JGG 2017;65:271-283

Review

The interplay between fascia, skeletal muscle, nerves, adipose tissue, inflammation and mechanical stress in musculo-fascial regeneration

A. Zullo[1, 2], F.P. Mancini[1], R. Schleip[3], S. Wearing[4], L. Yahia[5], W. Klingler[3]

[1] Department of Sciences and Technologies, University of Sannio, Benevento, Italy; [2] CEINGE Advanced Biotechnologies, s.c.a r.l, Napoli, Italy; [3] Neurophysiological muscle laboratory, Department of Neurosurgery, Ulm University, Guenzburg, Germany; [4] School of Clinical Sciences & Institute of Health and Biomedical Innovation, Queensland University of Technology, Kelvin Grove, Australia; [5] Laboratory for Innovation and Analysis of Bio-Performance, École Polytechnique de Montréal, Succursale Centre-Ville Montréal, Quebec, Canada

Muscle and connective tissues are structures that provide stability and movement to organisms belonging to the animal kingdom. Fascia and muscle tissues structurally and functionally integrate to form a musculofascial system with exceptional biomechanical properties, which allow animals to accomplish complex tasks. Mechanical overload through intense exercise or injury, however, may damage these tissues over the course of a lifetime. Although mammalian skeletal muscle and fascia both retain a good regenerative potential in the adulthood, regeneration is very sensitive to alterations in the biochemical and physical environment. In this review, the reciprocal role of fascial tissue and skeletal muscle in their regeneration processes are explored. The involvement of adipose and nervous tissue in the regulation of muscle and fascia regeneration are also revised. It is hypothesised, for the first time, that for effective regeneration of skeletal muscle, both muscle and fascial tissues are necessary, and that nervous and adipose tissues contribute and deeply influence this process.

Key words: Skeletal muscle, Fascia, Musculo-fascial regeneration, Extracellular matrix, Muscle loading, Muscle innervation, Intramuscular adipose tissue

INTRODUCTION

Mammalian skeletal muscle has a complex structure composed by different elements, such as myofibres, connective tissue, blood vessels, nerves, adipose cells and immune cells. Trauma, such as strain injuries, which are very common in sport, damage the muscles. Moreover, chronic degenerative neuromuscular diseases, forced inactivity, and aging can significantly impair muscle structure, mass and functionality [1]. Damaged skeletal muscle can regenerate through the coordinated action of different cells and mechanisms, but this process is very complex and unfavorable factors can lead to ineffective muscle repair. Much has been already understood about skeletal muscle regeneration, but many of its aspects, such as the contribution of fascia, nerves, and adipose tissue still remain poorly investigated.

STRUCTURE OF FASCIAL TISSUE

Fascia is a fibrous connective tissue, which envelopes and infiltrates all the organs and structures of the human body, including muscles, vessels, nerves, and joints [2]. Although the gross anatomical structure of fascia varies throughout the human body, all fascial tissue is composed of cells and an extracellular matrix (ECM).

Received: August 24, 2017 - Accepted: December 03, 2017

Correspondence: Alberto Zullo, Department of Sciences and Technologies, University of Sannio, via Port'Arsa 11, 82100 Benevento, Italy - E-mail: albzullo@unisannio.it

FASCIAL CELLS

The main cell types within fascia include fibroblasts, myofibroblasts, fibrocytes, adipocytes, telocytes and migrating white blood cells [3-5]. Fibroblasts are mesenchyme-derived cells that produce ECM, release cytokines, and can be transformed, under the certain stimuli, into myofibroblasts [6]. Myofibroblasts express α smooth muscle actin (αSMA) and myosin and have contractile properties [7] Myofibroblasts, therefore, have the capacity to generate tension in the surrounding ECM, and in addition to secreting cytokines, can also migrate, and respond to chemokines [8]. Fibrocytes are monocyte-derived cells that have features of both macrophages and fibroblasts [7]. Fibrocytes migrate into the injured tissue, where they can differentiate into macrophages or into fibroblasts/myofibroblasts [7]. Adipocytes are connective-tissue cells that store and synthesize fat, and white blood cells belong to the immune system. Telocytes are interstitial cells with mechanotransduction features, which are involved in inter-cellular signaling, survival of satellite cells, angiogenesis, and maintenance of tissue homeostasis. These specific properties of telocytes, make them key elements for tissue regeneration [9 10].

FASCIA ASSOCIATED WITH SKELETAL MUSCLE TISSUE

Fascia and muscle tissue strictly cooperate to achieve the higher-order function of the whole human body. The fascia of the muscle tissue can be divided into a superficial fascia, a denser deep fascia, which includes muscular septi (like the lateral septum of the upper arm) and a muscle-specific fascial tissue, which includes three different layers: epimysium, perimysium, and endomysium [2].

Each of these muscle-specific fascial layers has a different molecular and structural composition [11]. Endomysium, which envelopes myofibers, contains a matrix layer and a particular structure, the basement membrane. The matrix layer is formed by collagen type I, FN, HA, chondroitin sulfate and dermatan sulfate proteoglycans [12 13]. The basement membrane is composed mainly of matrix proteins; laminins, type IV and VI collagen, nidogens and the HSPG perlecan [14]. These proteins are organized in highly specialized and complex networks that make contacts with integrins and dystroglycan on the surface of myofibers [13 15 16]. Perimysium, which separates muscle fascicles, is associated with a reticular network composed of collagen fibers, through which it controls the number of mitochondria in the subsarcolemmal space. Epimysium is a thick collagenous sheath that extends from the tendons and envelops many muscle fascicles. Endomisium is a thin layer of connective tissue that surrounds every single muscle fibre. Epimysium, perimysium, and endomysium are predominantly constituted by collagen type I, II and III, which is synthesized by fibroblasts [17]. Cells, via integrins, and extracellular- and basement-membrane proteins, therefore, participate in the formation of a complex 3D network within and surrounding muscle tissue [18].

Fascia acts as a mechanical bridge between different tissues. It transfers mechanical stress, which originates in a specific point of the body, to different areas and tissues. This force transmission allows these different body regions to sense the tensile stimulus and activate proprioceptors and mechanoreceptors [19]. Interstitial connective tissue contains and support blood vessels, nerves, and lymphatic tissue, and is responsible for the transfer of mechanical force from the muscle to the skeleton [20]. Nevertheless, fascia is not just a passive force transmitter. It also regulates mechanical stress by adsorbing, storing and releasing kinetic energy [2].

The main structural component of fascia, the ECM, reflects the biomechanical environment of muscle tissue, with a structural and a functional role that is based on its constituting proteins: laminin, entactin, fibrinogen, types I and VI collagen, and vitronectin [21 22]. The ECM of fascia not only provides mechanical support to the muscle, but is also fundamental for the transduction of mechanical tension to muscle cells and of muscle contraction to the surrounding tissues [11]. At the molecular level, this efficient mechanical communication between different tissues is achieved by means of the ECM, cellular membrane receptors linked to the ECM fibers, and cytoplasmic/cytoskeletal proteins linked to membrane receptors [11].

The ECM also governs cell orientation and organization [23] and enhances muscle regeneration by regulating muscle cell proliferation, migration, fragmentation and fusion [24]. Another important function of the ECM is the regulation of growth factor activity during muscle regeneration, through protein-protein interactions [18]. Pathologic changes in the ECM, such as fibrotic ECM, can reduce the regenerative potential of muscle fibers, independently of the myogenic potential itself [25]. The role of the ECM in maintaining tissue and cellular stability is also highlighted by the phenomenon called anoikis; a programmed cell death process (apoptosis) arising from the interruption of the interaction between ECM and adjacent cells [22].

There is a constant balance between the amounts of connective tissue and parenchyma in any tissue [26]. Healthy skeletal muscle has a high volume ratio of muscle cells to fibroblasts, with fibroblast nuclei comprising

8-15% of all nuclei in the tissue [27]. For normal tissue turnover, cells also release enzymes that degrade the ECM, such as cathepsins, heparanase, hyaluronidases, and metalloproteases (MMPs) [22]. Excessive accumulation of connective tissue, in the form of collagen fibers and myofibroblasts, leads to muscle fibrosis, and diseases, such as Dupuytren Disease [28-30].

