subject [6], neither physical activity, normally associated with an increased activity of Akt/mTOR/p70S6K in young individuals, is able to stimulate this anabolic pathway in sarcopenic animal models [53]. In contrast, cancer cachexia significantly reduces the rates of muscle protein synthesis.

Among proteolytic systems ubiquitin-,calpain-, caspase-, and autophagy-mediated protein and organelles degradation are the principal pathways activated in several pathologies, leading to myofiber degeneration, wasting and impaired muscle regeneration [54].

6. THE UBIQUITIN-PROTEASOME SYSTEM

The ubiquitin-proteasome system is one of the signal transduction pathways that mediates the turnover of muscle protein. It has been demonstrated that several pathologic conditions, leading to muscle wasting (*e.g.* glucocorticoid treatment, sepsis, fasting, cancer, and acidosis) [55-58] or associated with muscle atrophy [59-61] over-activate a program of gene expression in which the ubiquitin-proteasome pathway represents the major player.

The pathway involves an enzymatic cascade starting with the conjugation of protein substrates with ubiquitin and terminating with the degradation of targeted protein to small peptides and amino acids (Fig. **2**). E1 enzymes activate the ubiquitin proteins, which move from E1 to the E2 ubiquitin-conjugating enzyme. The last step of the reaction involves the ubiquitin-protein ligases E3, which recognize the protein that will be ubiquitinated (Fig. **2**). The polyubiquitineted proteins are transferred, in an ATP-dependent reaction, in the 26S proteasome complex where the proteins are degraded (Fig. **2**). The ubiquitin itself is then released and reused for a new enzymatic reaction.

Two critical players of the ubiquitin-proteasome system are the muscle E3 ubiquitin ligases atrogin-1(MAFbx) and Muscle RING Finger 1 (MuRF-1), which are up-regulated in several pathologic conditions associated with muscle atrophy [56, 62, 63]. The most critical players of muscle atrophy are the members of the transcription factor FoxO (Forkhead box O) family, which represents the pivotal transcriptional activator of MAFbx/atrogin and MuRF-1during atrophy [64]. Indeed, constitutively active FOXO3a expression or muscle-specific FOXO1 overexpression was sufficient to induce muscle atrophy [64, 65]. Of note, knockout animals in which MAFbx or MuRF1 was ableted resulted resistant to atrophy [62], whereas their overexpression produce atrophy and alteration of the thick filaments within the sarcomere structure [57]. MuRF-1 targets myofibrillar proteins [66, 67], whereas atrogin-1 has been shown to target both the muscle regulatory factor MyoD and the eukaryotic initiation factor of protein synthesis eIF-e [68, 69]. MuRF1, but not Atrogin-1, can be also transcriptionally activated by NF-kB, a critical player activated by several catabolic stimuli, including oxidative and inflammatory cytokines, promoting muscle atrophy. Of note, other factors implicated in the regulation of muscle

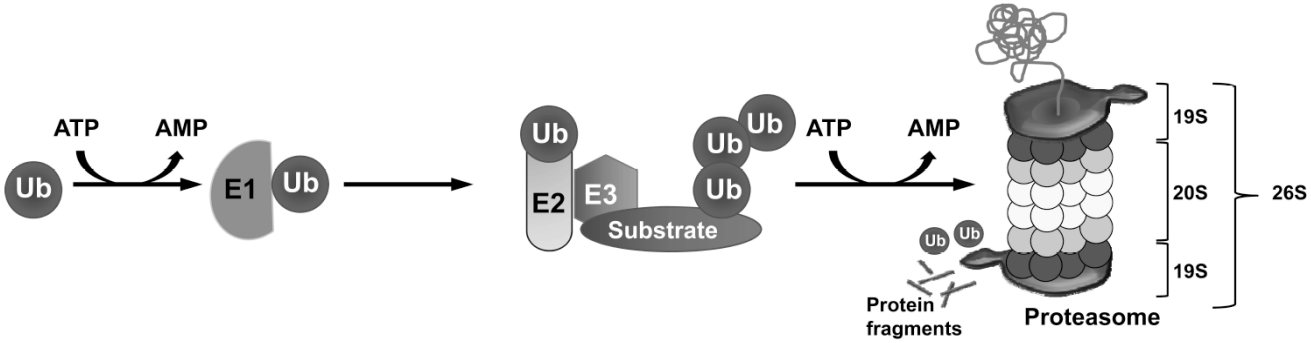

Fig. (2). A schematic representation of the ubiquitin-mediated protein degradation. The ubiquitin-activating enzyme-E1 activates ubiquitin, which is then transferred to a ubiquitin-conjugating enzyme-E2. The ubiquitin-protein ligase-E3 binds to E2 and to protein substrate which becomes polyubiquitinated and subsequently degraded by the proteasome, generating short peptides and free ubiquitin that can be further reused.

fiber size such as GSK3β, MEK and calcineurin [22, 62, 70], do not have direct roles in the regulation of atrogin-1 expression [64]. Two additional targets of FoxO activity have also been identified: Specific of Muscle Atrophy and Regulated by Transcription (SMART) and Muscle Ubiquitin ligase of SCF complex in Atrophy-1 (MUSA1) [71, 72]. MUSA1 is a novel ubiquitin ligase that plays a critical role for the induction of muscle atrophy during denervation and fasting [71, 72]. Ablation of MUSA1, by RNA interference, prevents muscle atrophy, whereas increased expression of MUSA1 exacerbates muscle loss, promoting muscle cachexia [72]. It has been also demonstrated that FoxO3 regulates another proteolytic pathways, namely autophagy (discussed below), coordinating two proteolytic systems, namely the proteasome, which is involved in removal of proteins, and the autophagy, involved in organelles clearance.

Several factors and stimuli including glucocorticoids, oxidative stress, and inflammatory cytokines might activate the ubiquitin-proteasome system [57]. Activation of the ubiquitin-proteasome system has been reported in both experimental model of cancer cachexia and human patients with advanced cancer. It has been reported that the decreased activity of signaling pathway mediated by IGF-1, associated with different model of cancer cahexia, can lead to muscle atrophy [62], *via* the activation of the "atrophy program". The reduction in IGF-1 expression and activity results in the inhibition of the AKT [73, 74]. Under anabolic conditions, AKT blocks, by phosphorylation and subsequent sequestration in the cytoplasm, the function of the FoxO transcription factors family [75]. Reduction in anabolic factors, including IGF-1, causes the inhibition of the PI3K/AKT pathway. In such conditions, the dephosphorylated active form of FoxO factors entering the nucleus which transactivate the expression of atrogin-1 and MuRF1 [64]. The ubiquitin-proteasome system can be also activated, although there are conflicting findings, in aging-related muscle wasting, a state known as sarcopenia [76, 77]. It has been demonstrated that the decline in muscle mass during aging is associated with a reduction in IGF-1/AKT pathway with a consequent activation of atrogin-1 and MuRF1 [7, 11, 78-82]. Nevertheless, due to different discrepancies among different studies [78, 83], the involvement of MuRF1 and atrogin-1 in aging related muscle loss require additional study. Other chronic diseases such as Chronic Obstructive Pulmonary Disease (COPD), diabetes, Chronic Kidney Disease (CKD), and Congestive Heart Failure (CHF) impinge the IGF-1 signaling and activate the ubiquitin-proteasome system [61, 63, 84-87]. Of note, a common feature of these complex diseases is the increase in angiotensin II (Ang II), which promotes muscle wasting through multiple mechan [87]. AngII can: i) reduce IGF-1 expression and increase cytokine signaling such as glucocorticoid and IL-6, resulting in increased protein breakdown; ii) increase oxidative stress; iii) inhibit AMPK, impinging energy balance; iv) inhibit satellite cell function and muscle regeneration.

7. THE CALPAIN PATHWAY

Calpains, which have been identified in many organisms [88], are calcium-activated cysteine proteases characterized by the presence of two subunits, a large subunit of 80kDa that contains protease activity and a smaller subunit of 30kDa, functioning as a regulator of calpain activity [89]. Based on Ca^{2+} concentration dependence, two ubiquitous calpain isoforms, μ and m, are well characterized, and are commonly referred as the ubiquitous/conventional/typical calpains. Among the different subtypes, calpain 3 is predominantly expressed in skeletal muscle and it maintains proteolytic activity at physiological Ca^{2+} level [89]. More recently, it has been demonstrated that calmodulin (CaM), a known transducer of the calcium signal, binds and facilitates Calpain 3 autolytic activation in skeletal muscle [90]. At physiological levels, calpains participate in many cellular processes, including cytoskeletal remodeling [91], cell mobility [92], myofibril maintenance [93], signal transduction [94], cell cycle progression [95], regulation of gene expression [96] apoptosis [88], and long term potentiation [97].

Calpains are also activated by several stimuli affecting Ca^{2+} homeostasis, causing sarcomeric alterations [98], mitochondrial swelling, sarcoplasmic reticulum vacuolization [99, 100], disruption of the contractile tissue [101], and muscle atrophy [102-104] (Fig. **3**).

Calpains are generally localized at the level of the Z disk of the sarcomere [105] where they initiate the proteolytic cleavage of muscle proteins that anchor the sarcomere, including titin, nebulin, desmin, filamin, troponin and tropomyosin, [106, 107]. Another molecular target of calpains is fodrin, the non-erythroid spectrin protein, whose alteration impinges muscle physiology and myofibers survival [106-108].

The activity of calpains is regulated by the endogenous inhibitor calpastatin, which prevents both enzyme activation and expression of catalytic activity, by modulating intracellular Ca^{2+} concentration [109, 110] (Fig. **3**).

Of note calpains, conversely to other proteolytic systems, cleave target proteins at specific sites, leaving large polypeptide fragments with altered physiological properties. Moreover, the calpains target proteins can be "marked" for degradation by post-translational modifications, such as phosphorylation [111].

The activity of calpains can play also a role in muscle injury under physiological, such as exercise, [112] or pathologic, such as Duchenne and Becker muscular dystrophies (DMD/BMD), conditions [113]. Of interest is the observation that calpain3 is associated with limb-girdle muscular dystrophy type 2A [114], and might control myoblast fusion and maturation, suggesting a role in muscle function [115]. More recently, two sarcomeric tropomodulin (Tmod) isoforms, Tmod1 and Tmod4, have been characterized, which represent novel proteolytic targets of m-calpain [116]. The levels of m-calpain resulted elevated in dystrophic soleus muscle of mdx mice and were associated with loss of Tmod1 from the thin filament pointed ends, suggesting that Tmod proteolysis, by m-calpain, represents a novel mechanisms that may contribute to DMD pathology [116]. In other reports, the catabolic activity of calpains, along with the ubiquitin-proteasome pathway, has been associated with muscle atrophy induced by chronic hypobaric hypoxia and with sarcopenia [117]. Sarcopenia is a pathologic consequence of aging, associated with loss of muscle mass and

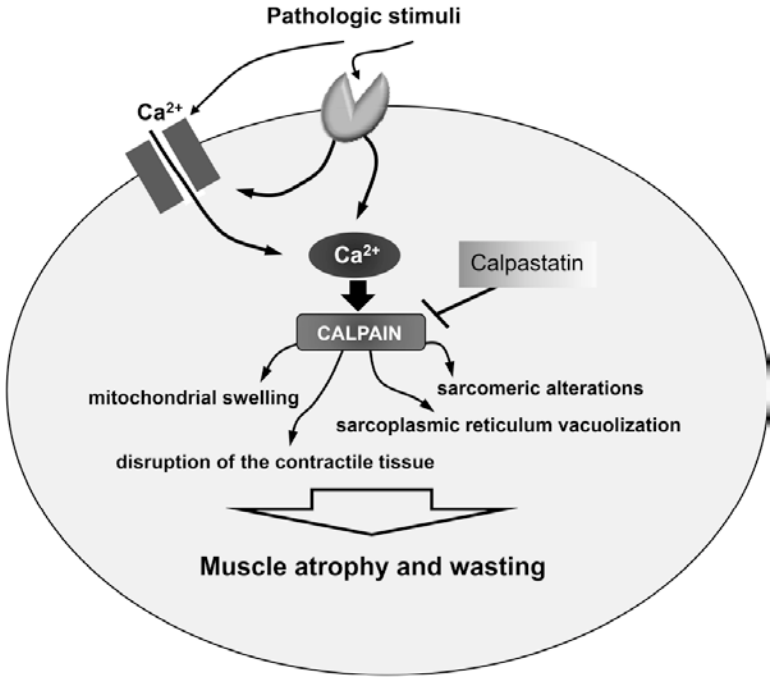

Fig. (3). Schematic representation of the calpain-mediated muscle atrophy and wasting. Calpains are Ca^{2+}-activated cysteine proteases, which might target different intracellular components, such as mitochondria, sarcoplasmic reticulum, myofilaments, leading to muscle atrophy and wasting. Calpastatin represents the negative regulator of calpain activity.

function. It has been demonstrated that muscle-specific overexpression of calpastatin greatly reduces sarcopenia [118].

