Physiol Rev
84: 209–238, 2004; 10.1152/physrev.00019.2003.

Cellular and Molecular Regulation of Muscle Regeneration

SOPHIE B. P. CHARGÉ AND MICHAEL A. RUDNICKI

Ottawa Health Research Institute, Ottawa, Canada

Chargé, Sophie B. P., and Michael A. Rudnicki. Cellular and Molecular Regulation of Muscle Regeneration. *Physiol Rev* 84: 209–238, 2004; 10.1152/physrev.00019.2003.—Under normal circumstances, mammalian adult skeletal muscle is a stable tissue with very little turnover of nuclei. However, upon injury, skeletal muscle has the remarkable ability to initiate a rapid and extensive repair process preventing the loss of muscle mass. Skeletal muscle repair is a highly synchronized process involving the activation of various cellular responses. The initial phase of muscle repair is characterized by necrosis of the damaged tissue and activation of an inflammatory response. This phase is rapidly followed by activation of myogenic cells to proliferate, differentiate, and fuse leading to new myofiber formation and reconstitution of a functional contractile apparatus. Activation of adult muscle satellite cells is a key element in this process. Muscle satellite cell activation resembles embryonic myogenesis in several ways including the de novo induction of the myogenic regulatory factors. Signaling factors released during the regenerating process have been identified, but their functions remain to be fully defined. In addition, recent evidence supports the possible contribution of adult stem cells in the muscle regeneration process. In particular, bone marrow-derived and muscle-derived stem cells contribute to new myofiber formation and to the satellite cell pool after injury.

I. INTRODUCTION

The primary functions of skeletal musculature are locomotor activity, postural behavior, and breathing. However, skeletal muscle is susceptible to injury after direct trauma (e.g., intensive physical activities, lacerations) or resulting from indirect causes such as neurological dysfunction or innate genetic defects. If left unrepaired, these injuries may lead to loss of muscle mass, locomotive deficiency, and in the worse cases lethality. The maintenance of a working skeletal musculature is conferred by its remarkable ability to regenerate. Indeed, upon muscle injury a finely orchestrated set of cellular responses is activated, resulting in the regeneration of a well-innervated, fully vascularized, and contractile muscle apparatus. The advances of molecular biology techniques combined with the identification and development of rodent models for muscular dystrophy have contributed to the identification of molecular pathways involved in muscle regeneration. In particular, the identification of muscle satellite cells has led to major advances in our understanding of muscle regeneration. Significant research into the biology of satellite cells has elucidated the cellular and molecular mechanisms during muscle regeneration. These studies have also led to insight regarding the development of therapeutic strategies that may alleviate some of the pathological conditions associated with poor muscle regenerative capacity, such as the one observed in muscular dystrophy patients and in the course of normal aging. More recently, the identification of multipotent stem cells capable of myogenic differentiation in the course of muscle regeneration has extended our view on the muscle regenerative process and opened new perspectives for the development of novel therapies. However, despite extensive research to unravel the process of skeletal muscle regeneration, the complex regulatory pathways remain poorly understood.

In this review, the current understanding of the cellular and molecular processes of skeletal muscle regeneration is presented. First, the embryonic development as well as the structure and function of normal skeletal muscle are briefly presented. Indeed, muscle regeneration appears to recapitulate to some extent the embryonic developmental process. Second, the morphological characteristics of injured muscle and the various experimental models used to study skeletal muscle regeneration are introduced. Third, the role of adult muscle satellite cells in muscle repair is discussed, focusing on the morphological and molecular characterization of these cells and their activation after damage. Fourth, the current knowledge on the role for nonmuscle and muscle resident adult stem cells in muscle regeneration is reviewed. Finally, the Urodele dedifferentiation process is discussed in the context of mammalian muscle regeneration.

A. Skeletal Muscle Development: An Overview

All vertebrate skeletal muscles (apart from head muscles) are derived from mesodermal precursor cells originating from the somites (epithelial spheres of paraxial mesoderm) (for review, see Ref. 15). During embryonic development, specification of mesodermal precursor cells to the myogenic lineage is regulated by positive and negative signals from surrounding tissues (summarized in Fig. 1*A*). Specification to the myogenic lineage requires the upregulation of *MyoD* and *Myf5*, basic helix-loop-helix transcriptional activators of the myogenic regulatory factor family (MRF) (Fig. 1). This is demonstrated by the total loss of skeletal muscle in *MyoD:Myf5* double knockout mice and the observation that putative muscle progenitor cells remain multipotential and contribute to nonmuscle tissues in the trunk and limbs of these mice (155, 232, 259; for review, see Ref. 15). Proliferative *MyoD* and/or *Myf5* positive myogenic cells are termed myoblasts. Proliferating myoblasts withdraw from the cell cycle to become terminally differentiated myocytes that express the "late" MRFs, *Myogenin* and *MRF4*, and subsequently muscle-specific genes such as myosin heavy chain (*MHC*) and muscle creatine kinase (*MCK*) (Fig. 1*B*). *Myogenin*-deficient embryos die perinatally due to a deficit in myoblast differentiation as evidenced by an almost total absence of myofibers in these mutants (141, 223). Similarly, *MRF4*-deficient mice display a range of phenotypes consistent with a late role for *MRF4* in the myogenic pathway (236, 249, 357, 348). Finally, mononucleated myocytes specifically fuse to each other to form multinucleated syncytium, which eventually mature into contracting muscle fibers (Fig. 1*B*). During the course of muscle development, a distinct subpopulation of myoblasts fails to differentiate, but remains associated with the surface of the developing myofiber as quiescent muscle satellite cells (Fig. 1*B* and discussed below). After sexual maturity, skeletal muscle is a stable tissue characterized by multinucleated postmitotic muscle fibers (85, 266).

