Pathophysiology and mechanisms of primary sarcopenia (Review)

HIROKI NISHIKAWA[1,2], SHINYA FUKUNISHI[1,2], AKIRA ASAI[1], KEISUKE YOKOHAMA[1], SHUHEI NISHIGUCHI[3] and KAZUHIDE HIGUCHI[1]

[1]The Second Department of Internal Medicine, [2]Premier Departmental Research of Medicine, Osaka Medical and Pharmaceutical University, Takatsuki, Osaka 569-8686; [3]Department of Internal Medicine, Kano General Hospital, Osaka 531-0041, Japan

DOI: 10.3892/ijmm_xxxxxxxx

Abstract. Aging causes skeletal muscle atrophy, and myofiber loss can be a critical component of this process. In 1989, Rosenberg emphasized the importance of the loss of skeletal muscle mass that occurs with aging and coined the term 'sarcopenia'. Since then, sarcopenia has attracted considerable attention due to the aging population in developed countries. The presence of sarcopenia is closely related to staggering, falls and even frailty in the elderly, which in turn leads to the need for nursing care. Sarcopenia is often associated with a poor prognosis in the elderly. Therefore, it is crucial to investigate the causes and pathogenesis of sarcopenia, and to develop and introduce interventional strategies in line with these causes and pathogenesis. Sarcopenia can be a primary component of physical frailty. The association between sarcopenia, frailty and locomotive syndrome is complex; however, sarcopenia is a muscle-specific concept that is relatively easy to approach in research. In the elderly, a lack of exercise, malnutrition and hormonal changes lead to neuromuscular junction insufficiency, impaired capillary blood flow, reduced repair and regeneration capacity due to a decrease in the number of muscle satellite cells, the infiltration of inflammatory cells and oxidative stress, resulting in muscle protein degradation exceeding synthesis. In addition, mitochondrial dysfunction causes metabolic abnormalities, such as insulin resistance, which may lead to quantitative and qualitative abnormalities in skeletal muscle, resulting in sarcopenia. The present review article focuses on age-related primary sarcopenia and outlines its pathogenesis and mechanisms.

Correspondence to: Dr Hiroki Nishikawa, The Second Department of Internal Medicine, Osaka Medical and Pharmaceutical University, 2-7 Daigakumachi, Takatsuki, Osaka 569-8686, Japan
E-mail: nishikawa_6392_0207@yahoo.co.jp

Abbreviations: IGF-1, insulin-like growth factor-1; mTOR, mammalian target of rapamycin; mTORC1, mammalian target of rapamycin complex 1; GH, growth hormone; PI3K, phosphoinositide3-kinase; TNF-α, tumor necrosis factor-α; ROS, reactive oxygen species; FGF-2, fibroblast growth factor 2; HGF, hepatocyte growth factor; MAPK, mitogen-activated protein kinase; RAS, renin-angiotensin system; ACE, angiotensin-converting enzyme

Key words: primary sarcopenia, mechanism, myofiber, satellite cell, myokine

Contents

1. Introduction

In 1989, Rosenberg (1) emphasized the importance of the loss of skeletal muscle mass that occurs with aging and coined the term 'sarcopenia'. Since then, sarcopenia has attracted considerable attention due to the aging population in developed countries. Lexell *et al* (2) reported that skeletal muscle mass was reduced by ~50% in elderly compared with young individuals, based on analyses using muscles obtained from autopsies. In general, the skeletal muscle area and muscle strength of elderly individuals decreases by 25-30% and 30-40%, respectively, compared with those in their 20s, and muscle mass decreases by 1-2% each year after the age of 50 (3,4). The presence of sarcopenia is closely related to staggering, falls and even frailty in the elderly, which in turn leads to the need for nursing care (3). Therefore, it is crucial to investigate the causes and pathogenesis of sarcopenia, and to develop and introduce interventional strategies in line with these causes and pathogenesis. In addition, nursing care prevention, and medical and nursing care policies also require attention in Japan which has entered a super-aging society (3).

The association between sarcopenia, frailty and locomotive syndrome is complex; however, sarcopenia is a muscle-specific concept that is relatively easy to approach in research. At the organ level, it is known that specific changes in the muscles of the elderly involve a decrease in fast-twitch muscle components and the accumulation of fat in muscles, and at the cellular

level, a mitochondrial dysfunction occurs (5-7). Age-related sarcopenia is termed primary sarcopenia, and disease-related sarcopenia is termed secondary sarcopenia (8,9). Sarcopenia can be a primary component of physical frailty. Sarcopenia is also a main health concern in the era of the COVID-19 pandemic. Sarcopenia can be an adverse predictor in elderly patients with COVID-19 infection (10,11).

The present review article focuses on age-related primary sarcopenia and outlines its pathogenesis and mechanisms.

2. Myofiber and muscle satellite cells

Multiple factors have been proposed to explain the pathogenesis of primary sarcopenia. Myofibers are multinucleated cells formed by the fusion of satellite cells. Skeletal muscle is an organ that is susceptible to damage from overload and trauma; however, it has a notable ability to regenerate. Satellite cells, known as skeletal muscle-specific somatic stem cells, play a central role in the process of muscle regeneration (12-15). Satellite cells are normally dormant; however, when nearby muscle fibers are damaged, they are stimulated by damaged myofibers to become active and form muscle progenitor cells. Cells that proliferate by division fuse with each other or with existing muscle fibers, contributing to the formation, repair and hypertrophy of new myofibers (12-15). Myofibers are classified into two major types (four subtypes) according to the isoform of myosin heavy chain: Type I, IIa, IIx and IIb (16). Myofibers are commanded to contract and relax by neuromuscular junctions, and receive blood flow from surrounding capillaries (5-7). Damaged myofibers are repaired and regenerated by satellite cells, which are bone marrow stem cells (5-7). In addition, mitochondria are abundant in the cells and are involved not only in energy production, mainly through fatty acid beta-oxidation, but also in metabolic regulation, such as insulin sensitivity (15).