Other reports provided additional insights into the pathogenic role of calpain in sarcopenia. One of the leading mechanistic theories for aging is the oxidative damage hypothesis, based also on the evidences that age-related changes accelerate with elevated oxidative stress [119, 120]. It has been demonstrated that calpains activate the degradation of oxidated myofibrillar proteins [121], suggesting a mechanistic link between oxidative stress and proteolysis [121, 122]. In addition, it has been reported that the reduced calpain activity, by forced expression of a calpastatin transgene, attenuates muscle wasting and prevents the shift from slow to fast fiber type in muscle unloading [123]. Another potential target of calpain is Akt *via* the chaperone heat shock protein 90 (HSP90), which under physiological conditions protects phosphorylated active form of Akt. It has been demonstrated that calpain activation reduces HSP90-client proteins binding, thus reducing Akt activation [124]. Calpain activation might also regulate other factors and pathways, such as STAT, NF-kB, PKC, calcineurin, Cdc42/RhoA leading to muscle atrophy and wasting [125-130]. More recently, it has been demonstrated that calpain activity increased, while calpastatin expression is reduced in the muscle of experimental models of cancer cachexia, namely AH-130 hepatoma and the C26 colon carcinoma [131]. Nevertheless, a specific inhibition of Ca^{2+}-dependent proteases did not block muscle wasting [131], suggesting that i) loss of myofibrillar proteins in muscle wasting requires the synergistic action of at least two proteolytic systems and ii) targeting the catabolic stimuli rather than the proteolytic activity of a single pathway would likely be the most efficient therapeutic approach for complex diseases as cancer cachexia.

Collectively these studies indicate that calpains exert critical role in both muscle homeostasis and diseases and represent potential therapeutic targets.

8. THE CASPASE PROTEOLYTIC SYSTEM

Another proteolytic system dysregulated in several diseases, is represented by Caspases [132, 133] which comprise two major groups, namely the initiator caspases and the effector caspases.

Caspase-2, -8, -9 and -10 belong to the initiator caspases and as such initiate the proteolitic cascade, whereas caspases-3, -6 and -7 are members of the effector caspases, which are activated by initiator caspases and are responsible for the proteolytic cleavage of a broad spectrum of cellular targets, which ultimately lead to cell death [132] (Fig. **4**).

Caspases can be activated by either extrinsic death stimuli, in which specific ligands triggers a cascade of events leading first to the activation of initiator caspases, or alternatively by intrinsic death stimuli inducing mitochondrial release of cytochrome c (Fig. **4**), which binding to Apaf1 leads to the formation of the apoptosome [132, 133].

A key role of caspases is to inactivate proteins that protect living cells from apoptosis. Caspases can stimulate the caspase-activated deoxyribonuclease (CAD), leading to DNA fragmentation [134, 135]. Caspases can also cleave structural proteins such as lamins, promoting nuclear lamina destruction and chromatin condensation [136, 137], desmin, generating a small product (N-desmin) that activates apoptotic pathways [138], gelsolin, altering cell motility and morphology, β-catenin, altering cell-cell contacts and cell-matrix focal adhesions, DNA repair, mRNA splicing, and DNA replication [133, 139, 140].

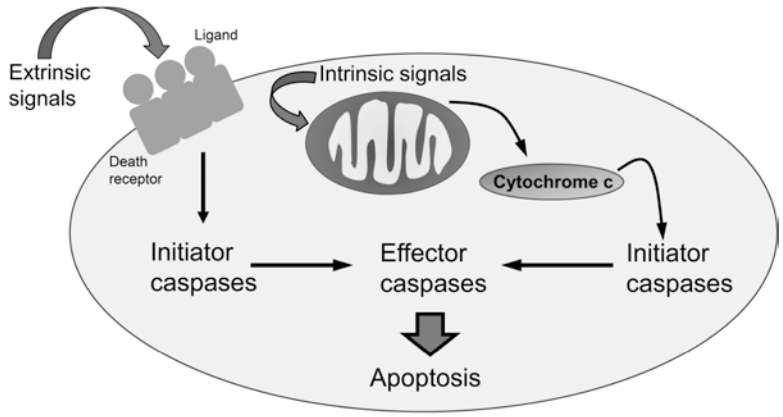

Fig. (4). Schematic representation of the caspase-mediated apoptosis. Caspases can be activated by either extrinsic or intrinsic death stimuli. Extrinsic death signals activate the initiator caspases, which in turn induce the effector caspases, promoting the proteolytic cleavage of cellular targets. Intrinsic death stimuli induce mitochondrial damage, resulting in the cytosolic release of cytochrome c that binding to Apaf1, resulting in the activation of initiator caspases and then to the induction of effector caspases and subsequently to cell death.

The activation of apoptotic pathway by Caspase activity is also associated with different muscular diseases, including Duchenne and facioscapulo-humeral dystrophies [141]. One of the proposed mechanisms that mediates the activation of caspase pathway in muscular dystrophy is the Endoplasmic Reticulum (ER) stress [142-145] that activates the initiator caspases-12 and-4 and in turn the caspase-9 and 3, leading to apoptosis [146-150].

Activated caspase-12 can also carry out apoptotic events directly into the nucleus [151], whereas deleting caspase-12 preserves muscle function in the mdx dystrophic mouse model, resulting in improved force generation [152].

Caspases are also involved in cancer-associated cachexia and in the inhibition of muscle differentiation [153].

The inflammatory cytokines Tumor Necrosis Factor-alpha (TNFα) and other factors such as IL-6, oncostatin M, leukemia inhibitory factor, and ciliary neurotrophic factor can activate and amplify caspase activity, leading to altered muscle differentiation [154-158] and cancer cachexia [159-163].

Pharmacological and/or genetic inhibition of intracellular mediator of the inflammatory cytokines counteract cancer-associated cachexia, age-induced sarcopenia, and muscular dystrophy, stimulating muscle regeneration [159, 160, 164-170].

Of note, caspases activity has been also associated to different cell functions, which are independent of inducing apoptosis [171]. Caspase activity is involved, along with the ubiquitin protesome system, in the modulation of satellite cell activity during muscle growth and regeneration, promoting Pax7 degradation; thus guaranteeing the acquisition of a differentiation competent state [172, 173]. The opposite and mutually exclusive effects exerted by caspases, namely death and non-death caspase function, depend on the duration of the signaling cascade, where cell death is characterized by sustained caspase activity, whereas non-death function is guaranteed by a transient activity responses [172, 174-176].

Moreover, increased proteolysis can be achieved by synergic activity of different proteolytic system, such as caspase and ubiquitin-proteasome [177].

9. THE AUTOPHAGIC PATHWAY

Autophagy is an important homeostatic mechanism for all eukaryotic organisms and modulates different processes such as proliferation, cell death, differentiation during both embryogenesis and postnatal development [178]. The activation of autophagy involves several distinct steps, namely i) induction, ii) phagophore formation, involving donor double-membrane structures, iii) autophagosome formation and sequestration of cellular components, iv) autophagosome-lysosome fusion, v) degradation of sequestered components (Fig. **5**).

Critical factors in autophagosome formation are the small ubiquitin-like molecules, such as LC3, which are transferred to isolation membranes to trigger their growth [179-182] (Fig. **5**).

Basal level of autophagy is essential for physiological turnover of old and damaged organelles, whereas the activation of autophagy beyond a certain threshold promote cell alterations and muscle atrophy [183-190]. For example, exercise promotes beneficial effects on muscle by improving overall mitochondrial quality through the selective removal of damaged or dysfunctional mitochondria [191, 192]. Indeed, long-life regular exercise in humans has been shown to preserve functional autophagy, guaranteeing better muscle mass and strength in senior sportsmen than elderly sedentary subjects [193]. Danon Disease (DD) and Glycogen Storage Disease type II (GSDII) represent two autophagic vacuolar myopathies, in which an autophagy block correlates with the severity of the disease [194]. Activity-dependent regulation of autophagy is also important in stabilizing the neuromuscular junction. It has been reported that muscle-specific loss of Atg7, a principal component of the autophagy machinery leads to neuromuscular junctions fragmentation, precocious synaptic dysfunction, and partial denervation [193]. Of note, basal autophagy is also essential to maintain the satellite cells quiescent state in mice [195].

Interestingly, it has been demonstrated that activated autophagy, by low-protein diet, is able to counteract myofiber apoptosis and to improve mitochondrial function in patients affected by COL6/collagen VI-related myopathies [196].

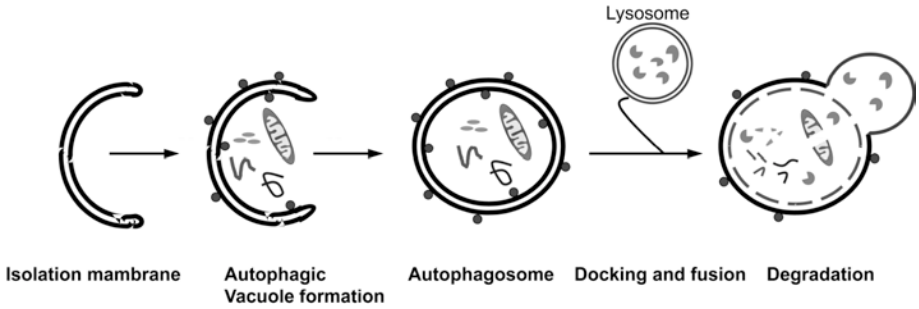

Fig. (5). Schematic representation of the autophay-mediated degradation. Autophagy involves the formation of a cytosolic double-membrane vesicle, an autophagosome, which sequesters and degrades components of the cytoplasm. The formation of autophagosome occurs in different steps: a portion of cytoplasm is first enclosed in a double structure, forming an autophagic vacuole surrounding proteins and cytoplasmic organels. The final step is characterized by the fusion of the autophagic vacuole with the lysosome, in which the lysosomal hydrolases degrade proteins, organelles and cellular components. The circle in the figure represents LC3.

On the other hand, excess of autophagy can be associated with cancer-associated cachexia [197] and in different pathological conditions including absence of nutrients [198, 199], sepsis [200], cirrhosis [201], skeletal muscle myopenia induced by bile duct ligation [202], disuse [203], denervation [204, 205], muscle atrophy induced by local expression of mutant SOD1 [16, 206].

Recently, a set of guidelines for the examination of macroautophagy and related processes has been reported [207].

10. ROLE OF EXERCISE AND PROTEIN INTAKE AS EFFECTIVE COUNTERMEASURES TO THE DELETERIOUS CONDITION OF MUSCLE ATROPHY AND WASTING

Several interventions have been suggested to counteract muscle atrophy and wasting.

Physical exercise can counteract muscle atrophy through multiple metabolic and transcriptional adaptations. Exercise acts on signaling pathways involved in muscle mass maintenance, such as the IGF-1/Akt/mTOR axis and on catabolic pathways, such as those mediated by FoxOs transcriptional factors [208-210].

Physical exercise can also attenuate muscle atrophy through the modulation of muscle catabolic pathways. Interestingly, it has been demonstrated that 12 weeks of resistance training in young women are sufficient to modulate the basal expression level of two molecular marker of the ubiquitin proteasome dependent proteolysis, MuRF-1 and FOXO3A [211]. Moreover, it has been demonstrated that exercise training enhances the cellular autophagic flux, inducing protein turnover and cellular organelles quality control processes [212, 213]. All these data demonstrate that physical exercise can counteract muscle atrophy favorably modulating the catabolic pathway.

Of note, nutrition also plays an important role in muscle homeostasis. It has been demonstrated that food intake can regulate muscle anabolic stimuli. In particular, the Essential Amino Acids (EAAs) have been described as the most important nutritional input for synthesis of new proteins; indeed the EAA Leucine is considered as the major nutritional regulator of protein anabolism for its ability to activate mTOR pathway and to inhibit the proteasome ubiquitin one [214].