B. Adult Skeletal Muscle Characteristics

The muscle fibers are the basic contractile units of skeletal muscles. They are individually surrounded by a connective tissue layer and grouped into bundles to form a skeletal muscle (Fig. 2). As well as being rich in connective tissue, skeletal muscles are highly vascularized to provide essential nutrients for muscle function (Fig. 2*A*, arrowhead). As the myofiber matures, it is contacted by a single motor neuron and expresses characteristic molecules for contractile function, principally different MHC isoforms and metabolic enzymes (Fig. 2*B*). Both the motor neuron and the myoblast origin have been implicated to play a role in specifying the myofiber contractile prop-

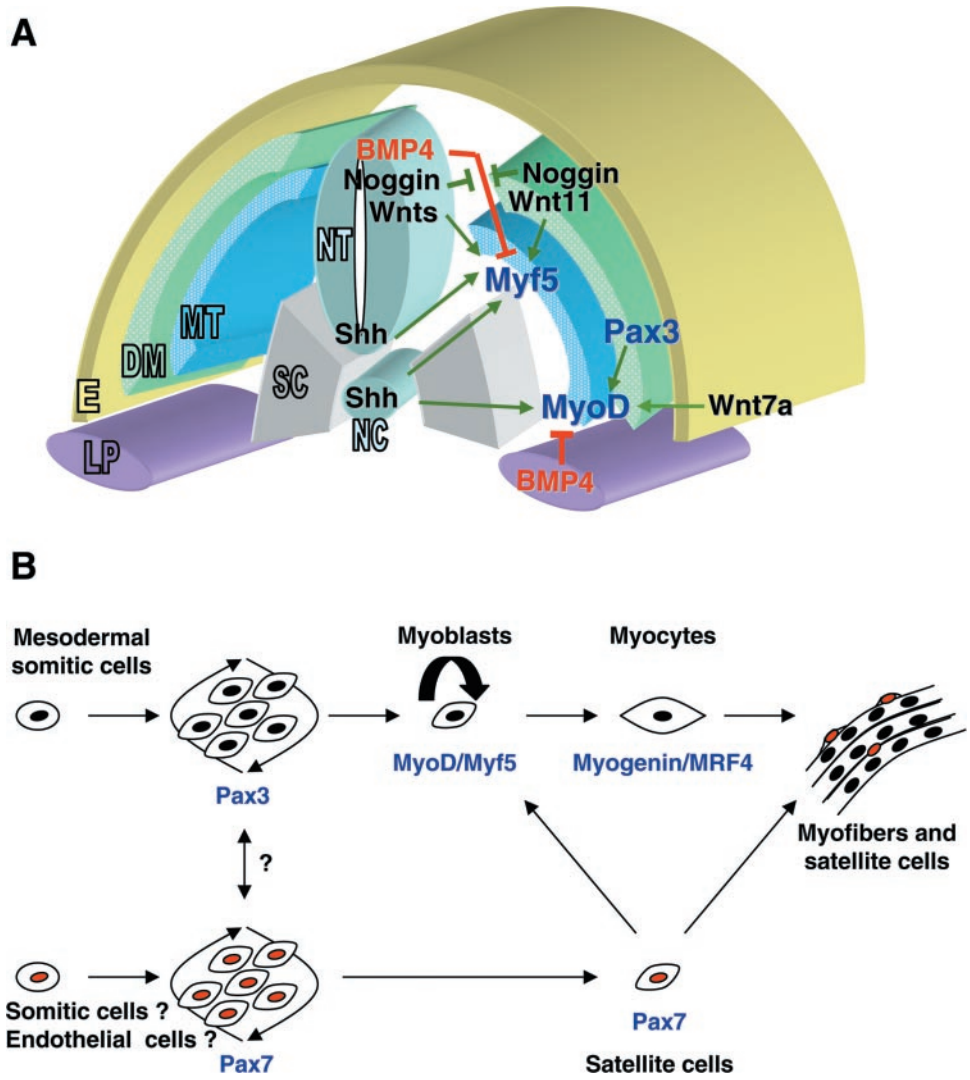

FIG. 1. Signaling factors and cellular events involved in embryonic skeletal muscle formation. *A*: mesodermal somitic cells located in the dorsal part of the somite [dermomyotome (DM)] receive signals from surrounding tissues, which induce [*Wnts*, Sonic hedgehog (*Shh*), *Noggin*] or inhibit (BMP4) the expression of the primary MRFs (*Myf5* and *MyoD*) and commitment to the myogenic lineage. Commited myoblasts migrate laterally to form the myotome (MT), which eventually forms the skeletal musculature. *Pax3* promotes myogenesis in the lateral myotome. E, ectoderm; LP, lateral plate; SC, sclerotome; NC, notochord; NT, neural tube. *B: Pax3* expression in precursor cells contributes to myogenic cell expansion. After *Myf5* and/or *MyoD* induction, mesodermal somitic cells are committed to the myogenic lineage (myoblasts). Later, upregulation of the secondary MRFs (myogenin and MRF4) induces terminal differentiation of myoblasts into myocytes. Finally, myocyte fusion gives rise to multinucleated myofibers. During the later phase of embryonic myogenesis, a distinct population of myoblasts, derived from satellite cells, fuses to existing myofibers enabling myofiber growth. Some satellite cells remain closely associated with myofibers in a quiescent undifferentiated state. The embryonic origin of satellite cells remains to be determined; however, *Pax7* expression is essential for the specification/expansion of the satellite cell population.