EXTRACELLULAR MATRIX

The ECM fills the extracellular space both within and between each tissue, thus forming the interstitial connective tissue, where it acts as a structural scaffold and functional regulator [22]. The principal components of the ECM are structural proteins (collagens, laminins, fibronectin, vitronectin, tenascin-c, and elastin), specialized proteins (growth factors, small matricellular proteins and small integrin-binding glycoproteins (SIBLINGS)), glycosaminoglycans and proteoglycans, such as metalloproteinases and cytokines, and water [14]. Although the precise composition of the ECM varies between different tissues, collectively the proteins of the ECM confer adhesion, strength, and elasticity to fascial tissues [18]. Moreover, they also play an important role in inter-cellular signaling, due to their capacity to store and release growth factors, such as TGFs, and IGFs [14].

Structural proteins

Fibronectin (FN) binds to several ECM molecules, including collagen and tenascin, and promotes cell adhesion and migration [14]. Also, FN binding to integrins is a crucial step in the fibril assembly process [18]. Tenascin C regulates the activity of integrins and syndecan and has a fundamental role in muscle innervation and neural function [14]. Elastin forms polymeric elastic fibers that cross-link in an extensible 3D network, which moderate the extensibility of tissues when exposed to low-level force. Collagen is a collective term commonly used to refer to a family of structural proteins of the ECM that provide resistance to tension and link together different ECM proteins. The fibrous components of fascia mainly consist of types I, III, IV, V, VI, XI, XII, XIV, XXI collagen [4]. Vitronectin is an adhesion protein binding cells to the ECM. Laminins, on the other hand, bind to integrins and the dystrophin-glycoprotein complex (DGC) [15].

Specialized proteins

The ECM is linked to cells by means of adhesion proteins (e.g. integrin receptors, mechanosensitive ion channels and tyrosine kinases), localized on the cell membrane and acting as a bridge between ECM fibers and the intracellular cytoskeleton. Integrin receptors bind to laminins, collagens, fibronectin, tenascin and link ECM proteins to the intracellular environment, via interaction with actin. This organization enables a fast mechanical communication between the extracellular and the intracellular environments [15]. Upon receiving a mechanical input, cells can modify their morphology, 3D orientation, and activity. Therefore, tissue restructuring can be influenced by mechanical stress [26].

Glycosaminoglycans and Proteoglycans

Heparan sulfate proteoglycans (HSPGs) are components of the ECM and the basement membrane of skeletal muscle. HSPGs form multimeric fibrils and bind to many proteins in the ECM, such as FGF-2, thanks to their negative charges [14]. Glycosaminoglycans confer gel properties to the ECM and act as important storage system for growth factors and cytokines [21]. Hyaluronan (HA) is a linear polysaccharide whose main function is to provide lubrication, hydration, and resistance to compression [31]. It is also present as in an aggregated form, which increases the viscosity of ECM and reduces the gliding of connective and muscle tissue layers [32].

STRUCTURE OF SKELETAL MUSCLE TISSUE

In mammals, skeletal muscle can account for up to 40% of the total body mass, thus representing the most abundant tissue. It maintains posture and enables voluntary body movements under the control of the cortical areas of the central nervous system.

Skeletal muscle is defined as “striated” muscle because of its band-patterned appearance under microscopic examination [33]. Muscle fiber is the smallest functional unit of skeletal muscle. It is a long, multinucleated cylindrical cell composed of a large number of contractile elements, the myofibrils. Skeletal muscle is composed of hundreds to thousands of multinucleated muscle fibers. Many individual muscle fibers are bundled in fascicles by a layer of connective tissue, the endomysium. Fascicles are grouped, in turn, in the muscle by a sheet of connective tissue, the perimysium. Two more connective tissue layers, the epimysium, and the deep fascia, wrap the muscle and separate it from other muscles, respectively [34].

Muscle cells establish a connection to the ECM via dystrophin- and integrin-associated complexes [15 35 36]. These junctions allow the transmission of mechanical traction generated by muscle fibers to the ECM. Another important filament-like protein that contributes to the formation of the bridge between the extracellular matrix and the cytoskeleton of muscle cells is syncoilin [37]. This protein belongs to the dystrophin-associated protein complex and is localized to the sarcolemma of muscle fibers [37]. Studies on mice have demonstrated

that Syncoilin is fundamental for the lateral transmission of muscle tension to the extracellular environment [37]. Disease, forced inactivity, injury, malnutrition, and aging can significantly, and dangerously, deplete the mass and functionality of muscles [38]. The potential of natural or induced skeletal muscle repair is crucial to re-establish tissue integrity after muscular injury.

REGENERATION OF MUSCLE AND FASCIAL TISSUE

To preserve body integrity and function, both fascia and skeletal muscle tissues act synergistically to aid each other in regeneration. In fact, previous studies have shown that both satellite cells and resident fibroblasts are fundamental for effective regeneration of connective tissue and muscle (Fig. 1). In particular, fibroblasts can enhance satellite cell proliferation and self-renewal [39], and skeletal muscle cells participate in the rebuilding of connective tissue [14].

The ECM is a central player in muscle development and regeneration. It is a very dynamic structure that constantly adapts to the environment by continuously switching between degradation and re-assembly. It influences cell proliferation, survival, shape, migration, and differentiation. No wonder its alteration may lead to disease [18].

Metalloproteinases (MMPs) and A Disintegrin and Metalloprotease (ADAMs) are proteases involved in the degradation of ECM proteins. MMPs are produced intra-cellularly as inactive precursors and are activated by extracellular proteolytic activity. ADAMs, in contrast, are transmembrane proteins located on the membrane of muscle cell [14]. The balanced control of MMP activity is achieved by the secretion of tissue inhibitors of MMPs (TIMPs) that allow MMP action to be finely tuned [14]. Proteolytic degradation of the ECM is also a crucial step for the movement of endothelial cells during regenerative neovascularization [40]. ECM degradation, therefore, not only destroys the extracellular protein network but also generates bioactive peptides and releases growth factors trapped within the ECM [18].

Given the strong interconnection between ECM and skeletal muscle, it is not surprising that skeletal muscle influences the remodeling of the ECM. In fact, during skeletal muscle regeneration extracellular proteases (MMP-2, MMP-7, MMP-9, MMP-24, MMP-25, plasmin and the plasminogen activation system), secreted by muscle cells, inflammatory cells and other cells, participate in the degradation of the ECM [41]. MMPs can both degrade ECM proteins, such as collagen fibers, elastin, proteoglycans, and fibronectin as well as proteolytically activate other proteins, such as the MMPs themselves [42]. For example, plasmin can degrade fibronectin and laminin and also activate MMPs and several latent growth factors [43]. Moreover, there is some evidence that activated satellite cell influence the remodeling of ECM [14 25].

The formation of new muscle mass is a process involving muscle cells and non-muscle cells with myogenic potential, such as adipose-tissue derived stem cells, mesoangioblasts, pericytes, side-population cells, Ac133 + cells, stem and/or precursor cells from muscle endothelium, and synovium [44]. Nevertheless, skeletal muscle regeneration relies mainly on the activity of satellite cells, i.e. muscular stem cells (MuSCs) with asymmetric division and self-renewal capabilities [45]. These cells are small, have a single nucleus, and are located between the myofiber sarcolemma and the basal lamina, and are, therefore, in close contact with the ECM. The paired box 7 (Pax7) transcription factor and tetraspanin KAI/CD82 are the molecular markers that identify satellite cells [46 47]. Pax7+ satellite cells are capable of asymmetric division, thus contributing to the regeneration of muscle fibers and, at the same time, maintenance of the stem cell pool [48]. Furthermore, MuSCs express different molecular markers in the quiescent state and the proliferating/differentiating state [46]. Even though satellite cells constitute the vast majority of MuSCs, it has been observed that non-satellite cells like mesenchymal stem cells may also acquire the skeletal muscle features under appropriate conditions [49]. Upon skeletal muscle injury, satellite cells are activated and start proliferating to execute the dual program of self-renewing and differentiation into new muscular fibers [50]. The critical balance between post-replicative differentiation and stemness is regulated by the presence of myogenic and self-renewing subpopulations of satellite cells [51]. On the other hand, the satellite niche, which is the MuSCs microenvironment, provides specific signals driving satellite cells toward either differentiation or stemness fate [46]. These signals include cell-to-matrix and cell-to-cell interactions, autocrine and paracrine stimuli, as well as other biochemical factors.