The age-related loss of skeletal muscle mass is caused by a decrease in the number of myofibers and the atrophy of individual myofibers, while disuse muscle atrophy due to a long-term bed ridden status and related disuse, which is the cause of secondary sarcopenia, is mainly due to a decrease in the cross-sectional area of myofibers (Table I) (5,14). In disuse atrophy, the time course is acute, the degree is severe, the recovery is often reversible, and the slow-twitch muscles are mainly affected, whereas in primary sarcopenia, the time course is chronic, the degree is mild, the recovery is sometimes irreversible, and the fast-twitch muscles are mainly affected (Table I) (17). As mentioned above, skeletal myofibers are classified into two major types: Type I (slow-twitch fibers) and type II (fast-twitch fibers) fibers, and a decrease in the number of type II fibers is observed from an early stage with aging, eventually resulting in a decrease in the number of both types of myofibers (5,6). The motor neurons that innervate myofibers are located in the spinal cord, and the nerve fibers that emerge from these neurons branch out in multiple directions to reach the muscle fibers (7). The motor neurons and the myofibers they innervate are collectively called motor units, and it is known that these motor units decrease with aging (7). In addition, it has been reported that aging causes morphological changes in neuromuscular synapses, resulting in the functional decline of skeletal muscles and muscle atrophy (18). Muscle satellite cells exist between the plasma membrane and basement membrane of muscle fibers and are normally dormant; however, they are activated by stimulation, proliferate, differentiate and fuse with existing muscle fibers, playing an important role in muscle regeneration (5-7). Aging causes a loss of function of muscle satellite cells, a decrease in the regenerative capacity of myofibers, and a decrease in the number of myofibers (12,19). Muscle regeneration is maintained by the infiltration of macrophages and the subsequent activation of satellite cells (12). The expression of notch ligand (Delta) is decreased in senescent muscle satellite cells, which may be involved in the decreased proliferative potential of satellite cells (20). In addition, it has been reported that Wnt signaling is also enhanced in senescent satellite cells, which promotes their differentiation into fibrogenic cells (21). The repair process of damaged skeletal muscle from the perspective of muscle satellite cells is illustrated in Fig. 1.

3. Protein synthesis and degradation in muscle

The atrophy or hypertrophy of myofibers is dependent on their protein content. Over 80% of the dry weight of muscle is comprised of protein (22). Theoretically, muscle hypertrophy occurs when muscle protein synthesis is increased and degradation is inhibited, while muscle atrophy occurs when degradation is increased and synthesis is inhibited. Muscle protein anabolism in muscle cells is known to be mediated by the following: i) Amino acids (branched chain amino acids, such as leucine); ii) exercise; iii) insulin and insulin-like growth factor-1 (IGF-1); and iv) hormones (23-26). All these factors induce the phosphorylation of mammalian target of rapamycin (mTOR) in myocytes (27). They also exhibit protein anabolism through the activation of 70-kDa ribosomal protein S6 kinase (p70S6K) and eukaryotic initiation factor 4E binding protein-1 (4E-BP1) (27).

The mTOR complex 1 (mTORC1) signaling pathway is a major regulator of protein metabolism (28). mTORC1 regulates protein synthesis and degradation by integrating a number of intracellular signals (28). For example, leucine intake and exercise activate mTORC1, leading to increased protein synthesis. On the other hand, during fasting, mTORC1 is inactivated and protein degradation is enhanced (28). The age-related loss of skeletal muscle mass is less likely to lead to the diet-induced enhancement of protein synthesis in the elderly due to the decreased sensitivity of mTORC1 to leucine (29). It has been shown that leucine is not only an organelle of muscle proteins, but also acts directly on muscle cells to induce protein synthesis (13). In addition, IGF-1, a potent anabolic factor, is regulated by growth hormone (GH) and is produced mainly in the liver (30). Ghrelin, a GH-promoting peptide, not only promotes GH secretion, but also has the function of promoting central or peripheral feeding (31). IGF-1 is involved in a number of anabolic pathways in skeletal muscle, including cell proliferation, differentiation and metabolism and muscle regeneration (32,33). As mentioned above, one of the causes of sarcopenia is decreased muscle synthesis; IGF-I activates the intracellular signaling pathways of phosphoinositide3-kinase (PI3K) and Akt, and further activates downstream mTOR, which enhances protein synthesis (34,35). The IGF-1/PI3K/mTOR system is important in muscle

Table I. Clinical and pathophysiological features of primary sarcopenia and disuse muscle atrophy.

Feature	Primary sarcopenia	Disuse muscle atrophy
Clinical course	Chronic	Acute
Degree of muscle damage	Mild	Severe
Recovery	Sometimes irreversible	Often reversible
Mainly affected muscle	Fast-twitch muscles	Slow-twitch muscles
Myofiber	Decrease in the number of myofiber. A decrease in the number of type II fibers is observed from an early stage with aging	Decrease in the cross-sectional area of myofibers
Motor neuron function	Often damaged	Often maintained

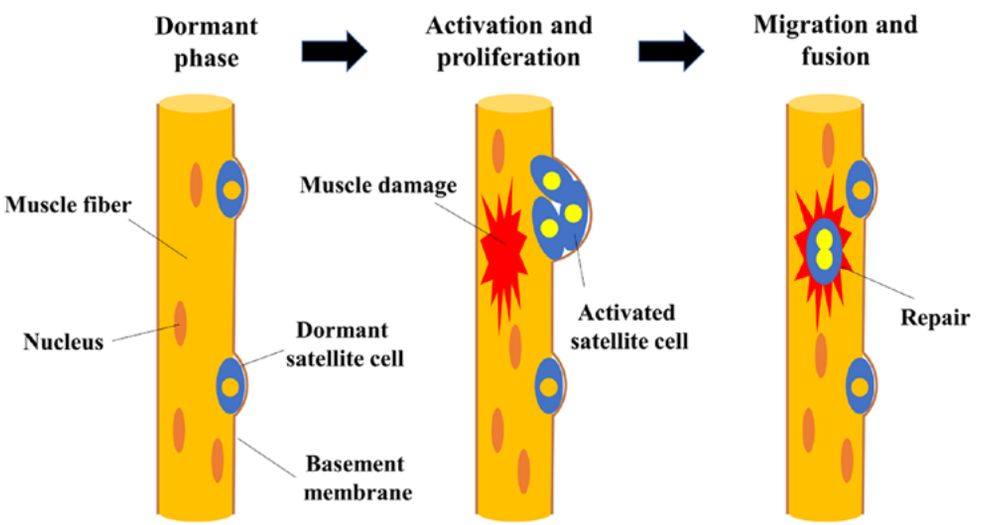

Figure 1. Muscle satellite cells during muscle regeneration. Muscle satellite cells reside between the muscle cell membrane and the basement membrane, and they are dormant. When a muscle fiber is damaged, satellite cells are activated, proliferate, and fuse to the muscle fiber to repair the damaged area. Some of the activated satellite cells return to their dormant state.

hypertrophy; however, its activity decreases with aging (36). The second is the enhancement of muscle breakdown. Ubiquitin is an approximately 8.5-kDa protein with a high degree of sequence conservation among different species and exists in a ubiquitinated (ubiquitylation, a type of protein modification) state (37,38). When a protein is ubiquitinated in the cell, the proteasome is able to degrade it. In 2001, the muscle-specific ubiquitin ligase genes, muscle-specific RING finger protein 1 (MuRF1) and Atrogin-1 (muscle atrophy-related factors), were identified (37,38). Atrorgin-1 is encoded by the Fbxo32 gene, which is also referred to as a muscle atrophy-related factor, and is upregulated in a wide range of pathological conditions, such as neurectomy and disuse; however, mice in which Atrorgin-1 is knocked out are less susceptible to neurectomy-induced muscle atrophy (37).