erties, although the precise mechanisms remain to be defined (reviewed by Ref. 335). Nevertheless, individual adult skeletal muscles are composed of a mixture of myofibers with different physiological properties, ranging from a slow-contracting/fatigue-resistant type to a fast-contracting/non-fatigue-resistant type. The proportion of each fiber type within a muscle determines its overall contractile property (Fig. 2*B*). Despite having different physiological properties, the basic mechanism of muscle contraction is similar in all myofiber types and is the result of a "sliding mechanism" of the myosin-rich thick filament over the actin-rich thin filament after neuronal activation (for review, see Ref. 147). The connective tissue framework in skeletal muscle combines the contractile myofibers into a functional unit, in which the contraction of myofibers is transformed into movement via myotendinous junctions at their ends, where myofibers attach to the skeleton by tendons. Thus the functional properties of skeletal muscle depend on the maintenance of a complex framework of myofibers, motor neurons, blood vessels, and extracellular connective tissue matrix. Although this review focuses on the regeneration process of the myofibers, it is understood that revascularization, reinnervation, and reconstitution of the extracellular matrix are also essential aspects of the muscle regeneration process.

C. Morphological Characteristics of Skeletal Muscle Regeneration

Adult mammalian skeletal muscle is a stable tissue with little turnover of nuclei (85, 266). Minor lesions inflicted by day-to-day wear and tear elicit only a slow turnover of its constituent multinucleated muscle fibers. It is estimated that in a normal adult rat muscle, no more than 1–2% of myonuclei are replaced every week (266). Nonetheless, mammalian skeletal muscle has the ability to complete a rapid and extensive regeneration in response to severe damage. Whether the muscle injury is inflicted by a direct trauma (i.e., extensive physical activ-

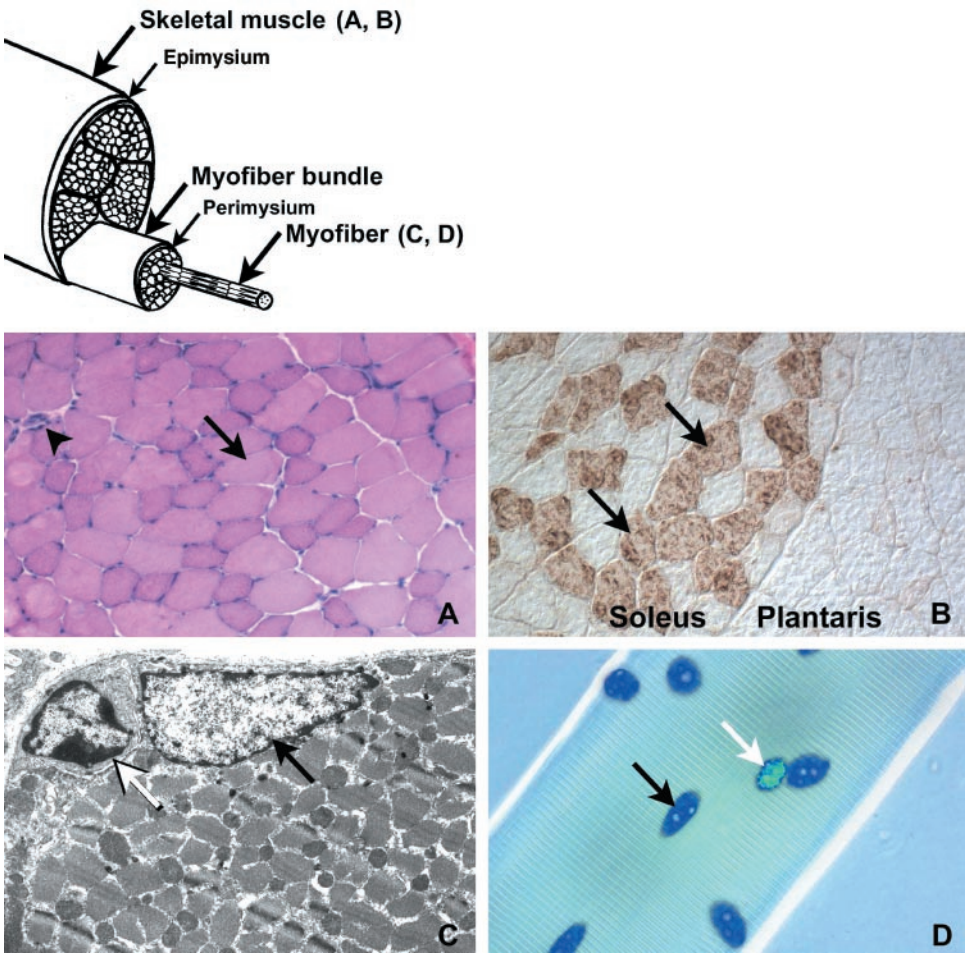

FIG. 2. Morphological characteristics of adult mammalian skeletal muscle. The primary components of skeletal muscles are the myofibers grouped in bundles within the perimysium. *A*: myofibers are multinucleated syncytia with their postmitotic myonuclei located at the periphery as seen in muscle cross-section stained with hematoxylin and eosin (arrow). Skeletal muscles are highly vascularized to provide essential nutrients for muscle function (arrowhead). *B*: myofibers are heterogeneous with respect to their contractile properties, ranging from slow/oxidative to fast/glycolytic types. The proportion of each fiber type within a muscle determines its overall contractile property. The slow contracting soleus muscle is rich in myofibers expressing the slow type I myosin heavy chain isoform as illustrated by the mosaic pattern displayed following immunostaining with an antibody specific to slow myosin heavy chain (arrows), whereas the fast contracting plantaris muscle is devoid of slow type I myofibers. *C* and *D*: the adult skeletal muscle contains a population of quiescent muscle satellite cells. Muscle satellite cells are closely associated with myofibers, located within the same basal lamina as seen by electron microscopy (*C*). Muscle satellite cell nuclei (white arrow) can be distinguished from myonuclei (black arrow) by their abundant heterochromatin reflecting their mitotic quiescence. Muscle satellite cells are present on myofibers isolated by mild enzymatic digestion (*D*) and are characterized by their high levels of *Pax7* expression as demonstrated by immunocytochemistry (white arrow) compared with myonuclei (black arrow).

ity and especially resistance training) or innate genetic defects, muscle regeneration is characterized by two phases: a degenerative phase and a regenerative phase (Fig. 3*A*).