Moreover, an antioxidant cocktail of rutin, vitamin E, vitamin A, zinc, and selenium is able to restore the defective leucine stimulation of protein synthesis and function of aged rats [215]. Taurine (2-aminoethane-sulfonic acid), a sulfur-containing semi-essential aminoacid has been demonstrated to regulate ubiquitin-proteasome system (UPS) and autophagy [216], promoting also the maintenance of cellular integrity and helping to overcome the resistance to anabolic stimuli and loss of muscle mass in elderly [217].

Recent literature has highlighted that the combination of exercise with increased protein intake can represent a plausible strategy to counteract muscle wasting. In this regard, it has been demonstrated that protein supplementation combined with both aerobic and resistance training is able to increase muscle mass, strength, and physical performance in elderly participants [218].

CONCLUSION

Muscle homeostasis is the result of a delicate balance between anabolic and catabolic processes, which if altered can lead to debilitating conditions. Thus, modulation of the anabolic and catabolic pathways represents an attractive therapeutic intervention to counteract muscle wasting and to guarantee the quality control of cellular components.

CONSENT FOR PUBLICATION

Not applicable.

CONFLICT OF INTEREST

The authors declare no conflict of interest, financial or otherwise.

ACKNOWLEDGEMENTS

We apologize to the colleagues whose studies were not cited. The work in the authors' laboratory has been supported by Fondazione Roma, Telethon, ASI, and Progetti di Ateneo. (Sapienza to AM and linea D.3.2 2015 to BMS).

REFERENCES

[1] Berridge, M.J.; Lipp, P.; Bootman, M.D. The versatility and universality of calcium signalling. *Nat. Rev. Mol. Cell Biol.*, **2000**, *1*(1), 11-21.

[2] Gillis, J.M. Membrane abnormalities and Ca homeostasis in mus-

cles of the mdx mouse, an animal model of the Duchenne muscular dystrophy: A review. *Acta Physiol. Scand.*, **1996**, *156*(3), 397-406.

[3] Orrenius, S.; Zhivotovsky, B.; Nicotera, P. Regulation of cell death: The calcium-apoptosis link. *Nat. Rev. Mol. Cell Biol.*, **2003**, *4*(7), 552-565.

[4] Schiaffino, S.; Dyar, K.A.; Ciciliot, S.; Blaauw, B.; Sandri, M. Mechanisms regulating skeletal muscle growth and atrophy. *FEBS J.*, **2013**, *280*(17), 4294-4314.

[5] Schiaffino, S.; Mammucari, C. Regulation of skeletal muscle growth by the IGF1-Akt/PKB pathway: Insights from genetic models. *Skelet. Muscle*, **2011**, *1*(1), 4. Available from: https://skeletalmusclejournal.biomedcentral.com/articles/10.1186/2044-5040-1-4

[6] Sandri, M.; Barberi, L.; Bijlsma, A.Y.; Blaauw, B.; Dyar, K.A.; Milan, G.; Mammucari, C.; Meskers, C.G.; Pallafacchina, G.; Paoli, A.; Pion, D.; Roceri, M.; Romanello, V.; Serrano, A.L.; Toniolo, L.; Larsson, L.; Maier, A.B.; Munoz-Canoves, P.; Musaro, A.; Pende, M.; Reggiani, C.; Rizzuto, R.; Schiaffino, S. Signalling pathways regulating muscle mass in ageing skeletal muscle: The role of the IGF1-Akt-mTOR-FoxO pathway. *Biogerontology*, **2013**, *14*(3), 303-323.

[7] Scicchitano, B.M.; Rizzuto, E.; Musaro, A. Counteracting muscle wasting in aging and neuromuscular diseases: The critical role of IGF-1. *Aging (Albany. N.Y.)*, **2009**, *1*(5), 451-457.

[8] Shavlakadze, T.; Winn, N.; Rosenthal, N.; Grounds, M.D. Reconciling data from transgenic mice that overexpress IGF-I specifically in skeletal muscle. *Growth Horm. IGF Res.*, **2005**, *15*(1), 4-18.

[9] Chew, S.L.; Lavender, P.; Clark, A.J.; Ross, R.J. An alternatively spliced human insulin-like growth factor-I transcript with hepatic tissue expression that diverts away from the mitogenic IBE1 peptide. *Endocrinology*, **1995**, *136*(5), 1939-1944.

[10] Matheny, R.W., Jr.; Nindl, B.C.; Adamo, M.L. Minireview: Mechano-growth factor: A putative product of IGF-I gene expression involved in tissue repair and regeneration. *Endocrinology*, **2010**, *151*(3), 865-875.

[11] Musaro, A.; McCullagh, K.; Paul, A.; Houghton, L.; Dobrowolny, G.; Molinaro, M.; Barton, E.R.; Sweeney, H.L.; Rosenthal, N. Localized Igf-1 transgene expression sustains hypertrophy and regeneration in senescent skeletal muscle. *Nat. Genet.*, **2001**, *27*(2), 195-200.

[12] Musaro, A.; Giacinti, C.; Borsellino, G.; Dobrowolny, G.; Pelosi, L.; Cairns, L.; Ottolenghi, S.; Cossu, G.; Bernardi, G.; Battistini, L.; Molinaro, M.; Rosenthal, N. Stem cell-mediated muscle regeneration is enhanced by local isoform of insulin-like growth factor 1. *Proc. Natl. Acad. Sci. U.S.A.*, **2004**, *101*(5), 1206-1210.

[13] Barton, E.R.; Morris, L.; Musaro, A.; Rosenthal, N.; Sweeney, H.L. Muscle-specific expression of insulin-like growth factor I counters muscle decline in mdx mice. *J. Cell Biol.*, **2002**, *157*(1), 137-148.

[14] Pelosi, L.; Giacinti, C.; Nardis, C.; Borsellino, G.; Rizzuto, E.; Nicoletti, C.; Wannenes, F.; Battistini, L.; Rosenthal, N.; Molinaro, M.; Musaro, A. Local expression of IGF-1 accelerates muscle regeneration by rapidly modulating inflammatory cytokines and chemokines. *FASEB J.*, **2007**, *21*(7), 1393-1402.

[15] Dobrowolny, G.; Giacinti, C.; Pelosi, L.; Nicoletti, C.; Winn, N.; Barberi, L.; Molinaro, M.; Rosenthal, N.; Musaro, A. Muscle expression of a local Igf-1 isoform protects motor neurons in an ALS mouse model. *J. Cell Biol.*, **2005**, *168*(2), 193-199.

[16] Dobrowolny, G.; Aucello, M.; Rizzuto, E.; Beccafico, S.; Mammucari, C.; Boncompagni, S.; Belia, S.; Wannenes, F.; Nicoletti, C.; Del Prete, Z.; Rosenthal, N.; Molinaro, M.; Protasi, F.; Fano, G.; Sandri, M.; Musaro, A. Skeletal muscle is a primary target of SOD1G93A-mediated toxicity. *Cell Metab.*, **2008**, *8*(5), 425-436.

[17] Palazzolo, I.; Stack, C.; Kong, L.; Musaro, A.; Adachi, H.; Katsuno, M.; Sobue, G.; Taylor, J.P.; Sumner, C.J.; Fischbeck, K.H.; Pennuto, M. Overexpression of IGF-1 in muscle attenuates disease in a mouse model of spinal and bulbar muscular atrophy. *Neuron*, **2009**, *63*(3), 316-328.

[18] Bosch-Marce, M.; Wee, C.D.; Martinez, T.L.; Lipkes, C.E.; Choe, D.W.; Kong, L.; Van Meerbeke, J.P.; Musaro, A.; Sumner, C.J. Increased IGF-1 in muscle modulates the phenotype of severe SMA mice. *Hum. Mol. Genet.*, **2011**, *20*(9), 1844-1853.

[19] Coolican, S.A.; Samuel, D.S.; Ewton, D.Z.; McWade, F.J.; Florini, J.R. The mitogenic and myogenic actions of insulin-like growth factors utilize distinct signaling pathways. *J. Biol. Chem.*, **1997**, *272*(10), 6653-6662.

[20] Engert, J.C.; Berglund, E.B.; Rosenthal, N. Proliferation precedes differentiation in IGF-I-stimulated myogenesis. *J. Cell Biol.*, **1996**, *135*(2), 431-440.

[21] Florini, J.R.; Ewton, D.Z.; Falen, S.L.; Van Wyk, J.J. Biphasic concentration dependency of stimulation of myoblast differentiation by somatomedins. *Am. J. Physiol.*, **1986**, *250*(5 Pt 1), C771-C778.

[22] Musaro, A.; McCullagh, K.J.; Naya, F.J.; Olson, E.N.; Rosenthal, N. IGF-1 induces skeletal myocyte hypertrophy through calcineurin in association with GATA-2 and NF-ATc1. *Nature*, **1999**, *400*(6744), 581-585. Available from: https://www.nature.com/articles/23060

[23] Musaro, A. Maturation of the myogenic program is induced by postmitotic expression of insulin-like growth factor I. **1999**, *19*(4), 3115-3124.

[24] Rosenthal, S.M.; Cheng, Z.Q. Opposing early and late effects of insulin-like growth factor I on differentiation and the cell cycle regulatory retinoblastoma protein in skeletal myoblasts. *Proc. Natl. Acad. Sci. U.S.A.*, **1995**, *92*(22), 10307-10311.

[25] Cross, D.A.; Alessi, D.R.; Cohen, P.; Andjelkovich, M.; Hemmings, B.A. Inhibition of glycogen synthase kinase-3 by insulin mediated by protein kinase B. *Nature*, **1995**, *378*(6559), 785-789. Available from: https://www.nature.com/articles/378785a0

[26] Hara, K.; Maruki, Y.; Long, X.; Yoshino, K.; Oshiro, N.; Hidayat, S.; Tokunaga, C.; Avruch, J.; Yonezawa, K. Raptor, a binding partner of target of rapamycin (TOR), mediates TOR action. *Cell*, **2002**, *110*(2), 177-189.

[27] Kim, D.H.; Sarbassov, D.D.; Ali, S.M.; Latek, R.R.; Guntur, K.V.; Erdjument-Bromage, H.; Tempst, P.; Sabatini, D.M. GbetaL, a positive regulator of the rapamycin-sensitive pathway required for the nutrient-sensitive interaction between raptor and mTOR. *Mol. Cell*, **2003**, *11*(4), 895-904.

[28] Kim, D.H.; Sarbassov, D.D.; Ali, S.M.; King, J.E.; Latek, R.R.; Erdjument-Bromage, H.; Tempst, P.; Sabatini, D.M. mTOR interacts with raptor to form a nutrient-sensitive complex that signals to the cell growth machinery. *Cell*, **2002**, *110*(2), 163-175.

[29] Jacinto, E.; Loewith, R.; Schmidt, A.; Lin, S.; Ruegg, M.A.; Hall, A.; Hall, M.N. Mammalian TOR complex 2 controls the actin cytoskeleton and is rapamycin insensitive. *Nat. Cell Biol.*, **2004**, *6*(11), 1122-1128.

[30] Sarbassov, D.D.; Ali, S.M.; Kim, D.H.; Guertin, D.A.; Latek, R.R.; Erdjument-Bromage, H.; Tempst, P.; Sabatini, D.M. Rictor, a novel binding partner of mTOR, defines a rapamycin-insensitive and raptor-independent pathway that regulates the cytoskeleton. *Curr. Biol.*, **2004**, *14*(14), 1296-1302.

[31] Peterson, T.R.; Laplante, M.; Thoreen, C.C.; Sancak, Y.; Kang, S.A.; Kuehl, W.M.; Gray, N.S.; Sabatini, D.M. DEPTOR is an mTOR inhibitor frequently overexpressed in multiple myeloma cells and required for their survival. *Cell*, **2009**, *137*(5), 873-886.

[32] Frias, M.A.; Thoreen, C.C.; Jaffe, J.D.; Schroder, W.; Sculley, T.; Carr, S.A.; Sabatini, D.M. mSin1 is necessary for Akt/PKB phosphorylation, and its isoforms define three distinct mTORC2s. *Curr. Biol.*, **2006**, *16*(18), 1865-1870.

[33] Jacinto, E.; Facchinetti, V.; Liu, D.; Soto, N.; Wei, S.; Jung, S.Y.; Huang, Q.; Qin, J.; Su, B. SIN1/MIP1 maintains rictor-mTOR complex integrity and regulates Akt phosphorylation and substrate specificity. *Cell*, **2006**, *127*(1), 125-137.