It has also been reported that muscle atrophy-related factor is increased in skeletal muscle of elderly individuals and aging rats (39,40). Protein synthesis in muscle decreases with aging, and protein anabolism is suppressed in the muscles of the elderly even when the same amounts of amino acids are present in the blood (i.e., anabolic resistance) (41). The mTOR activation response to amino acids, such as leucin that can stimulate mTOR phosphorylation is reduced in the elderly (41,42).

4. Immunological dysfunction and inflammation with aging

Elderly individuals are more likely to develop chronic inflammation, which is a persistent mild inflammation, due to the decline in immune function caused by aging. The risk of developing inflammatory diseases, such as infections and collagen diseases is increased in the elderly with an impaired immune function (43). These chronic inflammations are characterized by mildly elevated blood levels of pro-inflammatory cytokines, such as tumor necrosis factor-α (TNF-α), interleukin (IL)-1β, IL-6 and IL-18. C-reactive protein (CRP), an acute-phase protein produced by the liver in response to IL-6, is also upregulated during chronic inflammation (44). Blood levels of TNF-α, IL-1 β, and IL-6 have been reported to increase 2- to 4-fold in the elderly compared with healthy young adults (45). It has been also shown that the administration

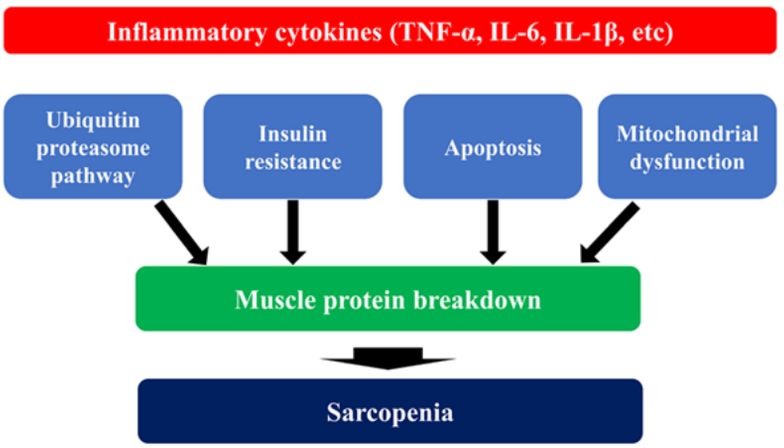

Figure 2. Inflammatory cytokines and sarcopenia. TNF-α, tumor necrosis factor-α; IL, interleukin.

of IL-6 and TNF-α to rats causes the degradation of skeletal muscle (46,47). Inflammatory cytokines cause the dysfunction of mitochondria, which are involved in energy production, resulting in a decreased ATP production, as well as in the excessive production of reactive oxygen species (ROS) (48,49). Excessive ROS production further exacerbates mitochondrial damage and subsequent metabolic abnormalities, and induces proteolysis by enhancing the ubiquitin-proteasome system, one of the major pathways for protein degradation as described above, resulting in skeletal muscle atrophy (50,51). Proteins labeled with ubiquitin are degraded by the proteasome, a large enzyme complex (52). Apoptosis, on the other hand, is a cell death mechanism that removes unnecessary cells. The activation of caspases, proteolytic enzymes, rapidly degrades intracellular proteins, which are ultimately phagocytosed by macrophages and other phagocytic cells (53). TNF-α is a major regulator of the apoptotic signaling pathway. TNF-α binds to TNF-α receptors in skeletal muscle and activates caspases through the Fas-associated death domain (FADD), thereby inducing apoptosis (54). Excessive apoptosis in skeletal muscle leads to increased degradation of muscle proteins, resulting in muscle atrophy (55).

Obesity is another important factor in the development of chronic inflammation. In recent years, it has been shown that adipose tissue interacts with immune cells, such as macrophages and neutrophils to induce chronic inflammation in obese individuals (56). TNF-α secreted by macrophages increases free fatty acids by promoting lipolysis through the Toll-like receptor 4 (TLR4) signaling pathway in adipose tissue (56). In addition, the macrophage response to free fatty acids increases the secretion of pro-inflammatory cytokines, such as TNF-α, IL-1β and IL-6, further exacerbating chronic inflammation (57). Inflammation-associated immune cell infiltration is found not only in adipose tissue, but also in skeletal muscle; in a study on critically ill hospitalized patients aged 50-59 years, the increased infiltration of CD68-positive macrophages into skeletal muscle was observed with atrophy of the rectus femoris muscle after 7 days of hospitalization (58). Thus, chronic inflammation induced by various factors in aging is considered to reduce muscle strength and function by increasing macrophage infiltration into skeletal muscle, decreasing muscle mass and increasing the accumulation of ectopic fat (59). Recently, sarcopenic obesity, a condition that involves both sarcopenia and obesity, has been attracting attention. Patients with sarcopenic obesity have a poorer prognosis than those with sarcopenia alone or obesity alone (60). The association between inflammatory cytokines and sarcopenia is illustrated in Fig. 2.

5. Myokines and sarcopenia

Biologically active substances produced by muscle cells are termed myokines, and IGF-1, IL-6, fibroblast growth factor 2 (FGF-2), hepatocyte growth factor (HGF) and IL-15 are representative myokines (61,62). Some myokines act endocrinologically on organs throughout the body (e.g., pancreas, brain, adipose tissue), while others act paracrine or autocrine on skeletal muscle itself (61,62). Pedersen *et al* (63) defined myokines as 'cytokines and peptides expressed in and secreted from skeletal myofibers that act in a paracrine and endocrine manner'. Myokines released from damaged myofibers act as messengers in the process of muscle regeneration by satellite cells upon muscle injury. When myofibers are damaged, cytokines and chemokines are first secreted by macrophages that migrate to the damaged area, and growth factors are also released from the damaged myofibers, which act on satellite cells to initiate muscle regeneration (64). Growth factors play a role in regulating the proliferation and differentiation of satellite cells (64). The expression of IGF-1 has also been found in skeletal muscle, where it is released from myofibers upon stimuli that damage the cell membrane, such as muscle overload (65). IL-6 is the oldest known myokine molecule, and its physiological effects include systemic metabolic regulation (66). HGF is also released extracellularly upon muscle injury and activates satellite cells (67). HGF activates mTOR signaling (68). FGF-2 is another growth factor that is secreted upon cell membrane damage (69,70). FGF-2 plays a role in regulating cell proliferation and differentiation by activating the mitogen-activated protein kinase (MAPK) signaling pathway in many cells (71). In satellite cells, p38α/βMAPK is activated upon entry from quiescence into the cell cycle, which is triggered by FGF-2 (62). It has also been shown that the activation of the Erk1/2 pathway by FGF-2 is essential in proliferating myocytes between G1 and S phases of the cell