The initial event of muscle degeneration is necrosis of the muscle fibers. This event is generally triggered by disruption of the myofiber sarcolemma resulting in increased myofiber permeability. Disruption of the myofiber

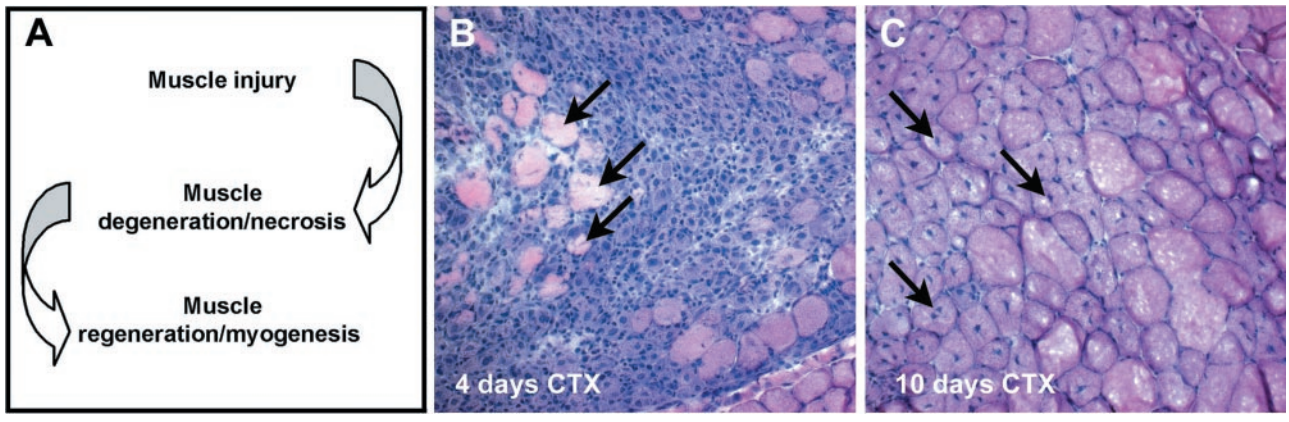

FIG. 3. Skeletal muscle repair process. *A*: mammalian skeletal muscle repair process is characterized by a degenerative phase followed by a regenerative phase. *B*: injury to the tibialis anterior muscle by cardiotoxin (CTX) injection results in the rapid necrosis of myofibers and the activation of an inflammatory response leading to the loss of muscle architecture (compare Fig. 3*B* with Fig. 2*A*). *C*: myofiber regeneration is characterized by the activation of myogenic cells to proliferate, differentiate, and fuse to necrotic fibers for repair or to each other for new fiber formation. Regenerating fibers are characterized by their small caliber and their centrally located myonuclei (arrows).

integrity is reflected by increased serum levels of muscle proteins, such as creatine kinase, which are usually restricted to the myofiber cytosol. In human and animal models, increased serum creatine kinase is observed after mechanical stress (e.g., extensive physical exercises) and in the course of muscle degenerative diseases such as muscular dystrophies, all of which are characterized by the induction of a muscle regeneration process (79, 226, 238; reviewed in Refs. 13, 184, 292, 355). Reciprocally, the uptake of low-molecular-weight dyes, such as Evans blue or procion orange, by the myofiber is a reliable indication of sarcolemmal damage and is also associated with strenuous exercise and muscle degenerative diseases (46, 135, 201, 231, 297, 298). It has been hypothesized that increased calcium influx after sarcolemmal or sarcoplasmic reticulum damage results in a loss of calcium homeostasis and increased calcium-dependent proteolysis that drives tissue degeneration (reviewed in Refs. 2, 12, 30). Calpains are calcium-activated proteases that can cleave myofibrillar and cytoskeletal proteins and hence are implicated in the process (180; reviewed in Refs. 30, 90). Thus disrupted myofibers undergo focal or total autolysis depending on the extent of the injury.

The early phase of muscle injury is usually accompanied by the activation of mononucleated cells, principally inflammatory cells and myogenic cells. Present reports suggest that factors released by the injured muscle activate inflammatory cells residing within the muscle, which in turn provide the chemotactic signals to circulating inflammatory cells (reviewed in Refs. 247, 315). Neutrophils are the first inflammatory cells to invade the injured muscle, with a significant increase in their number being observed as early as 1–6 h after myotoxin or exercise-induced muscle damage (104, 230). After neutrophil infiltration and ~48 h postinjury, macrophages become the predominant inflammatory cell type within the site of injury (230, 315). Macrophages infiltrate the injured site to phagocytose cellular debris and may affect other aspects of muscle regeneration by activating myogenic cells (7, 191, 212, 256). Moreover, studies demonstrating the stimulation of peritoneal macrophages after intensive physical exercise suggest that a systemic factor capable of inducing an inflammatory response throughout the body is released following muscle damage (99, 196, 339). Although several mediators involved in the activation of the inflammatory response have been characterized, further studies are necessary to demonstrate their potential role in the muscle regeneration process in vivo (reviewed in Ref. 315). Thus muscle fiber necrosis and increased number of nonmuscle mononucleate cells within the damaged site are the main histopathological characteristics of the early event following muscle injury (Fig. 3*B*).