[34] Yang, Q.; Inoki, K.; Ikenoue, T.; Guan, K.L. Identification of Sin1 as an essential TORC2 component required for complex formation and kinase activity. *Genes Dev.*, **2006**, *20*(20), 2820-2832.

[35] Pearce, L.R.; Huang, X.; Boudeau, J.; Pawlowski, R.; Wullschleger, S.; Deak, M.; Ibrahim, A.F.; Gourlay, R.; Magnuson, M.A.; Alessi, D.R. Identification of Protor as a novel Rictor-binding component of mTOR complex-2. *Biochem. J.*, **2007**, *405*(3), 513-522.

[36] Thedieck, K.; Polak, P.; Kim, M.L.; Molle, K.D.; Cohen, A.; Jeno, P.; Arrieumerlou, C.; Hall, M.N. PRAS40 and PRR5-like protein are new mTOR interactors that regulate apoptosis. *PLoS One*, **2007**, *2*(11), e1217. Available from: http://journals.plos.org/

plosone/article?id=10.1371/journal.pone.0001217

[37] Woo, S.Y.; Kim, D.H.; Jun, C.B.; Kim, Y.M.; Haar, E.V.; Lee, S.I.; Hegg, J.W.; Bandhakavi, S.; Griffin, T.J.; Kim, D.H. PRR5, a novel component of mTOR complex 2, regulates platelet-derived growth factor receptor beta expression and signaling. *J. Biol. Chem.*, **2007**, *282*(35), 25604-25612.

[38] Saxton, R.A.; Sabatini, D.M. mTOR signaling in growth, metabolism, and disease. *Cell*, **2017**, *168*(6), 960-976.

[39] Dorrello, N.V.; Peschiaroli, A.; Guardavaccaro, D.; Colburn, N.H.; Sherman, N.E.; Pagano, M. *Science*, **2006**, *314*(5798), 467-471. Available from: http://science.sciencemag.org/content/314/5798/467

[40] Holz, M.K.; Blenis, J. Identification of S6 kinase 1 as a novel mammalian target of rapamycin (mTOR)-phosphorylating kinase. *J. Biol. Chem*., **2005**, *280*(28), 26089-26093.

[41] Ma, X.M.; Yoon, S.O.; Richardson, C.J.; Julich, K.; Blenis, J. SKAR links pre-mRNA splicing to mTOR/S6K1-mediated enhanced translation efficiency of spliced mRNAs. *Cell*, **2008**, *133*(2), 303-313.

[42] Hoppeler, H. Molecular networks in skeletal muscle plasticity. *J. Exp. Biol*., **2016**, *219*(Pt 2), 205-213.

[43] Sarbassov, D.D.; Ali, S.M.; Sabatini, D.M. Growing roles for the mTOR pathway. *Curr. Opin. Cell Biol.*, **2005**, *17*(6), 596-603.

[44] Gan, X.; Wang, J.; Wang, C.; Sommer, E.; Kozasa, T.; Srinivasula, S.; Alessi, D.; Offermanns, S.; Simon, M.I.; Wu, D. PRR5L degradation promotes mTORC2-mediated PKC-delta phosphorylation and cell migration downstream of Galpha12. *Nat. Cell Biol.*, **2012**, *14*(7), 686-696.

[45] Li, X.; Gao, T. mTORC2 phosphorylates protein kinase Czeta to regulate its stability and activity. *EMBO Rep.*, **2014**, *15*(2), 191-198.

[46] Thomanetz, V.; Angliker, N.; Cloetta, D.; Lustenberger, R.M.; Schweighauser, M.; Oliveri, F.; Suzuki, N.; Ruegg, M.A. Ablation of the mTORC2 component rictor in brain or Purkinje cells affects size and neuron morphology. *J. Cell Biol.*, **2013**, *201*(2), 293-308.

[47] Dufresne, S.D.; Bjorbaek, C.; El Haschimi, K.; Zhao, Y.; Aschenbach, W.G.; Moller, D.E.; Goodyear, L.J. Altered extracellular signal-regulated kinase signaling and glycogen metabolism in skeletal muscle from p90 ribosomal S6 kinase 2 knockout mice. *Mol. Cell Biol.*, **2001**, *21*(1), 81-87.

[48] Pelosi, M.; Marampon, F.; Zani, B.M.; Prudente, S.; Perlas, E.; Caputo, V.; Cianetti, L.; Berno, V.; Narumiya, S.; Kang, S.W.; Musaro, A.; Rosenthal, N. ROCK2 and its alternatively spliced isoform ROCK2m positively control the maturation of the myogenic program. *Mol. Cell Biol.*, **2007**, *27*(17), 6163-6176.

[49] Roux, P.P.; Richards, S.A.; Blenis, J. Phosphorylation of p90 ribosomal S6 kinase (RSK) regulates extracellular signal-regulated kinase docking and RSK activity. *Mol. Cell Biol.*, **2003**, *23*(14), 4796-4804.

[50] Wang, X.; Li, W.; Williams, M.; Terada, N.; Alessi, D.R.; Proud, C.G. Regulation of elongation factor 2 kinase by p90(RSK1) and p70 S6 kinase. *EMBO J.*, **2001**, *20*(16), 4370-4379.

[51] Bonaldo, P.; Sandri, M. Cellular and molecular mechanisms of muscle atrophy. *Dis. Model. Mech.*, **2013**, *6*(1), 25-39.

[52] Cohen, S.; Nathan, J.A.; Goldberg, A.L. Muscle wasting in disease: Molecular mechanisms and promising therapies. *Nat. Rev. Drug Discov.*, **2015**, *14*(1), 58-74.

[53] Fry, C.S.; Drummond, M.J.; Glynn, E.L.; Dickinson, J.M.; Gundermann, D.M.; Timmerman, K.L.; Walker, D.K.; Dhanani, S.; Volpi, E.; Rasmussen, B.B. Aging impairs contraction-induced human skeletal muscle mTORC1 signaling and protein synthesis. *Skelet. Muscle*, **2011**, *1*(1), 11. Available from: https://skeletalmusclejournal.biomedcentral.com/articles/10.1186/2044-5040-1-11

[54] Vinciguerra, M.; Musaro, A.; Rosenthal, N. Regulation of muscle atrophy in aging and disease. *Adv. Exp. Med. Biol.*, **2010**, *694*, 211-233.

[55] Mitch, W.E.; Bailey, J.L.; Wang, X.; Jurkovitz, C.; Newby, D.; Price, S.R. Evaluation of signals activating ubiquitin-proteasome proteolysis in a model of muscle wasting. *Am. J. Physiol*., **1999**, *276*(5 Pt1), C1132-C1138.

[56] Mitch, W.E.; Price, S.R. Mechanisms activating proteolysis to cause muscle atrophy in catabolic conditions. *J. Ren. Nutr.*, **2003**, *13*(2), 149-152.

[57] Mitch, W.E.; Goldberg, A.L. Mechanisms of muscle wasting. The role of the ubiquitin-proteasome pathway. *N. Engl. J. Med.*, **1996**, *335*(25), 1897-1905.

[58] Zamir, O.; Hasselgren, P.O.; Kunkel, S.L.; Frederick, J.; Higashiguchi, T.; Fischer, J.E. Evidence that tumor necrosis factor participates in the regulation of muscle proteolysis during sepsis. *Arch. Surg.*, **1992**, *127*(2), 170-174.

[59] Jagoe, R.T.; Lecker, S.H.; Gomes, M.; Goldberg, A.L. Patterns of gene expression in atrophying skeletal muscles: response to food deprivation. *FASEB J.*, **2002**, *16*(13), 1697-1712.

[60] Jagoe, R.T.; Goldberg, A.L. What do we really know about the ubiquitin-proteasome pathway in muscle atrophy? *Curr. Opin. Clin. Nutr. Metab. Care*, **2001**, *4*(3), 183-190.

[61] Lecker, S.H.; Jagoe, R.T.; Gilbert, A.; Gomes, M.; Baracos, V.; Bailey, J.; Price, S.R.; Mitch, W.E.; Goldberg, A.L. Multiple types of skeletal muscle atrophy involve a common program of changes in gene expression. *FASEB J.*, **2004**, *18*(1), 39-51.

[62] Bodine, S.C.; Stitt, T.N.; Gonzalez, M.; Kline, W.O.; Stover, G.L.; Bauerlein, R.; Zlotchenko, E.; Scrimgeour, A.; Lawrence, J.C.; Glass, D.J.; Yancopoulos, G.D. Akt/mTOR pathway is a crucial regulator of skeletal muscle hypertrophy and can prevent muscle atrophy *in vivo*. *Nat. Cell Biol.*, **2001**, *3*(11), 1014-1019.

[63] Gomes, M.D.; Lecker, S.H.; Jagoe, R.T.; Navon, A.; Goldberg, A.L. Atrogin-1, a muscle-specific F-box protein highly expressed during muscle atrophy. *Proc. Natl. Acad. Sci. U.S.A*., **2001**, *98*(25), 14440-14445.

[64] Sandri, M.; Sandri, C.; Gilbert, A.; Skurk, C.; Calabria, E.; Picard, A.; Walsh, K.; Schiaffino, S.; Lecker, S.H.; Goldberg, A.L. Foxo transcription factors induce the atrophy-related ubiquitin ligase atrogin-1 and cause skeletal muscle atrophy. *Cell*, **2004**, *117*(3), 399-412.

[65] Kamei, Y.; Miura, S.; Suzuki, M.; Kai, Y.; Mizukami, J.; Taniguchi, T.; Mochida, K.; Hata, T.; Matsuda, J.; Aburatani, H.; Nishino, I.; Ezaki, O. Skeletal muscle FOXO1 (FKHR) transgenic mice have less skeletal muscle mass, down-regulated Type I (slow twitch/red muscle) fiber genes, and impaired glycemic control. *J. Biol. Chem.*, **2004**, *279*(39), 41114-41123.

[66] Clarke, B.A.; Drujan, D.; Willis, M.S.; Murphy, L.O.; Corpina, R.A.; Burova, E.; Rakhilin, S.V.; Stitt, T.N.; Patterson, C.; Latres, E.; Glass, D.J. The E3 Ligase MuRF1 degrades myosin heavy chain protein in dexamethasone-treated skeletal muscle. *Cell Metab*., **2007**, *6*(5), 376-385.

[67] Cohen, S.; Brault, J.J.; Gygi, S.P.; Glass, D.J.; Valenzuela, D.M.; Gartner, C.; Latres, E.; Goldberg, A.L. During muscle atrophy, thick, but not thin, filament components are degraded by MuRF1-dependent ubiquitylation. *J. Cell Biol.*, **2009**, *185*(6), 1083-1095.

[68] Lagirand-Cantaloube, J.; Offner, N.; Csibi, A.; Leibovitch, M.P.; Batonnet-Pichon, S.; Tintignac, L.A.; Segura, C.T.; Leibovitch, S.A. The initiation factor eIF3-f is a major target for atrogin1/MAFbx function in skeletal muscle atrophy. *EMBO J.*, **2008**, *27*(8), 1266-1276.

[69] Tintignac, L.A.; Lagirand, J.; Batonnet, S.; Sirri, V.; Leibovitch, M.P.; Leibovitch, S.A. Degradation of MyoD mediated by the SCF (MAFbx) ubiquitin ligase. *J. Biol. Chem.*, **2005**, *280*(4), 2847-2856.

[70] Murgia, M.; Serrano, A.L.; Calabria, E.; Pallafacchina, G.; Lomo, T.; Schiaffino, S. Ras is involved in nerve-activity-dependent regulation of muscle genes. *Nat. Cell Biol.*, **2000**, *2*(3), 142-147.

[71] Milan, G.; Romanello, V.; Pescatore, F.; Armani, A.; Paik, J.H.; Frasson, L.; Seydel, A.; Zhao, J.; Abraham, R.; Goldberg, A.L.; Blaauw, B.; DePinho, R.A.; Sandri, M. Regulation of autophagy and the ubiquitin-proteasome system by the FoxO transcriptional network during muscle atrophy. *Nat. Commun.*, **2015**, *6*, 6670. Available from: https://pathology.weill.cornell.edu/regulation-autophagy-and-ubiquitin-proteasome-system-foxo-transcriptional-network-during-muscle

[72] Sartori, R.; Schirwis, E.; Blaauw, B.; Bortolanza, S.; Zhao, J.; Enzo, E.; Stantzou, A.; Mouisel, E.; Toniolo, L.; Ferry, A.; Stricker, S.; Goldberg, A.L.; Dupont, S.; Piccolo, S.; Amthor, H.; Sandri, M. BMP signaling controls muscle mass. *Nat. Genet.*, **2013**, *45*(11), 1309-1318.