Satellite cells live in a reversible 'G0' or quiescence state that is crucial for the maintenance of the pool of myogenic precursors. This state allows satellite cells to re-enter the cell cycle in response to specific stimuli, like muscular injury. Cell cycle entry and exit are controlled by the coordinated expression of an array of regulatory genes and factors. While genes inhibiting the cell cycle are highly expressed in quiescent satellite cells, those promoting the cell cycle are activated after muscular injury [52-55]. During muscle regeneration, proliferating satellite cells either exit the cell cycle to return to a quiescent state and restore the MuSC pool or, alternatively, differentiate into myoblasts and fuse with pre-existing

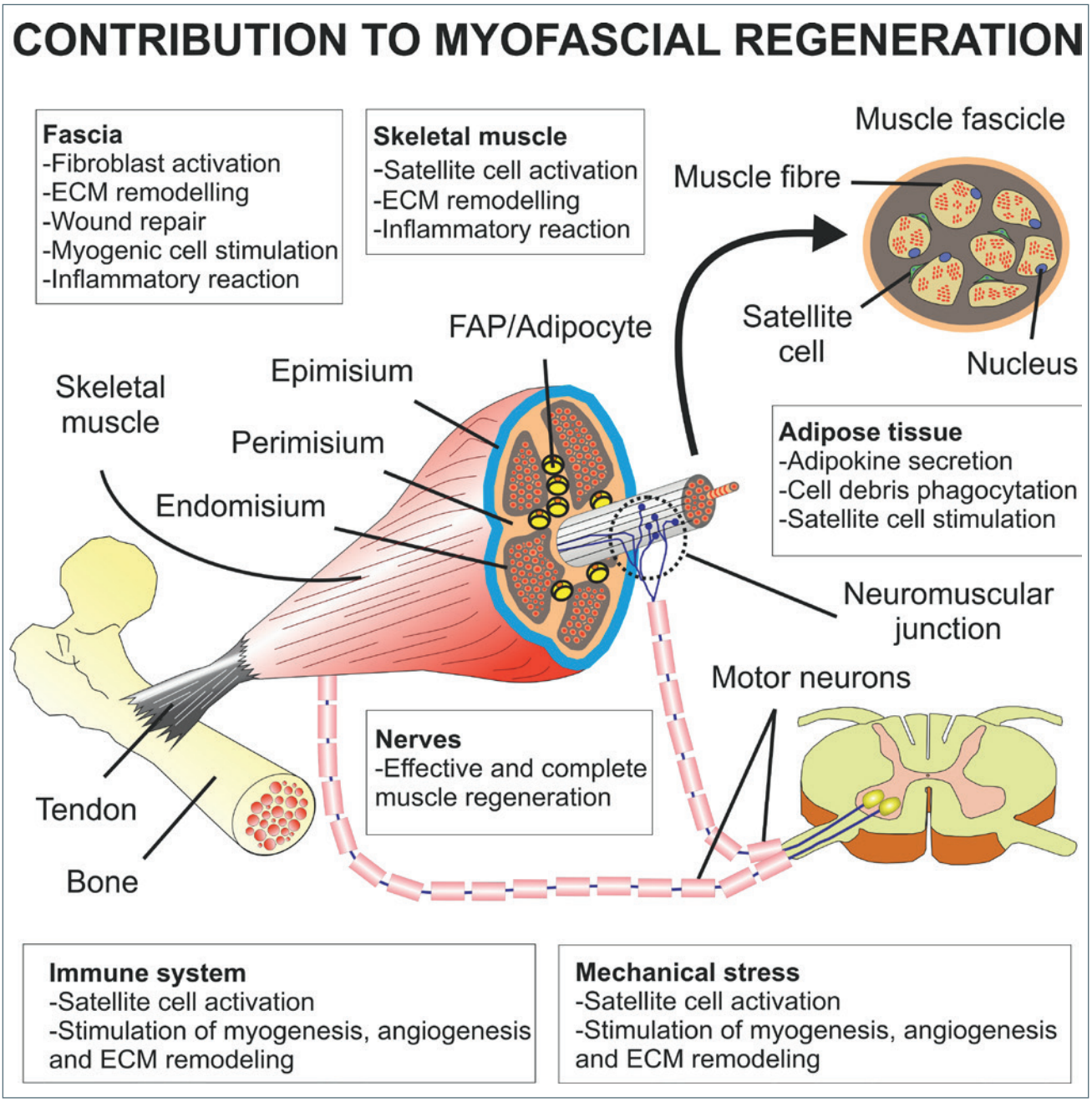

Figure 1. Scheme showing the contribution of skeletal muscle, fascia, nerves, intramuscular adipose tissue, immune system, and mechanical stress to myofascial regeneration.

muscle fibers to form new myofibers [56 57]. Collagen type VI, present in the ECM, and fibronectin, produced by activated satellite cells, induces the self-renewal of the satellite pool [58 59]. The post-mitotic cellular contacts within the niche also play a role in the determination of the satellite cell fate. For example, daughter cells in contact with the basement membrane are directed to the satellite cell self-renewal pathway, while daughter cells interacting with myofibers differentiate into myofibers [60].

ECM components also enhance muscle regeneration by regulating muscle cell proliferation, migration, fragmentation, and fusion [24]. Aberrant ECM remodeling after muscle injury, therefore, leads to ineffective muscle regeneration [61]. In the initial phase of skeletal muscle regeneration, both fibroblasts and satellite cells actively

proliferate, promoting each other's growth through a reciprocal positive feedback [39].

Cellular activities, as well as temporal and spatial production of ECM proteins, vary according to the different phases of the regeneration process [14]. Following tissue injury, fibroblasts migrate to the site of damage, proliferate and generate an extracellular network of disorganized collagen III fibers, most of which are subsequently transformed into collagen I fibers. Moreover, fibroblasts also secrete growth factors for the myogenic cells and differentiate into myofibroblasts, which contribute to the formation of new muscle fibers [39]. At this stage, the ECM collagen type I/III, which confer tensile strength, and laminin, tenascin-C, and fibronectin, which confer elastic properties, are produced, and TGF-ß1 is one important driver of such processes [62]. After this phase, a wound contracture process shortens the edges of the damaged area [63]. The shortening of these margins takes place because of the crosslinking and shortening of the collagen fibers. As result, a tight scar forms, thus increasing the mechanical resistance, and the functional performances of the damaged tissues. Afterward, collagen fibers aggregate and orientate to give better functional responses to mechanical stress, and to contractile activity [63]. This fibrotic response is beneficial in the initial phase of tissue regeneration because it preserves the tissue integrity. On the other hand, fibrotic elements should be removed, little by little, from the injured site, to replace the fibrotic tissue with functional skeletal muscle tissue [62].

During the regenerative phase, the regrown myofibers establish new and stronger lateral contacts (increasing the numbers of integrins on the adhesion sites) with the surrounding ECM, to allow some muscle activity and reduce the risk of a re-rupture [14]. The role of ECM in regenerative processes is so important that it has also been demonstrated in nonmammalian animal models, such as axolotl and *Octopus Vulgaris* [64 65].

ADIPOSE TISSUE

There is a close physical and biochemical link between connective tissue, muscle tissue, and adipose tissue. Therefore, they profoundly influence each other. In particular, skeletal muscle activity and regeneration are strongly affected by adipose tissue. Adipocytes are present in muscle tissue as "intramuscular fat", and secrete adipokines, proteic hormones that also regulate skeletal muscle development and function (Fig. 1).