Muscle degeneration is followed by the activation of a muscle repair process. Cellular proliferation is an important event necessary for muscle regeneration as demonstrated by the reduced muscle regenerative capacity after exposure to colchicine (an inhibitor of mitotic division) or after irradiation (241, 244, 325, 331). Notably, the expansion of myogenic cells provides a sufficient source of new myonuclei for muscle repair (reviewed in Refs. 48, 129, 142). Numerous nuclear radiolabeling experiments have demonstrated the contribution of dividing myogenic cells to regenerating myofibers, and it is well accepted that following the myogenic proliferation phase, new muscle fibers are formed much as during bona fide embryonic myogenesis; myogenic cells differentiate and fuse to existing damaged fibers for repair or to one another for new myofiber formation (82, 289, 290, 326). Long-standing histological characteristics are still used to identify the mammalian skeletal muscle regeneration process. On muscle cross-sections, these fundamental morphological characteristics are newly formed myofibers of small caliber and with centrally located myonuclei (Fig. 3*C*). Newly formed myofibers are often basophilic (reflecting high protein synthesis) and express embryonic/developmental forms of MHC (reflecting de novo fiber formation) (134, 333). On muscle longitudinal sections and in isolated single muscle fibers, central myonuclei are observed in discrete portions of regenerating fibers or along the entire new fiber, suggesting that cell fusion is not diffuse during regeneration but rather focal to the site of injury (41). Fiber splitting or branching is also a characteristic feature of muscle regeneration and is probably due to the incomplete fusion of fibers regenerating within the same basal lamina (38, 41, 43). Fiber splitting is commonly observed in muscles from patients suffering neuromuscular diseases, in hypertrophied muscles, and in aging mouse muscles, all of which are associated with abnormal regenerative capacity (42, 57, 61, 264). Once fusion of myogenic cells is completed, newly formed myofibers increase in size, and myonuclei move to the periphery of the muscle fiber. Under normal conditions, the regenerated muscle is morphologically and functionally indistinguishable from undamaged muscle.

D. Animal Models of Muscle Injury

Although the degenerative phase and the regenerative phase of the muscle regeneration process are similar among different muscle types and after varying causes of injuries, the kinetics and amplitude of each phase may vary depending on the extent of the injury, the muscle injured, or the animal model (149, 187, 217, 237, 255). To study the process of muscle regeneration in a controlled and reproducible way, it has therefore been necessary to develop animal models of muscle injury.

The use of myotoxins such as bupivacaine (Marcaine), cardiotoxin (CTX), and notexin (NTX) is perhaps the easiest and most reproducible way to induce muscle

regeneration (81, 133, 138, 139). These toxins have a wide range of biological activities, which are not entirely understood. For example, NTX is a phospholipase A_2 neurotoxin peptide extracted from snake venoms that block neuromuscular transmission by inhibition of acetylcholine release. CTX is also a peptide isolated from snake venoms, but it is a protein kinase C-specific inhibitor that appears to induce the depolarization and contraction of muscular cells, to disrupt membrane organization, and to lyse various cell types. In our laboratory, 25 μl of 10 μM CTX (from *Naja nigricollis* snake venom) injected in adult mouse tibialis anterior muscle induces muscle degeneration leading to a wound coagulum with mononuclear cell infiltration within 1 day of injection. Inflammatory response and mononuclear cell proliferation is most active within 1–4 days of injection (Fig. 3*B*). Myogenic cell differentiation and new myotube formation is observed ~5–6 days postinjection. By 10 days postinjection, the overall architecture of the muscle is restored, although most regenerated myofibers are smaller and display central myonuclei (Fig. 3*C*). The return to a morphologically and histochemically normal mature muscle is seen at ~3–4 wk postinjection. Although injection of CTX is a highly reproducible way of inducing muscle regeneration, the potentially unknown effects of this toxin on various muscle cell types including satellite cells is a potential "caveat" to this protocol.

Alternative methods to myotoxin injection, which are possibly more physiologically relevant, are available. For example, the direct infliction of a wound by crushing and/or freezing the muscle or the denervation-devascularization by transplantation of a single muscle will trigger the process of muscle regeneration (187, 272). Transplantation of the extensor digitorum longus in the rat leads to rapid degeneration (within 2–3 days) of the transplanted fibers followed by the rapid appearance of regenerating fibers (within 5 days) and leading to a normal muscle by 60 days (51, 136, 137). Muscle regeneration can also be induced by repeated bouts of intensive exercise, and in fact, eccentric exercise (lengthening contraction) is particularly potent at inducing muscle damage (149; reviewed in Refs. 13, 98). Thus appropriate procedures that promote muscle damage will induce a controlled regeneration process.

Laboratory mouse models with abnormal degeneration due to the spontaneous or artificial deregulation of

TABLE 1. *Targeted germline mutations in mice affecting muscle degeneration/regeneration process*

Targeted Mutation	Adult Muscle Phenotype	Muscle Regeneration Phenotype	Satellite Cell Phenotype (In Vitro)
	Transcription factors		
MyoD$^{-/-}$ (210, 260)	Minor alterations	Impaired	Increased proliferation Delayed differentiation
Pax7$^{-/-}$ (275) (P. Seale and S. Chargé, unpublished observations)	Growth deficit, satellite cells absent	Deficient	No satellite cell
Slug$^{-/-}$ (358)	Fairly normal	Impaired	ND
MNF$^{-/-}$ (114)	Growth deficit	Impaired	Decreased proliferation Normal differentiation
	DGC components		
MCK (Cre)-dystroglycan (LoxP) (68)	Increased degeneration without fibrosis or fat replacement, muscle mass increase	Efficient	ND
Dystrophin$^{-/-}$ (*mdx*) (42, 68, 235)	Increased degeneration with fibrosis and fat replacement	Impaired in older mice	Normal proliferation
δ-Sarcoglycan$^{-/-}$ (68)	Increased degeneration with fibrosis and fat replacement	Impaired in older mice	ND
	Growth factors		
FGF-6$^{-/-}$ (109)	Fairly normal	Impaired	ND
FGF-6$^{-/-}$ (106)	Normal	Normal	ND
LIF$^{-/-}$ (178)	Normal	Impaired	ND
MSTN$^{-/-}$ (208, 324)	Muscle mass increase (hyperplasia and hypertrophy)	Improved	ND
	Others		
M-cadherin$^{-/-}$ (144)	Normal	Normal	ND
Desmin$^{-/-}$ (193, 288)	Some degeneration	Delayed	ND