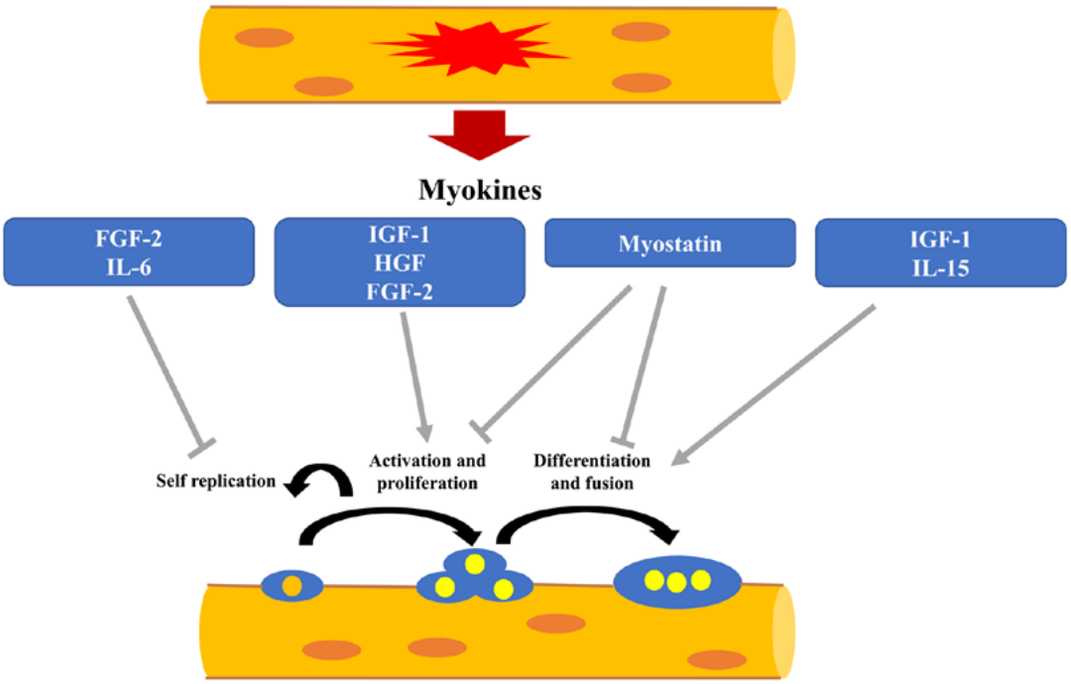

Figure 3. Role of myokines associated with the regulation for the functions of muscle satellite cells. FGF-2, fibroblast growth factor 2; IL, interleukin; IGF-1, insulin-like growth factor-1; HGF, hepatocyte growth factor.

cycle (72). IL-15 is a cytokine that is abundantly expressed in skeletal muscle and is recognized as a myokine that acts endocrinologically on adipose tissue and regulates whole body energy metabolism (73). On the other hand, IL-15 has anabolic effects and is considered to be involved in skeletal muscle hypertrophy (74). It has been shown that the muscle hypertrophic effect of IL-15 occurs in a pathway independent of IGF-1 (75). When cultured skeletal muscle cells are treated with IL-15, protein synthesis is increased and protein degradation is inhibited, resulting in hypertrophy of muscle fibers (75,76). While, it has been reported that the number of satellite cells decreases with aging, suggesting a link to reduced muscle regeneration capacity (77). This is attributed to the reduced self-replication capacity of satellite cells due to aging and the inability to secure the number of stem cells. On the other hand, myokines are also considered to play a part in the mechanism of inhibiting cancer growth by exercise, and one myokine that has actually been shown to inhibit cancer growth is secreted protein acidic and rich in cysteine (SPARC) (78).

Myostatin is a myokine that belongs to the TGF-family. In 1997, it was reported that skeletal muscle mass markedly increased in myostatin gene-knockout mice, which attracted attention as a factor regulating muscle mass (79). Myostatin binds to activin type IIB receptor and ALK4/ALK5 coreceptor, promotes phosphorylation of Smad2 and Smad3 proteins, and suppresses the expression of genes involved in skeletal muscle differentiation (80,81). Myostatin has also been reported to inhibit the PI3K/Akt signaling pathway (82). It has also been reported that myostatin secretion from muscle and adipocytes is increased in patients with severe obesity (83), and that weight loss decreases the expression of myostatin in muscle (84). Sarcopenic obesity can be associated with these observations. Follistatin and follistatin-related genes are known to be molecules that bind to and inhibit the function

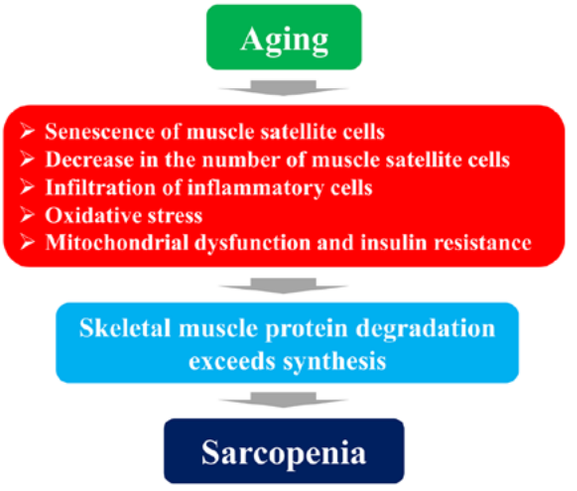

Figure 4. Schematic diagram of the pathogenesis of sarcopenia during the aging process.

of myostatin, and it is expected that these molecules can be used to increase muscle mass (85). During high-intensity exercise, myostatin is suppressed and muscle hypertrophy can occur through activation of the mTOR/IGF-1 system (86). The schematic explanation between myokines associated with the regulation for the functions of muscle satellite cells and the repair of damaged myofiber is illustrated in Fig. 3.