Different cell types, including mesenchymal progenitors and fibro/adipogenic progenitors (FAPs), have been shown to participate in the development of fat cells within the muscle tissue, however, the potential of satellite cells to generate intramuscular adipocytes has only been suggested [66]. FAPs are multipotent mesenchymal progenitor cells that are present in many tissues, such as skeletal muscle. These cells are fundamental in the process of muscle regeneration. In fact, after muscle injury, they transiently proliferate, phagocytize necrotic debris, and stimulate satellite cell expansion. Importantly, after this initial phase, the number of FAP must rapidly decline to allow efficient muscle regeneration [67 68]. Under disuse or chronic muscle diseases, such as dystrophic conditions, FAPs proliferate, differentiate into adipocytes and ECM-producing fibroblasts and persist in the tissue; thereby resulting in the accumulation of intra-muscular adipose tissue (IMAT) and fibrotic tissue [69-71]. In particular, FAPs can be induced to differentiate into ECM-producing cells by macrophages [72]. It has been shown in mouse models, that unloading inhibits IMAT accumulation in regenerating muscle [73]. The accumulation of fat tissue in the muscle inhibits its regenerative potential because it suppresses the proliferative capacity of satellite cells and may also inhibit the activity of macrophages [73 74]. In fact, obese mice have a very low skeletal muscle regenerative potential [74]. Fatty infiltration in the muscle tissue has also been proposed to arise from the differentiation of FAP into adipocytes when muscle regeneration is impaired [67]. The crosstalk among MuSCs, FAPs, and immune cells determines the fate of the muscle tissue; either toward regeneration or fibro-adipogenic degeneration [75].

In vitro, adipose stem cells can influence the activity of myofibroblasts. In particular, this cell-cell interaction leads to inhibition of actin expression and contractility in smooth muscle [76]. In the rat, it has been demonstrated that adipose tissue-derived regenerative cells (ADRCs) can stimulate the repair of injured skeletal muscle tissue, through their paracrine activity [77]. In particular, these cells promote angiogenesis and myogenesis and prevent fibrosis. Moreover, adipose-derived stem cells (ASCs) have been shown in vitro to differentiate into skeletal muscle cells [78]. Experimental evidence has also demonstrated that human ADSC can be transdifferentiated into fibroblast-like cells *in vitro* [79]. These observations underline the close interconnection between adipose and connective tissue.

The interaction among different tissues also relies on soluble factors that are released from one tissue and target other tissues. Adipose tissue, once believed to be merely an energy depot, secretes an extensive repertoire of molecules, collectively called adipokines, which can affect surrounding cells or distant cells, thus acting in a paracrine or endocrine fashion. Skeletal muscle expresses receptors for several adipokines, including adiponectin and leptin [44]. Globular adiponectin (gAD) is also involved in the proliferation and myogenic

differentiation of satellite cells, myoblasts, and mesoangioblasts [44]. gAd is also fundamental for the migration of satellite cells and macrophages towards the muscle tissue. Moreover, gAD causes the degradation of the extracellular matrix through the induction of MMP-2 secretion by satellite muscle cells [44]. Adiponectin is also secreted in an autocrine fashion by skeletal muscle tissue, and the inflamed environment stimulates the muscle production of gAd [44]. In this context, the importance of adiponectin is further underlined by the evidence that decreased plasma adiponectin is associated with reduced regenerative potential of skeletal muscle [44].
Studies on mice have also demonstrated that leptin affects muscle tissue by increasing skeletal muscle mass and altering its miRNA expression [80]. Similarly, Il-6, an adipo-myokine that is secreted both by adipose and muscle tissue has been shown to stimulate satellite cell activation and muscle repair at the muscle level [81]. It has also been recently discovered that a novel adipokine, CTRP3, is involved in the proliferation and differentiation of myoblasts. CTRP3 is expressed in muscle cells and acts in an autocrine/paracrine fashion [82]. In addition, other adipokines have also been shown to regulate myogenesis during muscle regeneration [82].

HUMORAL FACTORS

Soluble factors participate in the signaling process between the immune system, fascial tissue, and muscle tissue. HGF, FGFs, TGFβs, MSTN, IGF, TNFα, PDGF, IL-6, LIF, VEGF, EGF, KGF, are secreted by different cell types, including fibroblasts, myofibroblasts, muscle cells and immune cells [21]. Other factors, such as TGF-ß1, are trapped in the ECM via specific binding proteins (decorin and biglycan) and can be released by mechanical triggers [18 21 83 84].
In adult skeletal muscle, TGF-ß1 hinders muscle regeneration and promotes muscle atrophy and fibrosis. In particular, TGF-ß1 inhibits satellite cell proliferation, myofiber fusion, the activation of muscle-specific genes and, after injury, induces myoblast to myofibroblast conversion [85-87]. Furthermore, neutralization of TGF-ß1 reduces scar tissue formation and improves muscle tissue healing [62].

IMMUNE CELLS AND INFLAMMATION

The regenerative process in skeletal muscle also requires the participation of non-satellite cells, such as perivascular and immune cells (Fig. 1) [88]. Inflammatory cytokines (IFN-g and TNF), released at the site of injury, induce the polarization of infiltrated monocytes to M1 macrophages (classically proinflammatory). These cells, in turn, release other inflammatory cytokines and IGF-1, which activate muscle satellite cells [89 90]. Later, changes in the biochemical environment (cytokines) at the site of injury, induce the polarization of M1 macrophages to M2 anti-inflammatory phenotype. M2 macrophages inhibit inflammation by secreting IL-4, IL-10, TGF-b1, and support myogenesis by secreting IGF-1 [90]. M2 macrophages also induce angiogenesis and promote extracellular matrix remodeling [90-92]. The role of monocytes in the regeneration process of skeletal muscle has been demonstrated in mouse models, where a reduction of monocytes in the bloodstream or skeletal muscle has been shown to impair the regenerative potential of muscle [93 94]. In a mouse model of muscular dystrophy, suppression of proinflammatory macrophage activity by IL-10 reduces the severity of the disease [89]. This switch from M1 inflammatory to M2 anti-inflammatory and pro-regenerative macrophages is critical for an effective regeneration of the skeletal muscle [94-96]. In the early phases following muscle injury, inflammation favors muscle regeneration by inhibiting satellite cell death and promoting myoblast proliferation [44]. In the later phases, in contrast, Treg cells enhance the regenerative potential of skeletal muscle by protecting injured tissue from inflammatory damage, promoting satellite cells differentiation (by amphiregulin), and inhibiting fibrosis [97 98]. During chronic inflammation, such as those occurring in muscular dystrophies, the ECM accumulates and replaces muscle tissue with fibrotic tissue [67]. During muscle regeneration, anti-inflammatory macrophages induce fusion, growth, and differentiation of muscle cells by TGF-ß, low levels of TNF-a and IGF-1 [99 100].
The dense network of the ECM acts as a physical barrier against the movements of cells. Therefore it must be weakened by fragmentation to allow cells to penetrate it. The binding of integrins with laminins and type IV collagen on the basal lamina prevents neutrophils from reaching the skeletal muscle [14]. In the area of damage, different cells, such as fibroblasts, macrophages, endothelial cells release several proteases that fragment the ECM [14]. The ECM also has a positive role for cell invasion into the injury site. Fibronectin, type IV collagen, laminin, entactin and proteolytic fragments of fibronectin and laminin, all act as guides and chemoattractants for immune cells and myoblasts [14 101].
Interestingly, in aged organisms, the mass and strength of skeletal muscle are reduced, and satellite cells have impaired regenerative potential. Although the myogenic commitment of satellite cells is not affected by aging, their capacity to self-renew is reduced [102]. Emerging evidence suggests that changes occurring in the satellite cell niche contribute to the reduced regenerative potential of aged muscle.

MECHANICAL STRESS

Mechanical stress has a reciprocal influence on muscle and ECM. The ECM influences muscle cell alignment and activity, and muscle activity influences collagen fiber alignment [26]. It is well known that physical exercise is healthy for the organism, but only recently has it been appreciated that mechanical loading of skeletal muscle post-injury also triggers many beneficial responses that enhance skeletal muscle regeneration and reduce fibrosis (Fig. 1) [103-107]. Skeletal muscle loading can directly increase skeletal muscle regeneration by stimulating the activation of SCs, attracting exogenous myogenic stem cells and stimulating angiogenesis [106 107]. Moreover, mechanical loading of newly formed muscle fibers is crucial for maturation of the tissue and of the myotendinous junctions. Interestingly, mechanical stimulation, in the form of stretch activation, massage therapy, and physical manipulation can improve healing of muscle tissue after damage, exerting anti-inflammatory and anti-fibrotic effects [108-110].