MNF, myocyte nuclear factor; MCK, muscle creatine kinase; FGF, fibroblast growth factor; LIF, leukemia inhibitory factor; MSTN, myostatin; ND, not determined. Reference numbers are given in parentheses.

specific genes are also of interest (Table 1). For example, the *mdx* mouse is commonly used as an animal model of Duchenne muscular dystrophy (DMD) and as an alternative degeneration model for studying muscle repair (55; reviewed in Refs. 39, 328). The *mdx* mouse is a spontaneously occurring mouse line deficient for dystrophin because of a point mutation in exon 23 of the dystrophin gene, which forms a premature stop codon (283). Dystrophin is a major component of the dystrophin-glycoprotein complex (DGC), which links the myofiber cytoskeleton to the extracellular matrix. Disruption of this complex leads to increased susceptibility to contraction-induced injury and sarcolemmal damage leading to myofiber necrosis. Although *mdx* mice are normal at birth, skeletal muscles show extensive signs of muscle degeneration by 3–5 wk of age (79, 235, 304). This acute muscle degeneration phase is accompanied by an effective regeneration process leading to a transient muscle hypertrophy (79, 235). After this period, the degeneration/regeneration activity continues at lower and relatively constant levels throughout the life span of the animal. However, for reasons that remain unclear, in the older animals (~15 mo), muscle regeneration process is defective and the mice become extremely weak and die before wild-type littermates (68, 234, 235). To date there are various animal models in which the DGC has been disrupted by knocking out dystrophin-associated proteins such as dystroglycans and sarcoglycans (Table 1). These also provide useful insight in the degeneration/regeneration regulatory pathways (68, 297).

A large variety of animal models are available for studying muscle regeneration; however, the mouse strain influences muscle regeneration (125, 128, 149, 205, 217, 255). For example, the regeneration process in Swiss SJL/J mice is more efficient than in Balb/c, C57BL6J, and B6AF-1, while A/J mice appear the least efficient. The underlying mechanisms for such strain differences are unclear, but the correlation between regeneration capacity and genetic background suggests the involvement of modifier genes. Differential expressions of *Pax7* isoforms and *MyoD* have been correlated with efficiency of repair (165–167, 197). Furthermore, expression levels of basic fibroblast growth factor (FGF) have been correlated with the efficiency of repair within Balb/c and SJL/J muscles (10). Another experimental consideration is the observation that the muscle regeneration process follows a centripetal gradient (from the outer regions to the inner regions), which results in the formation of different zones within a regenerating muscle, each zone being in a different phase of degeneration or regeneration (51). The use of laboratory rodents has been fundamental in the understanding of mammalian skeletal muscle regeneration; through the use of such models the primary cellular component for muscle regeneration has been established to be the adult muscle satellite cell.

II. ADULT MUSCLE SATELLITE CELLS

A. Identification of Muscle Satellite Cells

Muscle satellite cells are a population of undifferentiated mononuclear myogenic cells found in mammalian (49, 113, 202), avian (140), reptilian (157), and amphibian (202, 242) skeletal muscles including muscle spindles. When cultured in vitro, satellite cells display specific characteristics allowing their distinction from embryonic and fetal myoblasts (75, 77, 140). Muscle satellite cells are apparent as a distinct population of myoblasts during the midfetal (E12–13) to late stage (E18) of avian development (100, 140), during the 10th to 14th week of human limb development (75) and from around E17.5 in limbs of mouse embryos (reviewed in Ref. 76). Thus, in all organisms analyzed to date, the muscle satellite cell population appears distinct from the embryonic and fetal myoblasts populations. Furthermore, the temporal appearance of satellite cells follows the appearance of both embryonic and fetal myoblasts.

Since their first description by Mauro (202), muscle satellite cells have been primarily identified in situ by their morphological characteristics. Indeed, muscle satellite cells can unequivocally be identified by electron microscopy due to their distinct location within the basal lamina surrounding individual myofibers, juxtaposed between the plasma membrane of the muscle fiber and the basement membrane, hence their name (Fig. 2*C*, white arrow). Other important morphological features of satellite cells are an increased nuclear-to-cytoplasmic ratio, a reduced organelle content, and a smaller nucleus size displaying increased amounts of heterochromatin compared with fiber myonuclei (Fig. 2*C*, white arrow). These characteristics reflect the finding that satellite cells are mitotically quiescent and transcriptionally less active than myonuclei (269, 291). The identification of satellite cells by light microscopy is more ambiguous, although the use of markers such as laminin and dystrophin to respectively identify the basal lamina and the myofiber sarcolemma facilitate their identification. Moreover, the development of techniques to isolate and study single muscle fibers with their resident satellite cells in vitro has allowed great advances in understanding this cell population (Fig. 2*D*) (33, 258). Nevertheless, the difficult in vivo identification of satellite cells has hindered the study of this cell population and in effect the understanding of skeletal muscle regeneration. To circumvent such difficulties, scientists are focusing on identifying molecular markers specific to this cell population (summarized in Table 2 and discussed herein) (reviewed in Ref. 142).