In recent years, it has also become clear that the myostatin gene is involved in the 'appropriateness for the running distance' of racing horses (87). There are three genetically distinct types of myostatin (C/C, C/T and T/T) in Thoroughbreds (88). It has been found that the difference of genetic types is associated with muscle mass and appropriateness for the running distance. In the C/C type muscle mass

tends to increase slightly, in the T/T type it tends to decrease slightly, and in the C/T type it tends to be in the middle (88). Therefore, racing horses with the C/C type tends to be suitable for a short distance, while those with the T/T type tends to be suitable for medium and long distances. Those with the C/T type tends to be suitable for a medium distance (88).

6. Renin-angiotensin system, sex hormones and sarcopenia

The renin-angiotensin system (RAS) is known from a report published in the Lancet in 2002, which demonstrated that continuous angiotensin-converting enzyme (ACE) inhibitor treatment suppressed knee extensor strength decline and walking speed decline (89). This report attracted attention to the suppression of the RAS. RAS activation is thought to cause sarcopenia through the following: i) Indirect effects, such as angiotensin II-induced decrease in anabolic hormones, induction of proinflammatory cytokines and increased muscle protein degradation via increased myostatin; and ii) direct oxidative stress via angiotensin II type 1 receptors (90,91). RAS suppression may contribute to the prevention of sarcopenia.

Age-related changes in reproductive endocrine organs are considered to be one of the most important functional changes associated with aging. In general, thyroid hormones and glucocorticoids maintain relatively constant levels in response to aging, whereas blood levels of sex steroid hormones, such as testosterone, are known to decrease with age in adults (92,93). The decline in blood testosterone levels with aging is considered to be associated with geriatric diseases and functional disabilities. In a cross-sectional study on men aged 24-90 years, serum testosterone levels were reported to be positively associated with skeletal muscle mass and muscle strength (94). In post-menopausal women, estrogen decline can cause endocrine and metabolic dysfunction, resulting in a predisposition to osteoporosis, metabolic syndrome and sarcopenia (95). Osteosarcopenia, a combined condition of osteoporosis and sarcopenia, increases the risk of developing frailty (96).

7. Conclusions

The present review outlined the pathogenesis of primary sarcopenia from the following viewpoints: i) Myofibers and muscle satellite cells; ii) protein synthesis and degradation; iii) immunocompetence and inflammation; iv) myokines; v) RAS; and vi) sex hormones. In the elderly, a lack of exercise, malnutrition and hormonal changes lead to neuromuscular junction insufficiency, an impaired capillary blood flow, a reduced repair and regeneration capacity due to the senescence of muscle satellite cells, a decrease in the number of muscle satellite cells, the infiltration of inflammatory cells and oxidative stress, resulting in muscle protein degradation exceeding synthesis. In addition, mitochondrial dysfunction causes metabolic abnormalities, such as insulin resistance, which may lead to quantitative and qualitative abnormalities in skeletal muscle, resulting in sarcopenia. A schematic diagram of the pathogenesis of sarcopenia during aging process is illustrated in Fig. 4. Skeletal muscle has been the subject of a great amount of research in recent years, and it is hoped that further drug discovery for sarcopenia based on pathological conditions will be developed in the future. The authors consider that the novelty of the present review article is that it outlines the pathogenesis of sarcopenia based on the latest evidence, with the aim of assisting in the development of novel drugs for sarcopenia.

Acknowledgements

Not applicable.

Funding

No funding was received.

Availability of data and materials

Not applicable.

Authors' contributions

HN wrote the review article. SF, AA, KY, SN and KH were involved in the editing and reviewing of the article. HN and KH confirm the authenticity of all the raw data. All authors have read and approved the final article.

Ethics approval and consent to participate

Not applicable.

Patient consent for publication

Not applicable.

Competing interests

The authors declare that they have no competing interests.

References

1. Rosenberg IH: Summary comments. Am J Clin Nutr 50: 1231-1233, 1989.
2. Lexell J, Taylor CC and Sjostrom M: What is the cause of the ageing atrophy? Total number, size and proportion of different fiber types studied in whole vastus lateralis muscle from 15- to 83-year-old men. J Neurol Sci 84: 275-294, 1988.
3. Kuzuya M: Aging-related frailty and sarcopenia. The concepts and diagnostic criteria of frailty. Clin Calcium 28: 1171-1176, 2018 (In Japanese).
4. Tournadre A, Vial G, Capel F, Soubrier M and Boirie Y: Sarcopenia. Joint Bone Spine 86: 309-314, 2019.
5. Ciciliot S, Rossi AC, Dyar KA, Blaauw B and Schiaffino S: Muscle type and fiber type specificity in muscle wasting. Int J Biochem Cell Biol 45: 2191-2199, 2013.
6. Nilwik R, Snijders T, Leenders M, Groen BB, van Kranenburg J, Verdijk LB and van Loon LJ: The decline in skeletal muscle mass with aging is mainly attributed to a reduction in type II muscle fiber size. Exp Gerontol 48: 492-498, 2013.
7. Deschenes MR: Effects of aging on muscle fibre type and size. Sports Med 34: 809-824, 2004.
8. Chen LK, Woo J, Assantachai P, Auyeung TW, Chou MY, Iijima K, Jang HC, Kang L, Kim M, Kim S, *et al*: Asian Working group for sarcopenia: 2019 consensus update on sarcopenia diagnosis and treatment. J Am Med Dir Assoc 21: 300-307.e2, 2020.
9. Nishikawa H, Shiraki M, Hiramatsu A, Moriya K, Hino K and Nishiguchi S: Japan Society of Hepatology guidelines for sarcopenia in liver disease (1st edition): Recommendation from the working group for creation of sarcopenia assessment criteria. Hepatol Res 46: 951-963, 2016.