[73] Asp, M.L.; Tian, M.; Wendel, A.A.; Belury, M.A. Evidence for the contribution of insulin resistance to the development of cachexia in tumor-bearing mice. *Int. J. Cancer*, **2010**, *126*(3), 756-763.

[74] Penna, F.; Bonetto, A.; Muscaritoli, M.; Costamagna, D.; Minero, V.G.; Bonelli, G.; Rossi, F.F.; Baccino, F.M.; Costelli, P. Muscle atrophy in experimental cancer cachexia: Is the IGF-1 signaling pathway involved? *Int. J. Cancer*, **2010**, *127*(7), 1706-1717.

[75] Brunet, A.; Bonni, A.; Zigmond, M.J.; Lin, M.Z.; Juo, P.; Hu, L.S.; Anderson, M.J.; Arden, K.C.; Blenis, J.; Greenberg, M.E. Akt promotes cell survival by phosphorylating and inhibiting a Forkhead transcription factor. *Cell*, **1999**, *96*(6), 857-868.

[76] Alnaqeeb, M.A.; Goldspink, G. Changes in fibre type, number and diameter in developing and ageing skeletal muscle. *J. Anat.*, **1987**, *153*, 31-45.

[77] Thompson, L.M.; Aiken, C.T.; Kaltenbach, L.S.; Agrawal, N.; Illes, K.; Khoshnan, A.; Martinez-Vincente, M.; Arrasate, M.; O'Rourke, J.G.; Khashwji, H.; Lukacsovich, T.; Zhu, Y.Z.; Lau, A.L.; Massey, A.; Hayden, M.R.; Zeitlin, S.O.; Finkbeiner, S.; Green, K.N.; LaFerla, F.M.; Bates, G.; Huang, L.; Patterson, P.H.; Lo, D.C.; Cuervo, A.M.; Marsh, J.L.; Steffan, J.S. IKK phosphorylates Huntingtin and targets it for degradation by the proteasome and lysosome. *J. Cell Biol.*, **2009**, *187*(7), 1083-1099.

[78] Altun, M.; Besche, H.C.; Overkleeft, H.S.; Piccirillo, R.; Edelmann, M.J.; Kessler, B.M.; Goldberg, A.L.; Ulfhake, B. Muscle wasting in aged, sarcopenic rats is associated with enhanced activity of the ubiquitin proteasome pathway. *J. Biol. Chem.*, **2010**, *285*(51), 39597-39608.

[79] Clavel, S.; Coldefy, A.S.; Kurkdjian, E.; Salles, J.; Margaritis, I.; Derijard, B. Atrophy-related ubiquitin ligases, atrogin-1 and MuRF1 are up-regulated in aged rat Tibialis Anterior muscle. *Mech. Ageing Dev.*, **2006**, *127*(10), 794-801.

[80] Grounds, M.D. Reasons for the degeneration of ageing skeletal muscle: A central role for IGF-1 signalling. *Biogerontology*, **2002**, *3*(1-2), 19-24.

[81] Rosen, C.J.; Pollak, M. Circulating IGF-I: New perspectives for a new century. *Trends Endocrinol. Metab.,* **1999**, *10*(4), 136-141.

[82] Rudman, D.; Kutner, M.H.; Rogers, C.M.; Lubin, M.F.; Fleming G.A.; Bain, R.P. Impaired growth hormone secretion in the adult population: relation to age and adiposity. *J. Clin. Invest*., **1981**, *67*(5), 1361-1369.

[83] Edstrom, E.; Altun, M.; Hagglund, M.; Ulfhake, B. Atrogin-1/MAFbx and MuRF1 are downregulated in aging-related loss of skeletal muscle. *J. Gerontol. A Biol. Sci. Med. Sci.*, **2006**, *61*(7), 663-674.

[84] Agusti, A.; Morla, M.; Sauleda, J.; Saus, C.; Busquets, X. NF-kappaB activation and iNOS upregulation in skeletal muscle of patients with COPD and low body weight. *Thorax*, **2004**, *59*(6), 483-487.

[85] Bailey, J.L.; Zheng, B.; Hu, Z.; Price, S.R.; Mitch, W.E. Chronic kidney disease causes defects in signaling through the insulin receptor substrate/phosphatidylinositol 3-kinase/Akt pathway: Implications for muscle atrophy. *J. Am. Soc. Nephrol.*, **2006**, *17*(5), 1388-1394.

[86] Wang, X.; Robbins, J. Heart failure and protein quality control. *Circ. Res.*, **2006**, *99*(12), 1315-1328.

[87] Yoshida, T.; Delafontaine, P. Mechanisms of Cachexia in chronic disease states. *Am. J. Med. Sci.*, **2015**, *350*(4), 250-256.

[88] Joyce, P.I.; Satija, R.; Chen, M.; Kuwabara, P.E. The atypical calpains: Evolutionary analyses and roles in Caenorhabditis elegans cellular degeneration. *PLoS Genet.*, **2012**, *8*(3), e1002602. Available from: http://journals.plos.org/plosgenetics/article?id=10.1371/journal.pgen.1002602

[89] Sorimachi, H.; Imajoh-Ohmi, S.; Emori, Y.; Kawasaki, H.; Ohno, S.; Minami, Y.; Suzuki, K. Molecular cloning of a novel mammalian calcium-dependent protease distinct from both m- and mu-types. Specific expression of the mRNA in skeletal muscle. *J. Biol. Chem.*, **1989**, *264*(33), 20106-20111.

[90] Ermolova, N.; Kramerova, I.; Spencer, M.J. Autolytic activation of calpain 3 proteinase is facilitated by calmodulin protein. *J. Biol. Chem.*, **2015**, *290*(2), 996-1004.

[91] Lebart, M.C.; Benyamin, Y. Calpain involvement in the remodeling of cytoskeletal anchorage complexes. *FEBS J.*, **2006**, *273*(15), 3415-3426.

[92] Franco, S.J.; Huttenlocher, A. Regulating cell migration: Calpains make the cut. *J. Cell Sci.*, **2005**, *118*(Pt17), 3829-3838.

[93] Goll, D.E.; Neti, G.; Mares, S.W.; Thompson, V.F. Myofibrillar protein turnover: the proteasome and the calpains. *J. Anim. Sci.*, **2008**, *86*(14 Suppl), E19-E35.

[94] Evans, J.S.; Turner, M.D. Emerging functions of the calpain superfamily of cysteine proteases in neuroendocrine secretory pathways. *J. Neurochem.*, **2007**, *103*(3), 849-859.

[95] Janossy, J.; Ubezio, P.; Apati, A.; Magocsi, M.; Tompa, P.; Friedrich, P. Calpain as a multi-site regulator of cell cycle. *Biochem. Pharmacol.,* **2004**, *67*(8), 1513-1521.

[96] Storr, S.J.; Carragher, N.O.; Frame, M.C.; Parr, T.; Martin, S.G. The calpain system and cancer. *Nat. Rev. Cancer*, **2011**, *11*(5), 364-374.

[97] Tomimatsu, Y.; Idemoto, S.; Moriguchi, S.; Watanabe, S.; Nakanishi, H. Proteases involved in long-term potentiation. *Life Sci.*, **2002**, *72*(4-5), 355-361.

[98] Belcastro, A.N.; Shewchuk, L.D.; Raj, D.A. Exercise-induced muscle injury: A calpain hypothesis. *Mol. Cell Biochem.*, **1998**, *179*(1-2), 135-145.

[99] Armstrong, R.B. Mechanisms of exercise-induced muscle fibre injury. *Sports Med.,* **1991**, *12*(3), 184-207.

[100] Friden, J.; Sfakianos, P.N.; Hargens, A.R. Blood indices of muscle injury associated with eccentric muscle contractions. *J. Orthop. Res.*, **1989**, *7*(1), 142-145.

[101] Cannon, J.G.; Meydani, S.N.; Fielding, R.A.; Fiatarone, M.A.; Meydani, M.; Farhangmehr, M.; Orencole, S.F.; Blumberg, J.B.; Evans, W.J. Acute phase response in exercise. II. Associations between vitamin E, cytokines, and muscle proteolysis. *Am. J. Physiol*., **1991**, *260*(6 Pt2), R1235-R1240.

[102] Fareed, M.U.; Evenson, A.R.; Wei, W.; Menconi, M.; Poylin, V.; Petkova, V.; Pignol, B.; Hasselgren, P.O. Treatment of rats with calpain inhibitors prevents sepsis-induced muscle proteolysis independent of atrogin-1/MAFbx and MuRF1 expression. *Am. J. Physiol. Regul. Integr. Comp. Physiol*., **2006**, *290*(6), R1589-R1597.

[103] Matsumoto, A.; Fujita, N.; Arakawa, T.; Fujino, H.; Miki, A. Influence of electrical stimulation on calpain and ubiquitin-proteasome systems in the denervated and unloaded rat tibialis anterior muscles. *Acta Histochem.,* **2014**, *116*(5), 936-942.

[104] Nelson, W.B.; Smuder, A.J.; Hudson, M.B.; Talbert, E.E.; Powers, S.K. Cross-talk between the calpain and caspase-3 proteolytic systems in the diaphragm during prolonged mechanical ventilation. *Crit. Care Med.*, **2012**, *40*(6), 1857-1863.

[105] Kumamoto, T.; Kleese, W.C.; Cong, J.Y.; Goll, D.E.; Pierce, P.R.; Allen, R.E. Localization of the Ca(2+)-dependent proteinases and their inhibitor in normal, fasted, and denervated rat skeletal muscle. *Anat. Rec.*, **1992**, *232*(1), 60-77.

[106] Huang, J.; Forsberg, N.E. Role of calpain in skeletal-muscle protein degradation. *Proc. Natl. Acad. Sci. U.S.A*., **1998**, *95*(21), 12100-12105.

[107] Martin, S.J.; O'Brien, G.A.; Nishioka, W.K.; McGahon, A.J.; Mahboubi, A.; Saido, T.C.; Green, D.R. Proteolysis of fodrin (non-erythroid spectrin) during apoptosis. *J. Biol. Chem.,* **1995**, *270*(12), 6425-6428.

[108] Planey, S.L.; Litwack, G. Glucocorticoid-induced apoptosis in lymphocytes. *Biochem. Biophys. Res. Commun.*, **2000**, *279*(2), 307-312.

[109] Goll, D.E.; Thompson, V.F.; Li, H.; Wei, W.; Cong, J. The calpain system. *Physiol. Rev.*, **2003**, *83*(3), 731-801.

[110] Saido, T.C.; Sorimachi, H.; Suzuki, K. Calpain: New perspectives in molecular diversity and physiological-pathological involvement. *FASEB J.*, **1994**, *8*(11), 814-822.

[111] Di Lisa, F.; De Tullio, R.; Salamino, F.; Barbato, R.; Melloni, E.; Siliprandi, N.; Schiaffino, S.; Pontremoli, S. Specific degradation of troponin T and I by mu-calpain and its modulation by substrate phosphorylation. *Biochem. J.*, **1995**, *308(Pt 1)*, 57-61.

[112] Kunimatsu, M.; Ma, X.J.; Ozaki, Y.; Narita, M.; Mizokami, M.; Sasaki, M. Neutrophil chemotactic N-acetyl peptides from the calpain small subunit are also chemotactic for immunocytes. *Biochem. Mol. Biol. Int.*, **1995**, *35*(2), 247-254.

[113] Spencer, M.J.; Croall, D.E.; Tidball, J.G. Calpains are activated in

necrotic fibers from mdx dystrophic mice. *J. Biol. Chem.*, **1995**, *270*(18), 10909-10914.

[114] Baghdiguian, S.; Martin, M.; Richard, I.; Pons, F.; Astier, C.; Bourg, N.; Hay, R.T.; Chemaly, R.; Halaby, G.; Loiselet, J.; Anderson, L.V.; Lopez, d.M.; Fardeau, M.; Mangeat, P.; Beckmann, J.S.; Lefranc, G. Calpain 3 deficiency is associated with myonuclear apoptosis and profound perturbation of the IkappaB alpha/NF-kappaB pathway in limb-girdle muscular dystrophy type 2A. *Nat. Med.*, **1999**, *5*(5), 503-511.