TABLE 2. *Satellite cell markers*

Molecular Markers	Satellite Cell Expression		Experimental Protocol
	Quiescent	Proliferating	
Cell surface			
M-cadherin (74, 150)	+/−	+	In vivo/in vitro
Syndecan-3 (73)	+	+	In vivo/in vitro
Syndecan-4 (73)	+	+	In vivo/in vitro
c-*met* (74)	+	+	In vivo/in vitro
VCAM-1 (153)	+	+	In vivo
NCAM (148)	+	+	In vivo
Glycoprotein Leu-19 (148, 267)	+	+	In vivo/in vitro
CD34 (28)	+/−	+/−	In vitro
Cytoskeletal			
Desmin (42, 74)	−	+	In vivo/in vitro
Transcription factors			
Pax7 (275)	+	+	In vivo/in vitro
Myf5 (28, 74)	+/−	+	In vivo/in vitro
MyoD (74)	−	+	In vivo/in vitro
MNF (115)	+	+	In vivo/in vitro
MSTN (73, 168, 211)	+	−/+	In vitro/in vivo
IRF-2 (153)	+	+	In vivo
Msx1 (73)	+	−	In vitro

MSTN, myostatin; VCAM-1, vascular cell adhesion molecule-1; NCAM, neural cell adhesion molecule; MNF, myocyte nuclear factor; IRF-2, interferon regulatory factor-2. Reference numbers are given in parentheses.

B. Dynamics of Muscle Satellite Cells

Satellite cells are present in all skeletal muscles and are associated with all muscle fiber types, albeit with unequal distribution. For instance, the percentage of satellite cells in adult slow soleus muscle is two- to threefold higher than in adult fast tibialis anterior or extensor digitorum longus muscles (Table 3) (117, 258, 265, 291). Similarly, high numbers of satellite cells are found associated with slow muscle fibers compared with fast fibers within the same muscle (117). Although these differences in satellite cell density between fiber types are well established, the regulatory mechanisms behind this phenomenon are less well understood. Increased density of satellite cells have been observed at the motor neuron junctions (312, 337) and adjacent to capillaries (265), suggesting that some factors emanating from these structures may play a role in homing satellite cells to specific muscle locations or in regulating the satellite cell pool by other means. The regulation of satellite cell density at the single fiber level is also suggestive of a role for the muscle fiber in regulating the satellite cell pool.

The satellite cell population varies also with age (Table 3). Compelling evidence suggests a decrease in satellite cell density over time. During postnatal muscle growth, there is a dramatic decrease in the proportion of satellite cell nuclei. This decrease is mainly due to the dramatic increase in myonuclei number following satellite cell fusion. However, in some glycolytic muscles, it is combined with a net decrease in total satellite cell number (118). At birth, satellite cells account for 30% of sublaminar muscle nuclei in mice followed by a decrease to <5% in a 2-mo-old adult (35). After sexual maturity, satellite cell number continues to decrease, albeit not as dramatically (42, 61, 118, 273). At 1–4 mo of age, most murine single myofibers in culture yield satellite cells, whereas from 9 to 12 mo of age, up to 50% of extensor digitorum longus fibers fail to yield any satellite cells under similar culture conditions (Table 3) (42, 61). Thus the overall satellite cell number appears to decrease as a function of age.

C. Embryonic Origin of Muscle Satellite Cells: Somitic Versus Endothelial

Satellite cells are believed to constitute a myogenic cell lineage distinct from embryonic and fetal myoblast lineages (Fig. 1*B*). However, although the origin of embryonic and fetal myoblasts has been extensively studied and researchers have unanimously concluded that these myogenic precursors originate from the multipotential mesodermal cells of the somites (reviewed in Refs. 15, 229), the origin of satellite cells has been the subject of fewer studies and remains unclear with, to date, two standing hypotheses: a somitic versus an endothelial origin.

TABLE 3. *Satellite cell number in skeletal muscle of different ages and type*

Animal Model	Muscle	Age, mo	Satellite Cell Nuclei, %	Number of Desmin$^+$ Cells/Fiber	Fiber Yielding No Cell, %
Mouse cross-sections (291)	EDL	5–7	1.2		
	Soleus	5–7	4.1		
Rat cross-sections (118)	EDL	1	7		
		12	2.9		
		24	1.9		
	Soleus	1	9.6		
		12	6.6		
		24	4.7		
Rat cross-sections (265)	TA	2	4		
	Soleus	2	11		
Mouse single fiber explant (42)	EDL	0.5		116 ± 16	0
		1		91 ± 11	0
		1.5		64 ± 5	0
		3.5		18 ± 4	25
		18		10 ± 3	50
Mouse single fiber explant (61)	EDL	1.5–2		30 ± 6	1
		4		37 ± 5	0
		12–13		3 ± 1	50

Number of desmin$^+$ cell/fiber represents mean ± SE of desmin$^+$ cells associated with an explanted single fiber following 90 h of culture in growth media (42) or 72 h in low growth factors media (61). Percent fiber yielding no cell represents the proportion of single fibers associated with no mononucleated cells after at least 72 h in culture. EDL, extensor digitorum longus; TA, tibialis anterior. Percent satellite cell represents the number of satellite cell nuclei per total number of satellite cell nuclei plus myonuclei. Reference numbers are given in parentheses.

The somitic origin hypothesis emanates from traditional transplantation studies performed in avian models (11). In this fate-mapping analysis, embryonic somites from donor quail embryos were introduced into host chick embryos. After embryonic development, the contribution of quail cells to the host satellite cell compartment is determined using ultrastructural characteristics specific to quail nuclei to identify the donor cells. In this study, donor somitic cells are found to migrate from the somites into the developing chick limb contributing to both terminally differentiated muscle fibers and the host satellite cell population. Although the identification of quail nuclei was never definite and the somitic domain generating satellite cells was never characterized, this study suggests a common somitic origin for all myogenic cell lineages, including muscle satellite cells.