The formation of scar tissue, secondary to an injury or pathogenic process, modifies the biomechanical properties of the connective tissue. It also leads to a reduction in gliding between adjacent tissues [63]. Several studies have shown that physical exercise in Duchenne muscle dystrophy and skeletal muscle-injured mice can prevent fibrosis and enhance muscle performance [111-114]. Parenthetically, removal of stress, via mechanical unloading, has been shown to reduce muscle regeneration after injury [115-117]. After this initial resting phase, however, the absence of mechanical loading reduces the extent of muscle regeneration [73]. As demonstrated in ovariectomized mice, mechanical stimulation of muscle fibers can inhibit fat accumulation in skeletal muscle tissue, a side effect of ovariectomy [66]. Mechanical stimulation has also been proved to prevent satellite cells differentiation toward an adipogenic fate [66].

The contractile activity of muscle cells stimulates the production of myokines, such as IL-6 [107]. Transient and short-term exercise-induced production of IL-6 is beneficial for muscle metabolism, however, long-term IL-6 production is detrimental and is associated with muscle atrophy [118].

Myokines are proteins secreted from skeletal muscle cells that exert autocrine and endocrine effects and allow communication with adipose tissue, liver, and pancreatic cells [107].

Mechanical stress is known to influence the structure the ECM. Deposition and alignment of the collagen fibers secreted into the extracellular space (to form the ECM matrix) is driven by the position and the orientation of the connective fibroblast-like cells [26]. At molecular level, the alignment of intracellular actin fibers in collagen-producing cells guides the parallel deposition of extracellular collagen [26]. Therefore, the stress produced by cytoskeletal proteins of cells can influence the structure of ECM [18].

Mechanical load also influences fiber alignment by acting on ECM protein synthesis and degradation [26]. Strain-stabilization, a mechanism relating mechanical stress and collagen fiber orientation, involves mechanical activation of enzymatic degradation of the collagen fibers [26]. ECM fiber orientation, in turn, affects cell activity, differentiation, and migration [119 120]. Mechanical stress, in the form of traction, also stimulates the regeneration of deep fascia in animals subjected to leg lengthening [28 121]. Shear stress applied to the tissue can also modify the alignment of collagen fibers [26].

The sensitivity of tissues to mechanical stress is dependent on the presence of TGF-ß1 in traction-sensitive protein complexes that can activate/deactivate TGF-ß1 in response to contractile force changes [83]. These mechanical stresses can originate in the extracellular environment (injury) or by cell contraction (myofibroblasts or muscle cells) [83]. The mechanical communication between extracellular and intracellular environments is due to specific cell-ECM and cell-cell interactions mediated by protein complexes [122]. Among these, integrins function as mechanotransducers that can transmit tension in the ECM to the cell and activate cellular responses [18]. Cells can sense changes in ECM stiffness and mechanical stress by cytoskeletal actomyosin contraction via integrin [122]. Proteins of the ECM interact with cells by integrins, assembled with other proteins in supramolecular adhesion complexes. The extracellular mechanical stress is transduced into intracellular events, by specific integrin-actin cytoskeleton interactions. Mechanical stimuli can alter the membrane assembly of these adhesion complexes and activate specific signaling processes, such as RhoA-induced actin stress fiber formation [123-127].

The input signals for the remodeling of the ECM are transmitted by ECM receptors, such as integrins, laminin receptors, syndecans, and proteases, including MMPs, serine protease (e.g. plasmin, plasminogen activator and uPAR) and cysteine protease (e.g.cathepsins) families and by structural tension provided by cells and extracellular proteins [18].

In the integer tissue, there is always a tensional homeostasis between cell traction and ECM stiffness [122]. There are distinct intracellular protein complexes, such as SPARC-ILK and Rho-A, that can also provide the tension required for FN assembly at the cell surface [18]. Moreover, FN can also induce cell contraction [18].

NERVOUS TISSUE

Reinnervation of injured skeletal muscle is crucial for effective and complete regeneration of the tissue (Fig. 1) [14]. In particular, the formation of neuromuscular junctions (NMJs) drives gene expression in the regenerating myotubes [128 129]. The complex ECM network acts as tissue stabilizer and guides the formation of new NMJs [130]. Therefore, ECM is essential for reinnervation of injured skeletal muscle [18]. Somewhat surprisingly, however, the presence of nerves does not appear to be critical in the initial phases of muscle regeneration [14]. After muscle injury, the regeneration process restores muscle tissue, ECM, nerves, and their connections. Due to specific proteins, such as agrin and s-laminin and components of the basal lamina (BL), skeletal muscle retains the memory of the original NMJ sites and can guide the formation and differentiation of new NMJs at same location as in pre-injured muscle [14 131]. Integrity of the BL surrounding muscle fibers and motor axons, however, is crucial for successful re-innervation and regeneration of muscle tissue [14]. The BL surrounding the nerve can act as a scaffold and a guide axon regeneration toward the original NMJ site. When the continuity of the BL is disrupted, re-innervation of the regenerating muscle is less efficient [14]. Nevertheless, even in the situation of BL disruption, NMJs can readily form at the original NMJ sites when contacted by a growing axon [14].

CONCLUSIONS

Regeneration is the evolutionary key to preserve tissue function during aging and after injury. In humans, different tissues and organs have different regenerative potentials. However, tissues with substantial regenerative capacity can fail to regenerate because of disease or antagonistic physical and biochemical factors. Skeletal muscle and fascial tissues have a remarkable ability to regenerate. Nevertheless, their regeneration is strongly dependent on their close interaction and the presence of nervous and adipose tissues. Moreover, the regeneration process can be easily hindered in favor of non-functional fibrotic tissue accumulation. This, in turn, results in loss of the original mechanical and biological function of the injured tissue, increasing the susceptibility to re-injury, and the development of chronic pain [110 132]. ECM and fibroblasts activity play a fundamental role in the reaction of skeletal muscle to injury [133]. Regenerative medicine aims to improve recovery and regeneration of damaged tissues. Over the last decade, cell therapy and bio-engineered synthetic scaffolds have been employed to enhance tissue repair [18 22 133-136]. These approaches aim to replace damaged tissue with new functional tissue by supplying ECM-like structures with active cells [18].

Although the results of regenerative medicine trials are still not sufficiently encouraging, a deeper comprehension of the complex mechanisms underlying tissue regeneration can significantly improve the outcome of future clinical studies.

References

1 Ciciliot S, Schiaffino S. *Regeneration of mammalian skeletal muscle: basic mechanisms and clinical implications*. Curr Pharm Des 2010;16:906-14.

2 Klingler W, Velders M, Hoppe K, et al. *Clinical relevance of fascial tissue and dysfunctions.* Curr Pain Headache Rep 2014;18:439.

3 Henry G, Garner WL. *Inflammatory mediators in wound healing.* Surg Clin North Am 2003;83:483-507.

4 Kumka M, Bonar J. *Fascia: a morphological description and classification system based on a literature review*. J Can Chiropr Assoc 2012;56:179-91.

5 Bucala R, Spiegel LA, Chesney J, et al. *Circulating fibrocytes define a new leukocyte subpopulation that mediates tissue repair.* Mol Med 1994;1:71-81.

6 Homer RJ, Elias JA, Lee CG, et al. *Modern concepts on the role of inflammation in pulmonary fibrosis*. Arch Pathol Lab Med 2011;135:780-8.

7 Reilkoff RA, Bucala R, Herzog EL. *Fibrocytes: emerging effector cells in chronic inflammation.* Nat Rev Immunol 2011;11:427-35.