[115] Spencer, M.J.; Guyon, J.R.; Sorimachi, H.; Potts, A.; Richard, I.; Herasse, M.; Chamberlain, J.; Dalkilic, I.; Kunkel, L.M.; Beckmann, J.S. Stable expression of calpain 3 from a muscle transgene *in vivo*: Immature muscle in transgenic mice suggests a role for calpain 3 in muscle maturation. *Proc. Natl. Acad. Sci. U.S.A.*, **2002**, *99*(13), 8874-8879.

[116] Gokhin, D.S.; Tierney, M.T.; Sui, Z.; Sacco, A.; Fowler, V.M. Calpain-mediated proteolysis of tropomodulin isoforms leads to thin filament elongation in dystrophic skeletal muscle. *Mol. Biol. Cell*, **2014**, *25*(6), 852-865.

[117] Chaudhary, P.; Suryakumar, G.; Prasad, R.; Singh, S.N.; Ali, S.; Ilavazhagan, G. Chronic hypobaric hypoxia mediated skeletal muscle atrophy: Role of ubiquitin-proteasome pathway and calpains. *Mol. Cell Biochem.,* **2012**, *364*(1-2), 101-113.

[118] Samengo, G.; Avik, A.; Fedor, B.; Whittaker, D.; Myung, K.H.; Wehling-Henricks, M.; Tidball, J.G. Age-related loss of nitric oxide synthase in skeletal muscle causes reductions in calpain S-nitrosylation that increase myofibril degradation and sarcopenia. *Aging Cell*, **2012**, *11*(6), 1036-1045.

[119] Barja, G. Free radicals and aging. *Trends Neurosci.*, **2004**, *27*(10), 595-600.

[120] Musaro, A.; Fulle, S.; Fano, G. Oxidative stress and muscle homeostasis. *Curr. Opin. Clin. Nutr. Metab. Care*, **2010**, *13*(3), 236-242.

[121] Smuder, A.J.; Kavazis, A.N.; Hudson, M.B.; Nelson, W.B.; Powers, S.K. Oxidation enhances myofibrillar protein degradation *via* calpain and caspase-3. *Free Radic. Biol. Med.*, **2010**, *49*(7), 1152-1160.

[122] McClung, J.M.; Judge, A.R.; Talbert, E.E.; Powers, S.K. Calpain-1 is required for hydrogen peroxide-induced myotube atrophy. *Am. J. Physiol. Cell Physiol.*, **2009**, *296*(2), C363-C371.

[123] Tidball, J.G.; Spencer, M.J. Expression of a calpastatin transgene slows muscle wasting and obviates changes in myosin isoform expression during murine muscle disuse. *J. Physiol.*, **2002**, *545*(Pt 3), 819-828.

[124] Stalker, T.J.; Skvarka, C.B.; Scalia, R. A novel role for calpains in the endothelial dysfunction of hyperglycemia. *FASEB J.*, **2003**, *17*(11), 1511-1513.

[125] Chockalingam, P.S.; Cholera, R.; Oak, S.A.; Zheng, Y.; Jarrett, H.W.; Thomason, D.B. Dystrophin-glycoprotein complex and Ras and Rho GTPase signaling are altered in muscle atrophy. *Am. J. Physiol. Cell Physiol.*, **2002**, *283*(2), C500-C511.

[126] Huang, J.; Zhu, X. The molecular mechanisms of calpains action on skeletal muscle atrophy. *Physiol. Res.,* **2016**, *65*(4), 547-560.

[127] Oda, A.; Wakao, H.; Fujita, H. Calpain is a signal transducer and activator of transcription (STAT) 3 and STAT5 protease. *Blood*, **2002**, *99*(5), 1850-1852.

[128] Sneddon, A.A.; Delday, M.I.; Maltin, C.A. Amelioration of denervation-induced atrophy by clenbuterol is associated with increased PKC-alpha activity. *Am. J. Physiol. Endocrinol. Metab.*, **2000**, *279*(1), E188-E195.

[129] Wang, K.K.; Roufogalis, B.D.; Villalobo, A. Characterization of the fragmented forms of calcineurin produced by calpain I. *Biochem. Cell Biol.*, **1989**, *67*(10), 703-711.

[130] Wei, L.; Sandbulte, M.R.; Thomas, P.G.; Webby, R.J.; Homayouni, R.; Pfeffer, L.M. NFkappaB negatively regulates interferon-induced gene expression and anti-influenza activity. *J. Biol. Chem.,* **2006**, *281*(17), 11678-11684.

[131] Pin, F.; Minero, V.G.; Penna, F.; Muscaritoli, M.; De Tullio, R.; Baccino, F.M.; Costelli, P. Interference with Ca^{2+}-dependent proteolysis does not alter the course of muscle wasting in experimental cancer Cachexia. *Front. Physiol.*, **2017**, *8*, 213. Available from: https://www.frontiersin.org/articles/10.3389/fphys.2017.00213/full

[132] Riedl, S.J.; Shi, Y. Molecular mechanisms of caspase regulation during apoptosis. *Nat. Rev. Mol. Cell Biol.*, **2004**, *5*(11), 897-907.

[133] Thornberry, N.A.; Lazebnik, Y. Caspases: Enemies within. *Science*, **1998**, *281*(5381), 1312-1316.

[134] Enari, M.; Sakahira, H.; Yokoyama, H.; Okawa, K.; Iwamatsu, A.; Nagata, S. A caspase-activated DNase that degrades DNA during apoptosis, and its inhibitor ICAD. *Nature*, **1998**, *391*(6662), 43-50. Available from: https://www.nature.com/articles/34112

[135] Liu, X.; Zou, H.; Slaughter, C.; Wang, X. DFF, a heterodimeric protein that functions downstream of caspase-3 to trigger DNA fragmentation during apoptosis. *Cell*, **1997**, *89*(2), 175-184.

[136] Orth, K.; Chinnaiyan, A.M.; Garg, M.; Froelich, C.J.; Dixit, V.M. The CED-3/ICE-like protease Mch2 is activated during apoptosis and cleaves the death substrate lamin A. *J. Biol. Chem.*, **1996**, *271*(28), 16443-16446.

[137] Takahashi, A.; Alnemri, E.S.; Lazebnik, Y.A.; Fernandes-Alnemri, T.; Litwack, G.; Moir, R.D.; Goldman, R.D.; Poirier, G.G.; Kaufmann, S.H.; Earnshaw, W.C. Cleavage of lamin A by Mch2 alpha but not CPP32: Multiple interleukin 1 beta-converting enzyme-related proteases with distinct substrate recognition properties are active in apoptosis. *Proc. Natl. Acad. Sci. U.S.A.*, **1996**, *93*(16), 8395-8400.

[138] Chen, F.; Chang, R.; Trivedi, M.; Capetanaki, Y.; Cryns, V.L. Caspase proteolysis of desmin produces a dominant-negative inhibitor of intermediate filaments and promotes apoptosis. *J. Biol. Chem.,* **2003**, *278*(9), 6848-6853.

[139] Cryns, V.; Yuan, J. Proteases to die for. *Genes Dev.*, **1998**, *12*(11), 1551-1570.

[140] Rheaume, E.; Cohen, L.Y.; Uhlmann, F.; Lazure, C.; Alam, A.; Hurwitz, J.; Sekaly, R.P.; Denis, F. The large subunit of replication factor C is a substrate for caspase-3 *in vitro* and is cleaved by a caspase-3-like protease during Fas-mediated apoptosis. *EMBO J.*, **1997**, *16*(21), 6346-6354.

[141] Sandri, M.; El Meslemani, A.H.; Sandri, C.; Schjerling, P.; Vissing, K.; Andersen, J.L.; Rossini, K.; Carraro, U.; Angelini, C. Caspase 3 expression correlates with skeletal muscle apoptosis in Duchenne and facioscapulo human muscular dystrophy. A potential target for pharmacological treatment? *J. Neuropathol. Exp. Neurol.*, **2001**, *60*(3), 302-312.

[142] Brostrom, M.A.; Brostrom ,C.O. Calcium dynamics and endoplasmic reticular function in the regulation of protein synthesis: Implications for cell growth and adaptability. *Cell Calcium,* **2003**, *34*(4-5), 345-363.

[143] Hotokezaka, Y.; van Leyen, K.; Lo, E.H.; Beatrix, B.; Katayama, I.; Jin, G.; Nakamura, T. alphaNAC depletion as an initiator of ER stress-induced apoptosis in hypoxia. *Cell Death Differ.,* **2009**, *16*(11), 1505-1514.

[144] Sawada, N.; Yao, J.; Hiramatsu, N.; Hayakawa, K.; Araki, I.; Takeda, M.; Kitamura, M. Involvement of hypoxia-triggered endoplasmic reticulum stress in outlet obstruction-induced apoptosis in the urinary bladder. *Lab. Invest.,* **2008**, *88*(5), 553-563.

[145] Tang, C.; Koulajian, K.; Schuiki, I.; Zhang, L.; Desai, T.; Ivovic, A.; Wang, P.; Robson-Doucette, C.; Wheeler, M.B.; Minassian, B.; Volchuk, A.; Giacca, A. Glucose-induced beta cell dysfunction *in vivo* in rats: Link between oxidative stress and endoplasmic reticulum stress. *Diabetologia*, **2012**, *55*(5), 1366-1379.

[146] Li, C.; Wei, J.; Li, Y.; He, X.; Zhou, Q.; Yan, J.; Zhang, J.; Liu, Y.; Liu, Y.; Shu, H.B. Transmembrane protein 214 (TMEM214) mediates endoplasmic reticulum stress-induced caspase 4 enzyme activation and apoptosis. *J. Biol. Chem.,* **2013**, *288*(24), 17908-17917.

[147] Morishima, N.; Nakanishi, K.; Takenouchi, H.; Shibata, T.; Yasuhiko, Y. An endoplasmic reticulum stress-specific caspase cascade in apoptosis. Cytochrome c-independent activation of caspase-9 by caspase-12. *J. Biol. Chem.*, **2002**, *277*(37), 34287-34294.

[148] Nakagawa, T.; Yuan, J. Cross-talk between two cysteine protease families. Activation of caspase-12 by calpain in apoptosis. *J. Cell Biol.*, **2000**, *150*(4), 887-894.

[149] Rutkowski, D.T.; Kaufman, R.J. That which does not kill me makes me stronger: Adapting to chronic ER stress. *Trends Biochem. Sci.*, **2007**, *32*(10), 469-476.

[150] Yukioka, F.; Matsuzaki, S.; Kawamoto, K.; Koyama, Y.; Hitomi, J.; Katayama, T.; Tohyama, M. Presenilin-1 mutation activates the signaling pathway of caspase-4 in endoplasmic reticulum stress-