10. Lim WS, Liang CK, Assantachai P, Auyeung TW, Kang L, Lee WJ, Lim JY, Sugimoto K, Akishita M, Chia SL, *et al*: COVID-19 and older people in Asia: Asian Working Group for Sarcopenia calls to actions. Geriatr Gerontol Int 20: 547-558, 2020.
11. Wang PY, Li Y and Wang Q: Sarcopenia: An underlying treatment target during the COVID-19 pandemic. Nutrition 84: 111104, 2021.
12. Shang M, Cappellesso F, Amorim R, Serneels J, Virga F, Eelen G, Carobbio S, Rincon MY, Maechler P, De Bock K, *et al*: Macrophage-derived glutamine boosts satellite cells and muscle regeneration. Nature 587: 626-631, 2020.
13. Chen X, Xiang L, Jia G, Liu G, Zhao H and Huang Z: Leucine regulates slow-twitch muscle fibers expression and mitochondrial function by Sirt1/AMPK signaling in porcine skeletal muscle satellite cells. Anim Sci J 90: 255-263, 2019.
14. Verdijk LB, Koopman R, Schaart G, Meijer K, Savelberg HH and van Loon LJ: Satellite cell content is specifically reduced in type II skeletal muscle fibers in the elderly. Am J Physiol Endocrinol Metab 292: E151-E157, 2007.
15. Fochi S, Giuriato G, De Simone T, Gomez-Lira M, Tamburin S, Del Piccolo L, Schena F, Venturelli M and Romanelli MG: Regulation of microRNAs in satellite cell renewal, muscle function, sarcopenia and the role of exercise. Int J Mol Sci 21: 6732, 2020.
16. Schiaffin S, Reggiani C and Murgia M: Fiber type diversity in skeletal muscle explored by mass spectrometry-based single fiber proteomics. Histol Histopathol 35: 239-246, 2020.
17. Wang Y and Pessin JE: Mechanisms for fiber-type specificity of skeletal muscle atrophy. Curr Opin Clin Nutr Metab Care 16: 243-250, 2013.
18. Lepore E, Casola I, Dobrowolny G and Musarò A: Neuromuscular Junction as an Entity of Nerve-Muscle Communication. Cells 8: 906, 2019.
19. Yamakawa H, Kusumoto D, Hashimoto H and Yuasa S: Stem cell aging in skeletal muscle regeneration and disease. Int J Mol Sci 21: 1830, 2020.
20. Liu L, Charville GW, Cheung TH, Yoo B, Santos PJ, Schroeder M and Rando TA: Impaired notch signaling leads to a decrease in p53 activity and mitotic catastrophe in aged muscle stem cells. Cell Stem Cell 23: 544-556.e4, 2018.
21. Brack AS, Conboy MJ, Roy S, Lee M, Kuo CJ, Keller C and Rando TA: Increased Wnt signaling during aging alters muscle stem cell fate and increases fibrosis. Science 317: 807-810, 2007.
22. Wilkinson DJ, Piasecki M and Atherton PJ: The age-related loss of skeletal muscle mass and function: Measurement and physiology of muscle fibre atrophy and muscle fibre loss in humans. Ageing Res Rev 47: 123-132, 2018.
23. Sartori R, Romanello V and Sandri M: Mechanisms of muscle atrophy and hypertrophy: Implications in health and disease. Nat Commun 12: 330, 2021.
24. Wilkinson DJ, Hossain T, Hill DS, Phillips BE, Crossland H, Williams J, Loughna P, Churchward-Venne TA, Breen L, Phillips SM, *et al*: Effects of leucine and its metabolite β-hydroxy-β-methylbutyrate on human skeletal muscle protein metabolism. J Physiol 591: 2911-2923, 2013.
25. Kruse R, Petersson SJ, Christensen LL, Kristensen JM, Sabaratnam R, Ørtenblad N, Andersen M and Højlund K: Effect of long-term testosterone therapy on molecular regulators of skeletal muscle mass and fibre-type distribution in aging men with subnormal testosterone. Metabolism 112: 154347, 2020.
26. de Alcantara Borba D, da Silva Alves E, Rosa JPP, Facundo LA, Costa CMA, Silva AC, Narciso FV, Silva A and de Mello MT: Can IGF-1 serum levels really be changed by acute physical exercise? A systematic review and meta-analysis. J Phys Act Health 17: 575-584, 2020.
27. Tan KT, Ang SJ and Tsai SY: Sarcopenia: Tilting the balance of protein homeostasis. Proteomics 20: e1800411, 2020.
28. Laplante M and Sabatini DM: mTOR signaling at a glance. J Cell Sci 122: 3589-3594, 2009.
29. D'Antona G and Nisoli E: mTOR signaling as a target of amino acid treatment of the age-related sarcopenia. Interdiscip. Top Gerontol 37: 115-141, 2010.
30. Giovannini S, Marzetti E, Borst SE and Leeuwenburgh C: Modulation of GH/IGF-1 axis: Potential strategies to counteract sarcopenia in older adults. Mech Ageing Dev 129: 593-601, 2008.
31. Akalu Y, Molla MD, Dessie G and Ayelign B: physiological effect of ghrelin on body systems. Int J Endocrinol 2020: 1385138, 2020.