8 Baum J, Duffy HS. *Fibroblasts and myofibroblasts: what are we talking about?* J Cardiovasc Pharmacol 2011;57:376-9.

9 Chaitow L. *Telocytes: connective tissue repair and communication cells.* J Body Mov Ther 2017;21:231-3.

10 Bei Y, Wang F, Yang C, et al. *Telocytes in regenerative medicine.* J Cell Mol Med 2015;19:1441-54.

11 Zhang C, Gao Y. *Finite element analysis of mechanics of lateral transmission of force in single muscle fiber.* J Biomech 2012;45.

12 Okita M, Yoshimura T, Nakano J, et al. *Effects of reduced joint mobility on sarcomere length, collagen fibril arrangement in the endomysium, and hyaluronan in rat soleus muscle.* J Muscle Res Cell Motil 2004;25:159-66.

13 Grounds M. *Complexity of extracellular matrix and skeletal muscle regeneration.* In: Schiaffino S, Partrige T (Eds.). *Skeletal muscle repair and regeneration*. The Netherlands: Springer 2008, pp. 269-302.

14 Schiaffino S, Partrige T (Eds.). *Skeletal muscle repair and regeneration*. The Netherlands: Springer 2008.

15 Grounds MD, Sorokin L, White J. *Strength at the extracellular matrix-muscle interface.* Scand J Med Sci Sports 2005;15381-91.

16 Sanes JR. *The basement membrane/basal lamina of skeletal muscle.* J Biol Chem 2003;278:12601-4.

[17] Shadrin IY, Khodabukus A, Bursac N. *Striated muscle function, regeneration, and repair.* Cell Mol Life Sci 2016;73:4175-202.

[18] Daley WP, Peters SB, Larsen M. *Extracellular matrix dynamics in development and regenerative medicine*. J Cell Sci 2008;121:255-64.

[19] Stecco C, Gagey O, Belloni A, et al. *Anatomy of the deep fascia of the upper limb. Second part: study of innervation*. Morphologie 2007;91:38-43.

[20] Kjaer M. *Role of extracellular matrix in adaptation of tendon and skeletal muscle to mechanical loading.* Physiol Rev 2004;84:649-98.

[21] Badylak SF. *Regenerative medicine and developmental biology: the role of the extracellular matrix*. Anat Rec New Anat 2005;287:36-41.

[22] Fuoco C, Petrilli LL, Cannata S, et al. *Matrix scaffolding for stem cell guidance toward skeletal muscle tissue engineering.* J Orthop Surg Res 2016;11:86.

[23] Chaturvedi V, Dye DE, Kinnear BF, et al. *Interactions between skeletal muscle myoblasts and their extracellular matrix revealed by a serum free culture system.* PLoS One 2015;10:1-27.

[24] Calve S, Odelberg SJ, Simon HG. *A transitional extracellular matrix instructs cell behavior during muscle regeneration.* Dev Biol 2010;344:259-71.

[25] Grounds MD. *The need to more precisely define aspects of skeletal muscle regeneration*. Int J Biochem Cell Biol 2014;56:56-65.

[26] Ghazanfari S, Khademhosseini A, Smit TH. *Mechanisms of lamellar collagen formation in connective tissues.* Biomaterials 2016;97:74-84.

[27] Shadrin Y, Khodabukus A, Bursac N. *Striated muscle function, regeneration, and repair.* Cell Mol Life Sci 2016;73:4175-202.

[28] Karkampouna S, Kreulen M, Obdeijn MC, et al. *Connective tissue degeneration: mechanisms of palmar fascia degeneration (Dupuytren's disease)*. Curr Mol Biol Reports 2016;2:133-40.

[29] Serrano AL, Muñoz-Cánoves P. *Regulation and dysregulation of fibrosis in skeletal muscle*. Exp Cell Res 2010;316:3050-8.

[30] Grimaldi V, De Pascale MR, Zullo A, et al. *Evidence of epigenetic tags in cardiac fibrosis.* J Cardiol 2017;69:401-8.

[31] McNary SM, Athanasiou KA, Reddi AH. *Engineering lubrication in articular cartilage.* Tissue Eng Part B Rev 2012;18:88-100.

[32] Cowman MK, Schmidt TA, Raghavan P, et al. *Viscoelastic properties of hyaluronan in physiological conditions*. F1000Research 2015;4:622.

[33] Hill JA, Olson EN, Griendling KK. *Muscle: fundamental biology and mechanisms of disease.* 1st edition. Academic Press 2012.

[34] Gillies AR, Lieber RL. *Structure and function of the skeletal muscle extracellular matrix*. Muscle Nerve 2011;44:318-31.

[35] Ervasti JM. *Dystrophin, its interactions with other proteins, and implications for muscular dystrophy*. Biochim Biophys Acta - Mol Basis Dis 2007;1772:108-17.

[36] Kääriäinen M, Kääriäinen J, Järvinen TL, et al. *Integrin and dystrophin associated adhesion protein complexes during regeneration of shearing-type muscle injury*. Neuromuscul Disord 2000;10:121-32.

[37] Zhang J, Bang M-L, Gokhin DS, et al. *Syncoilin is required for generating maximum isometric stress in skeletal muscle but dispensable for muscle cytoarchitecture*. Am J Physiol Cell Physiol 2008;294:C1175-82.

[38] Evans WJ. *Skeletal muscle loss: cachexia, sarcopenia, and inactivity*. Am J Clin Nutr 2010;91:1123S-7S.

[39] Murphy MM, Lawson JA, Mathew SJ, et al. *Satellite cells, connective tissue fibroblasts and their interactions are crucial for muscle regeneration.* Development 2011;138:3625-37.

[40] Carmeli E, Moas M, Reznick AZ, et al. *Matrix metalloproteinases and skeletal muscle: a brief review.* Muscle Nerve 2004;29:191-7.

[41] Nagamine Y, Muñoz-Cánoves P, Medcalf RL. *Transcriptional and posttranscriptional regulation of the plasminogen activator system.* Thromb Haemost 2005;93:661-75.

[42] Kherif S, Lafuma C, Dehaupas M, et al. *Expression of matrix metalloproteinases 2 and 9 in regenerating skeletal muscle: a study in experimentally injured and mdx muscles.* Dev Biol 1999;205:158-70.

[43] Suelves M, Vidal B, Ruiz V, et al. *The plasminogen activation system in skeletal muscle regeneration: antagonistic roles of urokinase-type plasminogen activator (uPA) and its inhibitor (PAI-1).* Front Biosci 2005;10:2978-85.

[44] Fiaschi T, Magherini F, Gamberi T, et al. *Adiponectin as a tissue regenerating hormone: more than a metabolic function.* Cell Mol Life Sci 2014;71:1917-25.

[45] Brack AS, Rando T. *Tissue-specific stem cells: lessons from the skeletal muscle satellite cell*. Cell Stem Cell 2012;10:504-14.

[46] Dumont NA, Wang YX, Rudnicki MA. *Intrinsic and extrinsic mechanisms regulating satellite cell function*. Development 2015;142:1572-81.

[47] Alexander MS, Rozkalne A, Colletta A, et al. *CD82 is a marker for prospective isolation of human muscle satellite cells and is linked to muscular dystrophies.* Cell Stem Cell 2016;19:800-7.

[48] Abou-Khalil R, Brack AS. *Muscle stem cells and reversible quiescence: the role of sprouty*. Cell Cycle 2010;9:2575-80.

[49] Fujimaki S, Machida M, Hidaka R, et al. *Intrinsic ability of adult stem cell in skeletal muscle: an effective and replenishable resource to the establishment of pluripotent stem cells.* Stem Cells Int 2013;2013:420164.

[50] Shi X, Garry DJ. *Muscle stem cells in development, regeneration, and disease*. Genes Dev 2006;20:1692-708.

[51] Rocheteau P, Gayraud-Morel B, Siegl-Cachedenier I, et al. *A subpopulation of adult skeletal muscle stem cells retains all template DNA strands after cell division*. Cell 2012;148:112-25.

[52] Rodgers JT, King KY, Brett JO, et al. *mTORC1 controls the adaptive transition of quiescent stem cells from G0 to G(Alert).* Nature 2014;510:393-6.

[53] Machida S, Spangenburg EE, Booth FW. *Forkhead transcription factor FoxO1 transduces insulin-like growth factor's signal to p27Kip1 in primary skeletal muscle satellite cells.* J Cell Physiol 2003;196:523-31.