induced apoptosis. *Neurochem. Int.,* **2008**, *52*(4-5), 683-687.
[151] Fujita, E.; Kouroku, Y.; Jimbo, A.; Isoai, A.; Maruyama, K.; Momoi, T. Caspase-12 processing and fragment translocation into nuclei of tunicamycin-treated cells. *Cell Death Differ.,* **2002**, *9*(10), 1108-1114.
[152] Moorwood, C.; Barton, E.R. Caspase-12 ablation preserves muscle function in the mdx mouse. *Hum. Mol. Genet.,* **2014**, *23*(20), 5325-5341.
[153] Belizario, J.E.; Lorite, M.J.; Tisdale, M.J. Cleavage of caspases-1, -3, -6, -8 and -9 substrates by proteases in skeletal muscles from mice undergoing cancer cachexia. *Br. J. Cancer*, **2001**, *84*(8), 1135-1140.
[154] Coletti, D.; Yang, E.; Marazzi, G.; Sassoon, D. TNFalpha inhibits skeletal myogenesis through a PW1-dependent pathway by recruitment of caspase pathways. *EMBO J.*, **2002**, *21*(4), 631-642.
[155] Israel, A. The IKK complex: An integrator of all signals that activate NF-kappaB? *Trends Cell Biol.,* **2000**, *10*(4), 129-133.
[156] Relaix, F.; Weng, X.; Marazzi, G.; Yang, E.; Copeland, N.; Jenkins, N.; Spence, S.E.; Sassoon, D. Pw1, a novel zinc finger gene implicated in the myogenic and neuronal lineages. *Dev. Biol.,* **1996**, *177*(2), 383-396.
[157] Tisdale, M.J. Biomedicine. Protein loss in cancer Cachexia. *Science*, **2000**, *289*(5488), 2293-2294. Available from: http://science.sciencemag.org/content/289/5488/2293.full
[158] Tisdale, M.J. Cachexia in cancer patients. *Nat. Rev. Cancer,* **2002**, *2*(11), 862-871.
[159] Bonetto, A.; Aydogdu, T.; Kunzevitzky, N.; Guttridge, D.C.; Khuri, S.; Koniaris, L.G.; Zimmers, T.A. STAT3 activation in skeletal muscle links muscle wasting and the acute phase response in cancer cachexia. *PLoS One*, **2011**, *6*(7), e22538. Available from: http://journals.plos.org/plosone/article?id=10.1371/journal.pone.0022538
[160] Gilabert, M.; Calvo, E.; Airoldi, A.; Hamidi, T.; Moutardier, V.; Turrini, O.; Iovanna, J. Pancreatic cancer-induced cachexia is Jak2-dependent in mice. *J. Cell Physiol*., **2014**, *229*(10), 1437-1443.
[161] Johns, N.; Stephens, N.A.; Fearon, K.C. Muscle wasting in cancer. *Int. J. Biochem. Cell Biol.*, **2013**, *45*(10), 2215-2229.
[162] Tisdale, M.J. New cachexic factors. *Curr. Opin. Clin. Nutr. Metab. Care,* **1998**, *1*(3), 253-256.
[163] Watchorn, T.M.; Dowidar, N.; Dejong, C.H.; Waddell, I.D.; Garden, O.J.; Ross, J.A. The cachectic mediator proteolysis inducing factor activates NF-kappaB and STAT3 in human Kupffer cells and monocytes. *Int. J. Oncol.,* **2005**, *27*(4), 1105-1111.
[164] Bonetto, A.; Aydogdu, T.; Jin, X.; Zhang, Z.; Zhan, R.; Puzis, L.; Koniaris, L.G.; Zimmers, T.A. JAK/STAT3 pathway inhibition blocks skeletal muscle wasting downstream of IL-6 and in experimental cancer cachexia. *Am. J. Physiol. Endocrinol. Metab.,* **2012**, *303*(3), E410-E421.
[165] Du, J.; Wang, X.; Miereles, C.; Bailey, J.L.; Debigare, R.; Zheng, B.; Price, S.R.; Mitch, W.E. Activation of caspase-3 is an initial step triggering accelerated muscle proteolysis in catabolic conditions. *J. Clin. Invest.,* **2004**, *113*(1), 115-123.
[166] Pelosi, L.; Coggi, A.; Forcina, L.; Musaro, A. MicroRNAs modulated by local mIGF-1 expression in mdx dystrophic mice. *Front. Aging Neurosci.,* **2015**, *7*, 69. Available from: https://www.frontiersin.org/articles/10.3389/fnagi.2015.00069/full
[167] Price, F.D.; von Maltzahn, J.; Bentzinger, C.F.; Dumont, N.A.; Yin, H.; Chang, N.C.; Wilson, D.H.; Frenette, J.; Rudnicki, M.A. Inhibition of JAK-STAT signaling stimulates adult satellite cell function. *Nat. Med.,* **2014**, *20*(10), 1174-1181.
[168] Silva, K.A.; Dong, J.; Dong, Y.; Dong, Y.; Schor, N.; Tweardy, D.J.; Zhang, L.; Mitch, W.E. Inhibition of Stat3 activation suppresses caspase-3 and the ubiquitin-proteasome system, leading to preservation of muscle mass in cancer cachexia. *J. Biol. Chem.,* **2015**, *290*(17), 11177-11187.
[169] Tierney, M.T.; Aydogdu, T.; Sala, D.; Malecova, B.; Gatto, S.; Puri, P.L.; Latella, L.; Sacco, A. STAT3 signaling controls satellite cell expansion and skeletal muscle repair. *Nat. Med.,* **2014**, *20*(10), 1182-1186.
[170] Wang, X.H.; Zhang, L.; Mitch, W.E.; LeDoux, J.M.; Hu, J.; Du, J. Caspase-3 cleaves specific 19 S proteasome subunits in skeletal muscle stimulating proteasome activity. *J. Biol. Chem.,* **2010**, *285*(28), 21249-21257.
[171] Dick, S.A.; Megeney, L.A. Cell death proteins: An evolutionary role in cellular adaptation before the advent of apoptosis. *Bioessays,* **2013**, *35*(11), 974-983.
[172] Dick, S.A.; Chang, N.C.; Dumont, N.A.; Bell, R.A.; Putinski, C.; Kawabe, Y.; Litchfield, D.W.; Rudnicki, M.A.; Megeney, L.A. Caspase 3 cleavage of Pax7 inhibits self-renewal of satellite cells. *Proc. Natl. Acad. Sci. U.S.A.,* **2015**, *112*(38), E5246-E5252.
[173] Olguin, H.C. Regulation of Pax7 protein levels by caspase-3 and proteasome activity in differentiating myoblasts. *Biol. Res.,* **2011**, *44*(4), 323-327.
[174] Murray, T.V.; McMahon, J.M.; Howley, B.A.; Stanley, A.; Ritter, T.; Mohr, A.; Zwacka, R.; Fearnhead, H.O. A non-apoptotic role for caspase-9 in muscle differentiation. *J. Cell Sci.*, **2008**, *121*(Pt 22), 3786-3793.
[175] Griffiths, G.S.; Doe, J.; Jijiwa, M.; Van Ry, P.; Cruz, V.; De, V.; Ramos, J.W.; Burkin, D.J.; Matter, M.L. Bit-1 is an essential regulator of myogenic differentiation. *J. Cell Sci.,* **2015**, *128*(9), 1707-1717.
[176] Bell, R.A.; Al Khalaf, M.; Megeney, L.A. The beneficial role of proteolysis in skeletal muscle growth and stress adaptation. *Skelet. Muscle*, **2016**, *6*, 16. Available from: https://skeletalmusclejournal.biomedcentral.com/articles/10.1186/s13395-016-0086-6
[177] Wang, X.H.; Mitch, W.E. Mechanisms of muscle wasting in chronic kidney disease. *Nat. Rev. Nephrol.,* **2014**, *10*(9), 504-516.
[178] Cecconi, F.; Levine, B. The role of autophagy in mammalian development: Cell makeover rather than cell death. *Dev. Cell*, **2008**, *15*(3), 344-357.
[179] Levine, B.; Kroemer, G. Autophagy in the pathogenesis of disease. *Cell*, **2008**, *132*(1), 27-42.
[180] Mizushima, N.; Yamamoto, A.; Matsui, M.; Yoshimori, T.; Ohsumi, Y. *In vivo* analysis of autophagy in response to nutrient starvation using transgenic mice expressing a fluorescent autophagosome marker. *Mol. Biol. Cell*, **2004**, *15*(3), 1101-1111.
[181] Tanida, I.; Ueno, T.; Kominami, E. LC3 conjugation system in mammalian autophagy. *Int. J. Biochem. Cell Biol.,* **2004**, *36*(12), 2503-2518.
[182] Tanida, I.; Ueno, T.; Kominami, E. Human light chain 3/MAP1LC3B is cleaved at its carboxyl-terminal Met121 to expose Gly120 for lipidation and targeting to autophagosomal membranes. *J. Biol. Chem.,* **2004**, *279*(46), 47704-47710.
[183] Galluzzi, L.; Vicencio, J.M.; Kepp, O.; Tasdemir, E.; Maiuri, M.C.; Kroemer, G. To die or not to die: That is the autophagic question. *Curr. Mol. Med.,* **2008**, *8*(2), 78-91.
[184] Grumati, P.; Coletto, L.; Schiavinato, A.; Castagnaro, S.; Bertaggia, E.; Sandri, M.; Bonaldo, P. Physical exercise stimulates autophagy in normal skeletal muscles but is detrimental for collagen VI-deficient muscles. *Autophagy*, **2011**, *7*(12), 1415-1423.
[185] Iqbal, S.; Hood, D.A. Oxidative stress-induced mitochondrial fragmentation and movement in skeletal muscle myoblasts. *Am. J. Physiol Cell Physiol.,* **2014**, *306*(12), C1176-C1183.
[186] Kirkwood, S.P.; Packer, L.; Brooks, G.A. Effects of endurance training on a mitochondrial reticulum in limb skeletal muscle. *Arch. Biochem. Biophys.,* **1987**, *255*(1), 80-88.
[187] Lira, V.A.; Okutsu, M.; Zhang, M.; Greene, N.P.; Laker, R.C.; Breen, D.S.; Hoehn, K.L.; Yan, Z. Autophagy is required for exercise training-induced skeletal muscle adaptation and improvement of physical performance. *FASEB J.,* **2013**, *27*(10), 4184-4193.
[188] Perry, B.D.; Wyckelsma, V.L.; Murphy, R.M.; Steward, C.H.; Anderson, M.; Levinger, I.; Petersen, A.C.; McKenna, M.J. Dissociation between short-term unloading and resistance training effects on skeletal muscle Na+,K+-ATPase, muscle function, and fatigue in humans. *J. Appl. Physiol.,* **2016**, *121*(5), 1074-1086.
[189] Picard, M.; Gentil, B.J.; McManus, M.J.; White, K.; St Louis, K.; Gartside, S.E.; Wallace, D.C.; Turnbull, D.M. Acute exercise remodels mitochondrial membrane interactions in mouse skeletal muscle. *J. Appl. Physiol.,* **2013**, *115*(10), 1562-1571.
[190] Sandri, M. Autophagy in skeletal muscle. *FEBS Lett.*, **2010**, *584*(7), 1411-1416.
[191] Safdar, A.; Little, J.P.; Stokl, A.J.; Hettinga, B.P.; Akhtar, M.; Tarnopolsky, M.A. Exercise increases mitochondrial PGC-1alpha content and promotes nuclear-mitochondrial cross-talk to coordi-

nate mitochondrial biogenesis. *J. Biol. Chem.,* **2011**, *286*(12), 10605-10617.

[192] Zampieri, S. Lifelong physical exercise delays age-associated skeletal muscle decline. *J. Gerontol. A Biol. Sci. Med. Sci.,* **2015**, *70*(2), 163-173.

[193] Carnio, S.; LoVerso, F.; Baraibar, M.A.; Longa, E.; Khan, M.M.; Maffei, M.; Reischl, M.; Canepari, M.; Loefler, S.; Kern, H.; Blaauw, B.; Friguet, B.; Bottinelli, R.; Rudolf, R.; Sandri, M. Autophagy impairment in muscle induces neuromuscular junction degeneration and precocious aging. *Cell Rep.,* **2014**, *8*(5), 1509-1521.

[194] Nascimbeni, A.C.; Fanin, M.; Angelini, C.; Sandri, M. Autophagy dysregulation in Danon disease. *Cell Death Dis.,* **2017**, *8*(1), e2565. Available from: https://www.nature.com/articles/cddis2016475

[195] Garcia-Prat, L.; Munoz-Canoves, P.; Martinez-Vicente, M. Dysfunctional autophagy is a driver of muscle stem cell functional decline with aging. *Autophagy,* **2016**, *12*(3), 612-613.

[196] Castagnaro, S.; Pellegrini, C.; Pellegrini, M.; Chrisam, M.; Sabatelli, P.; Toni, S.; Grumati, P.; Ripamonti, C.; Pratelli, L.; Maraldi, N.M.; Cocchi, D.; Righi, V.; Faldini, C.; Sandri, M.; Bonaldo, P.; Merlini, L. Autophagy activation in COL6 myopathic patients by a low-protein-diet pilot trial. *Autophagy,* **2016**, *12*(12), 2484-2495.

[197] Penna, F.; Costamagna, D.; Pin, F.; Camperi, A.; Fanzani, A.; Chiarpotto, E.M.; Cavallini, G.; Bonelli, G.; Baccino, F.M.; Costelli, P. Autophagic degradation contributes to muscle wasting in cancer cachexia. *Am. J. Pathol.,* **2013**, *182*(4), 1367-1378.

[198] Mammucari, C.; Milan, G.; Romanello, V.; Masiero, E.; Rudolf, R.; Del Piccolo, P.; Burden, S.J.; Di Lisi, R.; Sandri, C.; Zhao, J.; Goldberg, A.L.; Schiaffino, S.; Sandri, M. FoxO3 controls autophagy in skeletal muscle *in vivo*. *Cell Metab.,* **2007**, *6*(6), 458-471.