32. Lu Y, Bradley JS, McCoski SR, Gonzalez JM, Ealy AD and Johnson SE: Reduced skeletal muscle fiber size following caloric restriction is associated with calpainmediated proteolysis and attenuation of IGF-1 signaling. Am J Physiol Regul Integr Comp Physiol 312: R806-R815, 2017.
33. Matheny RW Jr, Carrigan CT, Abdalla MN, Geddis AV, Leandry LA, Aguilar CA, Hobbs SS and Urso ML: RNA transcript expression of IGF-I/PI3K pathway components in regenerating skeletal muscle is sensitive to initial injury intensity. Growth Horm IGF Res 32: 14-21, 2017.
34. Bodine SC, Stitt TN, Gonzalez M, Kline WO, Stover GL, Bauerlein R, Zlotchenko E, Scrimgeour A, Lawrence JC, Glass DJ and Yancopoulos GD: Akt/mTOR pathway is a crucial regulator of skeletal muscle hypertrophy and can prevent muscle atrophy in vivo. Nat Cell Biol 3: 1014-1019, 2001.
35. Rommel C, Bodine SC, Clarke BA, Rossman R, Nunez L, Stitt TN, Yancopoulos GD and Glass DJ: Mediation of IGF-1-induced skeletal myotube hypertrophy by PI(3)K/Akt/mTOR and PI(3)K/Akt/GSK3 pathways. Nat Cell Biol 3: 1009-1013, 2001.
36. Parkington JD, LeBrasseur NK, Siebert AP and Fielding RA: Contraction-mediated mTOR, p70S6k, and ERK1/2 phosphorylation in aged skeletal muscle. J Appl Physiol 1985. 97: 243-248, 2004.
37. Gomes MD, Lecker SH, Jagoe RT, Navon A and Goldberg AL: Atrogin-1, a muscle-specific F-box protein highly expressed during muscle atrophy. Proc Natl Acad Sci USA 98: 14440-14445, 2001.
38. Bodine SC, Latres E, Baumhueter S, Lai VK, Nunez L, Clarke BA, Poueymirou WT, Panaro FJ, Na E, Dharmarajan K, *et al*: Identification of ubiquitin ligases required for skeletal muscle atrophy. Science 294: 1704-1708, 2001.
39. Giresi PG, Stevenson EJ, Theilhaber J, Koncarevic A, Parkington J, Fielding RA and Kandarian SC: Identification of a molecular signature of sarcopenia. Physiol Genomics 21: 253-263, 2005.
40. Clavel S, Coldefy AS, Kurkdjian E, Salles J, Margaritis I and Derijard B: Atrophy-related ubiquitin ligases, atrogin-1 and MuRF1 are up-regulated in aged rat Tibialis Anterior muscle. Mech Ageing Dev 127: 794-801, 2006.
41. Burd NA, Gorissen SH and van Loon LJ: Anabolic resistance of muscle protein synthesis with aging. Exerc Sport Sci Rev 41: 169-73, 2013.
42. Marshall RN, Smeuninx B, Morgan PT and Breen L: Nutritional strategies to offset disuse-induced skeletal muscle atrophy and anabolic resistance in older adults: From whole-foods to isolated ingredients. Nutrients 12: 1533, 2020.
43. Wilson D, Jackson T, Sapey E and Lord JM: Frailty and sarcopenia: The potential role of an aged immune system. Ageing Res Rev 36: 1-10, 2017.
44. Lee JS, Auyeung TW, Kwok T, Lau EM, Leung PC and Woo J: Associated factors and health impact of sarcopenia in older Chinese men and women: A cross-sectional study. Gerontology 53: 404-410, 2008.
45. Baylis D, Bartlett DB, Patel HP and Roberts HC: Understanding how we age: Insights into inflammaging. Longev Healthspan 2: 8, 2013.
46. Goodman MN: Tumor necrosis factor induces skeletal muscle protein breakdown in rats. Am J Physiol 260: E727-E730, 1991.
47. Goodman MN: Interleukin-6 induces skeletal muscle protein breakdown in rats. Proc Soc Exp Biol Med 205: 182-185, 1994.
48. Ko F, Abadir P, Marx R, Westbrook R, Cooke C, Yang H and Walston J: Impaired mitochondrial degradation by autophagy in the skeletal muscle of the aged female interleukin 10 null mouse. Exp Gerontol 73: 23-27, 2016.
49. Correia-Melo C, Marques FD, Anderson R, Hewitt G, Hewitt R, Col J, Carroll BM, Miwa S, Birch J, Merz A, *et al*: Mitochondria are required for pro-ageing features of the senescent phenotype. EMBO J 35: 724-742, 2016.
50. Sriram S, Subramanian S, Sathiakumar D, Venkatesh R, Salerno MS, McFarlane CD, Kambadur R and Sharma M: Modulation of reactive oxygen species in skeletal muscle by myostatin is mediated through NF-κB. Aging Cell 10: 931-948, 2011.
51. Tang H, Inoki K, Brooks SV, Okazawa H, Lee M, Wang J, Kim M, Kennedy CL, Macpherson PCD, Ji X, *et al*: mTORC1 underlies age-related muscle fiber damage and loss by inducing oxidative stress and catabolism. Aging Cell 18: e12943, 2019.
52. Bodine SC and Baehr LM: Skeletal muscle atrophy and the E3 ubiquitin ligases MuRF1 and MAFbx/atrogin-1. Am J Physiol Endocrinol Metab 307: E469-E484, 2014.