[54] Yablonka-Reuveni Z, Seger R, Rivera AJ. *Fibroblast growth factor promotes recruitment of skeletal muscle satellite cells in young and old rats.* J Histochem Cytochem 1999;47:23-42.

[55] Fukada S, Uezumi A, Ikemoto M, et al. *Molecular signature of quiescent satellite cells in adult skeletal muscle.* Stem Cells 2007;25:2448-59.

[56] Chakkalakal J V, Christensen J, Xiang W, et al. *Early forming label-retaining muscle stem cells require p27kip1 for maintenance of the primitive state*. Development 2014 15;141:1649-59.

[57] Pajcini K V, Corbel SY, Sage J, et al. *Transient inactivation of Rb and ARF yields regenerative cells from postmitotic mammalian muscle.* Cell Stem Cell 2010;7:198-213.

[58] Urciuolo A, Quarta M, Morbidoni V, et al. *Collagen VI regulates satellite cell self-renewal and muscle regenerati*on. Nat Commun 2013;4:1964.

[59] Bentzinger CF, Wang YX, von Maltzahn J, et al. *Fibronectin regulates Wnt7a signaling and satellite cell expansion.* Cell Stem Cell 2013;12:75-87.

[60] Kuang S, Kuroda K, Le Grand F, et al. *Asymmetric self-renewal and commitment of satellite stem cells in muscle*. Cell 2007;129:999-1010.

[61] Vinarsky V, Atkinson DL, Stevenson TJ, et al. *Normal newt limb regeneration requires matrix metalloproteinase function.* Dev Biol 2005;279:86-98.

[62] Laumonier T, Menetrey J. *Muscle injuries and strategies for improving their repair.* J Exp Orthop 2016;3:15.

[63] Brown MT, Tyler WB, Brown CH. IAAF *Medical Manual 2012*.

[64] Phan AQ, Lee J, Oei M, et al. *Positional information in axolotl and mouse limb extracellular matrix is mediated via heparan sulfate and fibroblast growth factor during limb regeneration in the axolotl (Ambystoma mexicanum)*. Regen 2015;2:182-201.

[65] Nödl M-T, Fossati SM, Domingues P, et al. *The making of an octopus arm.* Evodevo 2015;6:19.

[66] Frechette DM, Krishnamoorthy D, Adler BJ, et al. *Diminished satellite cells and elevated adipogenic gene expression in muscle as caused by ovariectomy are averted by low-magnitude mechanical signals.* J Appl Physiol 2015;119:27-36.

[67] Mann CJ, Perdiguero E, Kharraz Y, et al. *Aberrant repair and fibrosis development in skeletal muscle.* Skelet Muscle 2011;1:21.

[68] Joe AWB, Yi L, Natarajan A, et al. *Muscle injury activates resident fibro/adipogenic progenitors that facilitate myogenesis.* Nat Cell Biol 2010;12:153-63.

[69] Uezumi A, Ito T, Morikawa D, et al. *Fibrosis and adipogenesis originate from a common mesenchymal progenitor in skeletal muscle.* J Cell Sci 2011;124:3654-64.

[70] Uezumi A, Fukada S, Yamamoto N, et al. *Mesenchymal progenitors distinct from satellite cells contribute to ectopic fat cell formation in skeletal muscle*. Nat Cell Biol 2010;12:143-52.

[71] Lemos DR, Paylor B, Chang C, et al. *Functionally convergent white adipogenic progenitors of different lineages participate in a diffused system supporting tissue regeneration*. Stem Cells 2012;30:1152-62.

[72] Chazaud B. *Inflammation during skeletal muscle regeneration and tissue remodeling: application to exercise-induced muscle damage management.* Immunol Cell Biol 2016;94:140-5.

[73] Pagano AF, Demangel R, Brioche T, et al. *Muscle regeneration with intermuscular adipose tissue (IMAT) Accumulation Is Modulated by Mechanical Constraints.* PLoS One 2015;10:e0144230.

[74] Akhmedov D, Berdeaux R. *The effects of obesity on skeletal muscle regeneration.* Front Physiol 2013;4:371

[75] Farup J, Madaro L, Puri PL, et al. *Interactions between muscle stem cells, mesenchymal-derived cells and immune cells in muscle homeostasis, regeneration and disease*. Cell Death Dis 2015;6:e1830.

[76] Verhoekx JSN, Mudera V, Walbeehm ET, et al. *Adipose-derived stem cells inhibit the contractile myofibroblast in Dupuytren's disease.* Plast Reconstr Surg 2013;132:1139-48.

[77] Mori R, Kamei N, Okawa S, et al. *Promotion of skeletal muscle repair in a rat skeletal muscle injury model by local injection of human adipose tissue-derived regenerative cells.* J Tissue Eng Regen Med 2015;9:1150-60.

[78] Zuk P. *Adipose-derived stem cells in tissue regeneration: a review*. Int Sch Res Not 2013;2013:e713959.

[79] Hung M, Wen M, Huang Y. *Fascia tissue engineering with human adipose-derived stem cells in a murine model: Implications for pelvic floor reconstruction*. J Formos Med Assoc 2014;113:704-15.

[80] Hamrick MW, Herberg S, Arounleut P, et al. *The adipokine leptin increases skeletal muscle mass and significantly alters skeletal muscle miRNA expression profile in aged mice*. Biochem Biophys Res Commun 2010;400:379-83.

[81] Lutosławska G. Interleukin-6 *As an adipokine and myokine: the regulatory role of cytokine in adipose tissue and skeletal muscle metabolism.* Hum Mov Versita 2012;13:372-9.

[82] Otani M, Furukawa S, Wakisaka S, et al. *A novel adipokine C1q/TNF-related protein 3 is expressed in developing skeletal muscle and controls myoblast proliferation and differentiation*. Mol Cell Biochem 2015;409:271-82.

[83] Hinz B. *The extracellular matrix and transforming growth factor-β1: tale of a strained relationship.* Matrix Biology 2015;47:54-65.

[84] Fiaschi T, Cirelli D, Comito G, et al. *Globular adiponectin induces differentiation and fusion of skeletal muscle cells*. Cell Res 2009;19:584-97.

[85] Kwon A-H, Qiu Z, Hirao Y. *Topical application of plasma fibronectin in full-thickness skin wound healing in rats*. Exp Biol Med 2007;232:935-41.

[86] Burks TN, Andres-Mateos E, Marx R, et al. *Losartan restores skeletal muscle remodeling and protects against disuse atrophy in sarcopenia*. Sci Transl Med 2011;3:82ra37.

[87] Mendias CL, Gumucio JP, Davis ME, et al. *Transforming growth factor-beta induces skeletal muscle atrophy and fibrosis through the induction of atrogin-1 and scleraxis*. Muscle Nerve 2012;45:55-9.

[88] Domingues-Faria C, Vasson M, Goncalves-Mendes N, et al. *Skeletal muscle regeneration and impact of aging and nutrition*. Ageing Res Rev 2016;26:22-36.

[89] Tidball JG, Villalta SA. *Regulatory interactions between muscle and the immune system during muscle regeneration.* Am J Physiol Regul Integr Comp Physiol 2010;298:R1173-87.

[90] Saclier M, Yacoub-Youssef H, Mackey AL, et al. *Differentially activated macrophages orchestrate myogenic precursor cell fate during human skeletal muscle regeneration*. Stem Cells 2013;31:384-96.

[91] Ogle ME, Segar CE, Sridhar S, et al. *Monocytes and macrophages in tissue repair: Implications for immunoregenerative biomaterial design*. Exp Biol Med 2016;241:1084-97.

[92] Ochoa O, Torres FM, Shireman PK. *Chemokines and diabetic wound healing*. Vascular 2007;15:350-5.

[93] Arnold L, Henry A, Poron F, et al. *Inflammatory monocytes recruited after skeletal muscle injury switch into antiinflammatory macrophages to support myogenesis.* J Exp Med 2007;204:1057-69.