[199] Mizushima, N.; Levine, B.; Cuervo, A.M.; Klionsky, D.J. Autophagy fights disease through cellular self-digestion. *Nature,* **2008**, *451*(7182), 1069-1075. Available from: https://www.nature.com/articles/nature06639

[200] Mofarrahi, M.; Sigala, I.; Guo, Y.; Godin, R.; Davis, E.C.; Petrof, B.; Sandri, M.; Burelle, Y.; Hussain, S.N. Autophagy and skeletal muscles in sepsis. *PLoS One,* **2012**, *7*(10), e47265. Available from: http://journals.plos.org/plosone/article?id=10.1371/journal.pone.0047265

[201] Qiu, J.; Tsien, C.; Thapalaya, S.; Narayanan, A.; Weihl, C.C.; Ching, J.K.; Eghtesad, B.; Singh, K.; Fu, X.; Dubyak, G.; McDonald, C.; Almasan, A.; Hazen, S.L.; Naga Prasad, S.V.; Dasarathy, S. Hyperammonemia-mediated autophagy in skeletal muscle contributes to sarcopenia of cirrhosis. *Am. J. Physiol. Endocrinol. Metab.,* **2012**, *303*(8), E983-E993.

[202] Giusto, M.; Barberi, L.; Di Sario, F.; Rizzuto, E.; Nicoletti, C.; Ascenzi, F.; Renzi, A.; Caporaso, N.; D'Argenio, G.; Gaudio, E.; Musaro, A.; Merli, M. Skeletal muscle myopenia in mice model of bile duct ligation and carbon tetrachloride-induced liver cirrhosis. *Physiol. Rep*., **2017**, *5*(7), e13153. Available from: http://physreports.physiology.org/content/5/7/e13153

[203] Brocca, L.; Cannavino, J.; Coletto, L.; Biolo, G.; Sandri, M.; Bottinelli, R.; Pellegrino, M.A. The time course of the adaptations of human muscle proteome to bed rest and the underlying mechanisms. *J. Physiol.,* **2012**, *590*(20), 5211-5230.

[204] O'Leary, M.F.; Vainshtein, A.; Carter, H.N.; Zhang, Y.; Hood, D.A. Denervation-induced mitochondrial dysfunction and autophagy in skeletal muscle of apoptosis-deficient animals. *Am. J. Physiol. Cell Physiol.,* **2012**, *303*(4), C447-C454.

[205] Zhao, J.; Brault, J.J.; Schild, A.; Cao, P.; Sandri, M.; Schiaffino, S.; Lecker, S.H.; Goldberg, A.L. FoxO3 coordinately activates protein degradation by the autophagic/lysosomal and proteasomal pathways in atrophying muscle cells. *Cell Metab.,* **2007**, *6*(6), 472-483.

[206] Aucello, M.; Dobrowolny, G.; Musaro, A. Localized accumulation of oxidative stress causes muscle atrophy through activation of an autophagic pathway. *Autophagy,* **2009**, *5*(4), 527-529.

[207] Klionsky, D.J.; Abdelmohsen, K.; Abe, A.; Abedin, M.J.; Abeliovich, H.; Acevedo, A.A.; Adachi, H.; Adams, C.M.; Adams, P.D.; Adeli, K.; Adhihetty, P.J.; Adler, S.G.; Agam, G.; Agarwal, R.; Aghi, M.K.; Agnello, M.; Agostinis, P.; Aguilar, P.V.; Aguirre-Ghiso, J.; Airoldi, E.M.; Ait-Si-Ali, S.; Akematsu, T.; Akporiaye, E.T.; Al Rubeai, M.; Albaiceta, G.M.; Albanese, C.; Albani, D.; Albert, M.L.; Aldudo, J.; Algul, H.; Alirezaei, M.; Alloza, I.; Almasan, A.; Almonte-Beceril, M.; Alnemri, E.S.; Alonso, C.; Altan-Bonnet, N.; Altieri, D.C.; Alvarez, S.; Alvarez-Erviti, L.; Alves, S.; Amadoro, G.; Amano, A.; Amantini, C.; Ambrosio, S.; Amelio, I.; Amer, A.O.; Amessou, M.; Amon, A.; An, Z.; Anania, F.A.; Andersen, S.U.; Andley, U.P.; Andreadi, C.K.; Andrieu-Abadie, N.; Anel, A.; Ann, D.K.; Anoopkumar-Dukie, S.; Antonioli, M.; Aoki, H.; Apostolova, N.; Aquila, S.; Aquilano, K.; Araki, K.; Arama, E.; Aranda, A.; Araya, J.; Arcaro, A.; Arias, E.; Arimoto, H.; Ariosa, A.R.; Armstrong, J.L.; Arnould, T.; Arsov, I.; Asanuma, K.; Askanas, V.; Asselin, E.; Atarashi, R.; Atherton, S.S.; Atkin, J.D.; Attardi, L.D.; Auberger, P.; Auburger, G.; Aurelian, L.; Autelli, R.; Avagliano, L.; Avantaggiati, M.L.; Avrahami, L.; Awale, S.; Azad, N.; Bachetti, T.; Backer, J.M.; Bae, D.H.; Bae, J.S.; Bae, O.N.; Bae, S.H.; Baehrecke, E.H.; Baek, S.H.; Baghdiguian, S.; Bagniewska-Zadworna, A.; Bai, H.; Bai, J.; Bai, X.Y.; Bailly, Y.; Balaji, K.N.; Balduini, W.; Ballabio, A.; Balzan, R.; Banerjee, R.; Banhegyi, G.; Bao, H.; Barbeau, B.; Barrachina, M.D.; Barreiro, E.; Bartel, B.; Bartolome, A.; Bassham, D.C.; Bassi, M.T.; Bast, R.C., Jr.; Basu, A.; Batista, M.T.; Batoko, H.; Battino, M.; Bauckman, K.; Baumgarner, B.L.; Bayer, K.U.; Beale, R.; Beaulieu, J.F.; Beck, G.R., Jr.; Becker, C.; Beckham, J.D.; Bedard, P.A.; Bednarski, P.J.; Begley, T.J.; Behl, C.; Behrends, C.; Behrens, G.M.; Behrns, K.E.; Bejarano, E.; Belaid, A.; Belleudi, F.; Benard, G.; Berchem, G.; Bergamaschi, D.; Bergami, M.; Berkhout, B.; Berliocchi, L.; Bernard, A.; Bernard, M.; Bernassola, F.; Bertolotti, A.; Bess, A.S.; Besteiro, S.; Bettuzzi, S.; Bhalla, S.; Bhattacharyya, S.; Bhutia, S.K.; Biagosch, C.; Bianchi, M.W.; Biard-Piechaczyk, M.; Billes, V.; Bincoletto, C.; Bingol, B.; Bird, S.W.; Bitoun, M.; Bjedov, I.; Blackstone, C.; Blanc, L.; Blanco, G.A.; Blomhoff, H.K.; Boada-Romero, E.; Bockler, S.; Boes, M.; Boesze-Battaglia, K.; Boise, L.H.; Bolino, A.; Boman, A.; Bonaldo, P.; Bordi, M.; Bosch, J.; Botana, L.M.; Botti, J.; Bou, G.; Bouche, M.; Bouchecareilh, M.; Boucher, M.J.; Boulton, M.E.; Bouret, S.G.; Boya, P.; Boyer-Guittaut, M.; Bozhkov, P.V.; Brady, N.; Braga, V.M.; Brancolini, C.; Braus, G.H.; Bravo-San Pedro, J.M.; Brennan, L.A.; Bresnick, E.H.; Brest, P.; Bridges, D.; Bringer, M.A.; Brini, M.; Brito, G.C.; Brodin, B.; Brookes, P.S.; Brown, E.J.; Brown, K.; Broxmeyer, H.E.; Bruhat, A.; Brum, P.C.; Brumell, J.H.; Brunetti-Pierri, N.; Bryson-Richardson, R.J.; Buch, S.; Buchan, A.M.; Budak, H.; Bulavin, D.V.; Bultman, S.J.; Bultynck, G.; Bumbasirevic, V.; Burelle, Y.; Burke, R.E.; Burmeister, M.; Butikofer, P.; Caberlotto, L.; Cadwell, K.; Cahova, M.; Cai, D.; Cai, J.; Cai, Q.; Calatayud, S.; Camougrand, N.; Campanella, M.; Campbell, G.R.; Campbell, M.; Campello, S.; Candau, R.; Caniggia, I.; Cantoni, L.; Cao, L.; Caplan, A.B.; Caraglia, M.; Cardinali, C.; Cardoso, S.M.; Carew, J.S.; Carleton, L.A.; Carlin, C.R.; Carloni, S.; Carlsson, S.R.; Carmona-Gutierrez, D.; Carneiro, L.A.; Carnevali, O.; Carra, S.; Carrier, A.; Carroll, B. Guidelines for the use and interpretation of assays for monitoring autophagy (3rd edition). *Autophagy,* **2016**, *12*(1), 1-222.

[208] Ahtiainen, J.P.; Hulmi, J.J.; Lehti, M.; Kraemer, W.J.; Nyman, K.; Selanne, H.; Alen, M.; Komulainen, J.; Kovanen, V.; Mero, A.A.; Philippou, A.; Laakkonen, E.K.; Hakkinen, K. Effects of resistance training on expression of IGF-I splice variants in younger and older men. *Eur. J. Sport Sci.,* **2016**, *16*(8), 1055-1063.

[209] Tsai, C.L.; Wang, C.H.; Pan, C.Y.; Chen, F.C. The effects of long-term resistance exercise on the relationship between neurocognitive performance and GH, IGF-1, and homocysteine levels in the elderly. *Front. Behav. Neurosci.,* **2015**, *9*, 23. Available from: https://www.frontiersin.org/articles/10.3389/fnbeh.2015.00023/full

[210] Arnarson, A.; Gudny, G.O.; Ramel, A.; Jonsson, P.V.; Thorsdottir, I. Insulin-like growth factor-1 and resistance exercise in community dwelling old adults. *J. Nutr. Health Aging,* **2015**, *19*(8), 856-860.

[211] Raue, U.; Slivka, D.; Jemiolo, B.; Hollon, C.; Trappe, S. Proteolytic gene expression differs at rest and after resistance exercise between young and old women. *J. Gerontol. A Biol. Sci. Med. Sci.,* **2007**, *62*(12), 1407-1412.

[212] Luo, L.; Lu, A.M.; Wang, Y.; Hong, A.; Chen, Y.; Hu, J.; Li, X.; Qin, Z.H. Chronic resistance training activates autophagy and reduces apoptosis of muscle cells by modulating IGF-1 and its receptors, Akt/mTOR and Akt/FOXO3a signaling in aged rats. *Exp. Gerontol.,* **2013**, *48*(4), 427-436.

[213] Kim, Y.; Triolo, M.; Hood, D.A. Impact of aging and exercise on mitochondrial quality control in skeletal muscle. *Oxid. Med. Cell. Longev.,* **2017**, *2017*, 3165396. Available from: https://www.hindawi.com/journals/omcl/2017/3165396/

[214] Anthony, T.G.; Anthony, J.C.; Yoshizawa, F.; Kimball, S.R.; Jefferson, L.S. Oral administration of leucine stimulates ribosomal protein mRNA translation but not global rates of protein synthesis in the liver of rats. *J. Nutr.,* **2001**, *131*(4), 1171-1176.

[215] Marzani, B.; Balage, M.; Venien, A.; Astruc, T.; Papet, I.; Dardevet, D.; Mosoni, L. Antioxidant supplementation restores defective leucine stimulation of protein synthesis in skeletal muscle from old rats. *J. Nutr.,* **2008**, *138*(11), 2205-2211.

[216] Jong, C.J.; Ito, T.; Schaffer, S.W. The ubiquitin-proteasome system and autophagy are defective in the taurine-deficient heart. *Amino Acids.,* **2015**, *47*(12), 2609-2622.

[217] Scicchitano, B.M.; Sica, G. The beneficial effects of taurine to counteract sarcopenia. *Curr. Protein Pept. Sci.,* **2016**.

[218] Xia, Z.; Cholewa, J.M.; Zhao, Y.; Yang, Y.; Shang, H.; Jiang, H.; Su, Q.; Zanchi, N.E. A potential strategy for counteracting age-related sarcopenia: Preliminary evidence of combined exercise training and leucine supplementation. *Food Funct.,* **2017**, *8*(12), 4528-4538.