53. Povea-Cabello S, Oropesa-Ávila M, de la Cruz-Ojeda P, Villanueva-Paz M, de la Mata M, Suárez-Rivero JM, Álvarez-Córdoba M, Villalón-García I, Cotán D, Ybot-González P and Sánchez-Alcázar JA: Dynamic reorganization of the cytoskeleton during apoptosis: The two coffins hypothesis. Int J Mol Sci 18: pii: E2393, 2017.
54. Phillips T and Leeuwenburgh C: Muscle fiber specific apoptosis and TNF-alpha signaling in sarcopenia are attenuated by life-long calorie restriction. FASEB J 19: 668-670, 2005.
55. Dupont-Versteegden EE: Apoptosis in muscle atrophy: Relevance to sarcopenia. Exp Gerontol 40: 473-481, 2005.
56. Wu H and Ballantyne CM: Skeletal muscle inflammation and insulin resistance in obesity. J Clin Invest 127: 43-54, 2017.
57. Tornatore L, Thotakura AK, Bennett J, Moretti M and Franzoso G: The nuclear factor kappa B signaling pathway: Integrating metabolism with inflammation. Trends Cell Biol 22: 557-566, 2012.
58. Puthucheary ZA, Rawal J, McPhail M, Connolly B, Ratnayake G, Chan P, Hopkinson NS, Phadke R, Dew T, Sidhu PS, *et al*: Acute skeletal muscle wasting in critical illness. JAMA 310: 1591-600, 2013.
59. Merritt EK, Stec MJ, Thalacker-Mercer A, Windham ST, Cross JM, Shelley DP, Craig Tuggle S, Kosek DJ, Kim JS and Bamman MM: Heightened muscle inflammation susceptibility may impair regenerative capacity in aging humans. J Appl Physiol 115: 937-948, 2013.
60. Nishikawa H, Enomoto H, Nishiguchi S and Iijima H: Sarcopenic obesity in liver cirrhosis: Possible mechanism and clinical impact. Int J Mol Sci 22: 1917, 2021.
61. Pedersen BK and Febbraio MA: Muscles, exercise and obesity: Skeletal muscle as a secretory organ. Nat Rev Endocrinol 8: 457-465, 2012.
62. Giudice J and Taylor JM: Muscle as a paracrine and endocrine organ. Curr Opin Pharmacol 34: 49-55, 2017.
63. Pedersen BK, Akerström TC, Nielsen AR and Fischer CP: Role of myokines in exercise and metabolism. J Appl Physiol 103: 1093-1098, 2007.
64. Ten Broek RW, Grefte S and Von den Hoff JW: Regulatory factors and cell populations involved in skeletal muscle regeneration. J Cell Physiol 224: 7-16, 2010.
65. Adams GR: Invited Review: Autocrine/paracrine IGF-I and skeletal muscle adaptation. J Appl Physiol 93: 1159-1167, 2002.
66. Pedersen BK and Febbraio MA: Muscle as an endocrine organ: Focus on muscle-derived interleukin-6. Physiol Rev 88: 1379-406, 2008.
67. Tatsumi R, Anderson JE, Nevoret CJ, Halevy O and Allen RE: HGF/SF is present in normal adult skeletal muscle and is capable of activating satellite cells. Dev Biol 194: 114-128, 1998.
68. Rodgers JT, King KY, Brett JO, Cromie MJ, Charville GW, Maguire KK, Brunson C, Mastey N, Liu L, Tsai CR, *et al*: mTORC1 controls the adaptive transition of quiescent stem cells from G0 to G(Alert). Nature 509: 393-396, 2014.
69. Clarke MS and Feeback DL: Mechanical load induces sarcoplasmic wounding and FGF release in differentiated human skeletal muscle cultures. Faseb J 10: 502-509, 1996.
70. Yablonka-Reuveni Z, Seger R and Rivera AJ: Fibroblast growth factor promotes recruitment of skeletal muscle satellite cells in young and old rats. J Histochem Cytochem 47: 23-42, 1999.
71. Jones NC, Tyner KJ, Nibarger L, Stanley HM, Cornelison DD, Fedorov YV and Olwin BB: The p38alpha/beta MAPK functions as a molecular switch to activate the quiescent satellite cell. J Cell Biol 169: 105-116, 2005.
72. Jones NC, Fedorov YV, Rosenthal RS and Olwin BB: ERK1/2 is required for myoblast proliferation but is dispensable for muscle gene expression and cell fusion. J Cell Physiol 186: 104-115, 2001.
73. Quinn LS, Anderson BG, Strait-bodey L, Stroud AM and Argile M: Oversecretion of interleukin-15 from skeletal muscle reduces adiposity. Am J Physiol Endocrinal Metab 296: E191-E202, 2009.
74. Furmanczyk PS and Quinn LS: Interleukin-15 increases myosin accretion in human skeletal myogenic cultures. Cell Biol Int 27: 845-851, 2003.
75. Quinn LS, Anderson BG, Drivdahl RH, Alvarez B and Argilés JM: Overexpression of interleukin-15 induces skeletal muscle hypertrophy in vitro: Implications for treatment of muscle wasting disorders. Exp Cell Res 280: 55-63, 2002.
76. Quinn LS, Haugk KL and Damon SE: Interleukin-15 stimulates C2 skeletal myoblast differentiation. Biochem Biophys Res Commun 239: 6-10, 1997.
77. Shefer G, Rauner G, Yablonka-Reuveni Z and Benayahu D: Reduced satellite cell numbers and myogenic capacity in aging can be alleviated by endurance exercise. PLoS One 5: e13307, 2010.
78. Aoi W, Naito Y, Takagi T, Tanimura Y, Takanami Y, Kawai Y, Sakuma K, Hang LP, Mizushima K, Hirai Y, *et al*: A novel myokine, secreted protein acidic and rich in cysteine (SPARC), suppresses colon tumorigenesis via regular exercise. Gut 62: 882-889, 2013.
79. McPherron AC, Lawler AM and Lee SJ: Regulation of skeletal muscle mass in mice by a new TGF-beta super family member. Nature 387: 83-90, 1997.
80. Saneyasu T, Honda K and Kamisoyama H: Myostatin Increases Smad2 phosphorylation and atrogin-1 expression in chick embryonic myotubes. J Poult Sci 56: 224-230, 2019.
81. Nikooie R, Jafari-Sardoie S, Sheibani V and Nejadvaziri Chatroudi A: Resistance training-induced muscle hypertrophy is mediated by TGF-β1-Smad signaling pathway in male Wistar rats. J Cell Physiol 235: 5649-5665, 2020.
82. Liu L, Hu R, You H, Li J, Liu Y, Li Q, Wu X, Huang J, Cai X, Wang M and Wei L: Formononetin ameliorates muscle atrophy by regulating myostatin-mediated PI3K/Akt/FoxO3a pathway and satellite cell function in chronic kidney disease. J Cell Mol Med 25: 1493-1506, 2021.
83. Hill M, Wernig A and Goldspink G: Muscle satellite stem cell activation during local tissue injury and repair. J Anat 203: 89-99, 2003.
84. Milan G, Dalla Nora E, Pilon C, Pagano C, Granzotto M, Manco M, Mingrone G and Vettor R: Changes in muscle myostatin expression in obese subjects after weight loss. J Clin Endocrinol Metab 89: 2724-2727, 2004.
85. Mafi F, Biglari S, Ghardashi Afousi A and Gaeini AA: Improvement in skeletal muscle strength and plasma levels of follistatin and myostatin induced by an 8-week resistance training and epicatechin supplementation in sarcopenic older adults. J Aging Phys Act 27: 384-391, 2019.
86. Biglari S, Afousi AG, Mafi F and Shabkhiz F: High-intensity interval training-induced hypertrophy in gastrocnemius muscle via improved IGF-I/Akt/FoxO and myostatin/Smad signaling pathways in rats. Physiol Int: Jul 7, 2020 (Epub ahead of print).
87. Hill EW, McGivney BA, Rooney MF, Katz LM, Parnell A and MacHugh DE: The contribution of myostatin (MSTN) and additional modifying genetic loci to race distance aptitude in Thoroughbred horses racing in different geographic regions. Equine Vet J 51: 625-633, 2019.
88. McGivney BA, Browne JA, Fonseca RG, Katz LM, Machugh DE, Whiston R and Hill EW: MSTN genotypes in Thoroughbred horses influence skeletal muscle gene expression and racetrack performance. Anim Genet 43: 810-812, 2012.
89. Onde G, Penninx BW, Balkrishnan R, Fried LP, Chaves PH, Williamson J, Carter C, Di Bari M, Guralnik JM and Pahor M: Relation between use of angiotensin-converting enzyme inhibitors and muscle strength and physical function in older women: An observational study. Lancet 359: 926-930, 2002.
90. Mogi M: Effect of renin-angiotensin system on senescence. Geriatr Gerontol Int 20: 520-525, 2020.
91. Yoshida T, Tabony AM, Galvez S, Mitch WE, Higashi Y, Sukhanov S and Delafontaine P: Molecular mechanisms and signaling pathways of angiotensin II-induced muscle wasting: Potential therapeutic targets for cardiac cachexia. Int J Biochem Cell Biol 45: 2322-32, 2013.
92. van den Beld AW, Kaufman JM, Zillikens MC, Lamberts SWJ, Egan JM and van der Lely AJ: The physiology of endocrine systems with ageing. Lancet Diabetes Endocrinol 6: 647-658, 2018.
93. Roy TA, Blackman MR, Harman SM, Tobin JD, Schrager M and Metter EJ: Interrelationships of serum testosterone and free testosterone index with FFM and strength in aging men. Am J Physiol Endocrinol Metab 283: E284-E294, 2002.
94. Sipila S, Narici M, Kjaer M, Pollanen E, Atkinson RA, Hansen M and Kovanen V: Sex hormones and skeletal muscle weakness. Biogerontology 14: 231-245, 2013.
95. Ikeda K, Horie-Inoue K and Inoue S: Functions of estrogen and estrogen receptor signaling on skeletal muscle. J Steroid Biochem Mol Biol 191: 105375, 2019.
96. Yoshimura N, Muraki S, Iidaka T, Oka H, Horii C, Kawaguchi H, Akune T, Nakamura K and Tanaka S: Prevalence and co-existence of locomotive syndrome, sarcopenia, and frailty: The third survey of research on osteoarthritis/osteoporosis against disability (ROAD) study. J Bone Miner Metab 37: 1058-1066, 2019.