[94] Ruffell D, Mourkioti F, Gambardella A, et al. *A CREB-C/EBPbeta cascade induces M2 macrophage-specific gene expression and promotes muscle injury repair.* Proc Natl Acad Sci U S A 2009;106:17475-80.

[95] Summan M, Warren GL, Mercer RR, et al. *Macrophages and skeletal muscle regeneration: a clodronate-containing liposome depletion study.* Am J Physiol Regul Integr Comp Physiol 2006;290:R1488-95.

[96] Mounier R, Théret M, Arnold L, et al. *AMPKα1 regulates macrophage skewing at the time of resolution of inflammation during skeletal muscle regeneration.* Cell Metab 2013;18:251-64.

[97] Burzyn D, Kuswanto W, Kolodin D, et al. *A special population of regulatory T cells potentiates muscle repair*. Cell 2013;155:1282-95.

[98] Tang T-T, Yuan J, Zhu Z-F, et al. *Regulatory T cells ameliorate cardiac remodeling after myocardial infarction.* Basic Res Cardiol 2012;107:232.

[99] Lu H, Huang D, Saederup N, et al. *Macrophages recruited via CCR2 produce insulin-like growth factor-1 to repair acute skeletal muscle injury.* FASEB J 2011;25:358-69.

[100] Tonkin J, Temmerman L, Sampson RD, et al. *Monocyte/macrophage-derived IGF-1 orchestrates murine skeletal muscle regeneration and modulates autocrine polarization.* Mol Ther 2015;23:1189-200.

[101] Sorokin L. *The impact of the extracellular matrix on inflammation*. Nat Rev Immunol 2010;10:712-23.

[102] Collins CA, Zammit PS, Ruiz AP, et al. *A population of myogenic stem cells that survives skeletal muscle aging*. Stem Cells 2007;25:885-94.

[103] Mancini A, Vitucci D, Labruna G, et al. *Effect of lifelong football training on the expression of muscle molecular markers involved in healthy longevity.* Eur J Appl Physiol 2017;117:721-30.

[104] Alfieri A, Martone D, Randers MB, et al. *Effects of long-term football training on the expression profile of genes involved in muscle oxidative metabolism.* Mol Cell Probes 2015;29:43-7.

[105] Martone D, Giacobbe M, Capobianco A, et al. *Exercise intensity and technical demands of small-sided soccer games for under-12 and under-14 players*. J Strength Cond Res 2017;31:1486-92.

[106] Teixeira E, Duarte JA. *Skeletal muscle loading changes its regenerative capacity*. Sport Med 2016;46:783-92.

[107] Raschke S, Eckel J. *Adipo-myokines: two sides of the same coin – Mediators of inflammation and mediators of exercise*. Mediators Inflamm 2013;2013:320724.

[108] Cezar CA, Roche ET, Vandenburgh HH, et al. *Biologic-free mechanically induced muscle regeneration*. Proc Natl Acad Sci U S A 2016;113:1534-9.

[109] Bove GM, Harris MY, Zhao H, et al. *Manual therapy as an effective treatment for fibrosis in a rat model of upper extremity overuse injury*. J Neurol Sci 2016;361:168-80.

[110] Klingler W, Jurkat-rott K, Lehmann-horn F, et al. *The role of fibrosis in Duchenne muscular dystrophy.* Acta Myol 2012;31:184-95

[111] De Luca A, Pierno S, Liantonio A, et al. *Enhanced dystrophic progression in mdx mice by exercise and beneficial effects of taurine and insulin-like growth factor-1*. J Pharmacol Exp Ther 2003;304:453-63.

[112] Ambrosio F, Ferrari RJ, Distefano G, et al. *The synergistic effect of treadmill running on stem-cell transplantation to heal injured skeletal muscle*. Tissue Eng Part A 2010;16:839-49.

[113] Pedersen BK, Febbraio MA. *Muscles, exercise and obesity: skeletal muscle as a secretory organ*. Nat Rev Endocrinol 2012;8:457-65.

[114] Heinemeier KM, Olesen JL, Schjerling P, et al. *Short-term strength training and the expression of myostatin and IGF-I isoforms in rat muscle and tendon: differential effects of specific contraction types.* J Appl Physiol 2007;102:573-81.

[115] Darr KC, Schultz E. *Hindlimb suspension suppresses muscle growth and satellite cell proliferation*. J Appl Physiol 1989;67:1827-34.

[116] Mozdziak PE, Truong Q, Macius A, et al. *Hindlimb suspension reduces muscle regeneration.* Eur J Appl Physiol Occup Physiol 1998;78:136-40.

[117] Wang XD, Kawano F, Matsuoka Y, et al. *Mechanical load-dependent regulation of satellite cell and fiber size in rat soleus muscle.* Am J Physiol Cell Physiol 2006;290:C981-9.

[118] Muñoz-Cánoves P, Scheele C, Pedersen BK, et al. *Interleukin-6 myokine signaling in skeletal muscle: a double-edged sword?* FEBS J 2013;280:4131-48.

[119] Subramony SD, Dargis BR, Castillo M, et al. *The guidance of stem cell differentiation by substrate alignment and mechanical stimulation*. Biomaterials 2013;34:1942-53.

[120] Perris R, Perissinotto D. *Role of the extracellular matrix during neural crest cell migration.* Mech Dev 2000;95:3-21.

[121] Wang H, Li X, Wu Z, et al. *The effect on the extracellular matrix of the deep fascia in response to leg lengthening.* BMC Musculoskelet Disord 2008;9:101.

[122] Freedman BR, Bade ND, Riggin CN, et al. *The (dys)functional extracellular matrix*. Biochim Biophys Acta-Mol Cell Res 2015;1853:3153-64.

[123] Schwartz MA. *Integrins and extracellular matrix in mechanotransduction.* Cold Spring Harb Perspect Biol 2010;2:a005066-a005066.

[124] Millon-Frémillon A, Bouvard D, Grichine A, et al. *Cell adaptive response to extracellular matrix density is controlled by ICAP-1-dependent beta1-integrin affinity*. J Cell Biol 2008;180:427-41.

[125] Moore SW, Roca-Cusachs P, Sheetz MP. *Stretchy proteins on stretchy substrates: the important elements of integrin-mediated rigidity sensing.* Dev Cell 2010;19:194-206.

[126] Hoffman BD, Grashoff C, Schwartz MA. *Dynamic molecular processes mediate cellular mechanotransductio*n. Nature 2011;475:316-23.

[127] Bershadsky AD, Balaban NQ, Geiger B. *Adhesion-dependent cell mechanosensitivity*. Annu Rev Cell Dev Biol 2003;19:677-95.

[128] Musarò A. *The basis of muscle regeneration*. Adv Biol 2014;2014:1-16.

[129] Wu H, Xiong WC, Mei L. *To build a synapse: signaling pathways in neuromuscular junction assembly.* Development 2010;137:1017-33.

[130] Lluri G, Langlois GD, McClellan B, et al. *Tissue inhibitor of metalloproteinase-2 (TIMP-2) regulates neuromuscular junction development via a beta1 integrin-mediated mechanism.* J Neurobiol 2006;66:1365-77.

[131] McMahan UJ, Slater CR. *The influence of basal lamina on the accumulation of acetylcholine receptors at synaptic sites in regenerating muscle.* J Cell Biol 1984;98:1453-73.

[132] Garg K, Corona BT, Walters TJ. *Therapeutic strategies for preventing skeletal muscle fibrosis after injury*. Front Pharmacol 2015;6:87.

[133] Sommese L, Zullo A, Schiano C, et al. *Possible muscle repair in the human cardiovascular system*. Stem Cell Rev 2017;13:170-91.

[134] Grimaldi V, Mancini FP, Casamassimi A, et al. *Potential benefits of cell therapy in coronary heart disease*. J Cardiol 2013;62:267-76.

[135] Grimaldi V, Schiano C, Casamassimi A, et al. *Imaging techniques to evaluate cell therapy in peripheral artery disease: state of the art and clinical trials*. Clin Physiol Funct Imaging 2016;36:165-78.

[136] Mertens JP, Sugg KB, Lee JD, et al. *Engineering muscle constructs for the creation of functional engineered musculoskeletal tissue.* Regen Med 2014;9:89-100.