Several recent observations have challenged the view of a somitic origin for satellite cells. For example, bone marrow-derived myogenic cells are able to participate in skeletal muscle regeneration, although at low frequency, when injected intravenously, suggesting that some myogenic cells with similar functional characteristics as satellite cells originate from bone marrow-derived stem cells (discussed in sect. v) (37, 103, 132, 181). Subsequently, more studies have demonstrated the ability of nonmuscle resident cells to follow the myogenic lineage (reviewed in Ref. 119). However, more definitive conclusions come from the detailed clonal analysis of different mouse tissues at various developmental stages by De Angelis et al. (83). In this elegant study, the authors demonstrate the presence of clonal myogenic precursors within the embryonic dorsal aorta. When cultured in vitro, these clones display similar morphological characteristics and express myogenic and endothelial markers similar to that of adult muscle satellite cells. Furthermore, the same study shows that myogenic precursor clones can be derived from limbs of *c-Met*$^{-/-}$ and *Pax3*$^{-/-}$ mutants, which lack appendicular musculature due to the absence of migratory myoblasts of somitic origin. Thus myogenic precursors derived from these mutant limbs may be of endothelial origin. When directly injected into regenerating host muscles, these cells are incorporated into newly regenerating fibers, a hallmark of bona fide satellite cells. When embryonic aortas are transplanted into muscles of newborn immunodeficient mice, they can also give rise to many myogenic cells within the treated muscles and also within collateral untreated muscles. Moreover, when fetal limbs are transplanted under the skin of host animals and become vascularized by the host, myogenic cells of host origin are observed within the transplant. Taken together, these results suggest the presence of a multipotential cell population within the embryonic vasculature, which may differentiate as a function of the tissue it perfuses (83, 216). In the case of the skeletal muscle, the progenitors have the capacity to adopt a myogenic fate (83). However, although it is clear that these endothelial cells can contribute to new muscle fiber formation during muscle development and regeneration, it remains to be determined whether these cells can contribute to the quiescent sublaminar cell population historically defined as a satellite cell population (202). Indeed, the endothelial progenitors could represent an alternative cell population to satellite cells capable of muscle growth and repair. It is noteworthy, however, that adult satellite cells, in contrast to embryonic and fetal myoblasts, express both endothelial and myogenic markers, such as CD34 and MRFs (28).

The recent view of an endothelial origin for satellite cells is not mutually exclusive with the more traditional view of a somitic origin. In fact, during early embryogenesis the aortic endothelium and the somites are adjacent, suggesting a close proximity in origin of these two lineages (reviewed in Refs. 228, 233). Thus the presence of myogenic cells within the embryonic dorsal aorta does not rule out the possibility of an indirect somitic origin of the satellite cells. Moreover, emerging evidence that satellite cells may represent a heterogeneous population may be a reflection of this dual origin (reviewed in Ref. 213). Moreover, in the course of differing physiological or pathological states, myogenic cells of different origin may contribute differently to the myogenic repair. Thus myogenic repair may involve the activation of various myogenic cells depending on the extent of the injury or the local environment, and in particular, in response to damaged vasculature. Taken together, these studies highlight the need for further experiments designed to unequivocally identify the embryonic origin(s) of muscle satellite cells. Such studies will require the use of classical chimeric studies combined with lineage analysis using retroviral or genetic labeling.

D. Specification/Expansion of Muscle Satellite Cells: Role of *Pax7*

Although the embryonic origin of satellite cells remains to be determined, the gene responsible for the specification of progenitor cells to the satellite cell lineage has been recently identified (275). Using representational difference analysis (RDA) of cDNAs, our group has isolated *Pax7* as a gene specifically expressed in cultured satellite cells and demonstrated its expression in quiescent and activated satellite cells in vivo (146, 275) (Fig. 2*D*). The *Pax7* gene is a member of the paired box containing gene family of transcription factors implicated in development of the skeletal muscle of the trunk and limbs, as well as elements of the central nervous system (reviewed in Refs. 65, 198). *Pax7* is closely related to *Pax3*, based on highly similar protein structures and partially overlapping expression patterns during embryonic development (121, 154). Interestingly, *Pax3* is a key reg-

ulator of somitic myogenesis (200, 303). Detailed analysis of the distribution of *Pax7* mRNA using Northern blot analysis by Seale et al. (275) demonstrated the expression of *Pax7* in proliferating satellite cell-derived myoblasts and a rapid downregulation of *Pax7* transcripts upon myogenic differentiation. Low levels of *Pax7* expression were also detected in proliferating C_2C_{12} mouse myoblasts, which is an established cell line originally derived from satellite cells (40, 344). However, *Pax7* was not expressed at detectable levels in a variety of nonmuscle cell lines. In addition, analysis of poly(A)$^+$ RNA from selected mouse tissues revealed expression of *Pax7* at low levels in adult skeletal muscles only. Specific expression of *Pax7* within muscle satellite cells in vivo was confirmed by in situ hybridization and immunocytochemistry analyses on fresh frozen muscle sections. *Pax7* mRNA and protein were found in a subset of nuclei (~5%) in discrete peripheral locations within undamaged wild-type skeletal muscle. Furthermore, the number of *Pax7*-positive cells increased in muscles undergoing regeneration such as in *MyoD*$^{-/-}$, *mdx*, and *mdx:MyoD*$^{-/-}$ skeletal muscles. Centrally located nuclei within newly regenerated muscle fibers were also associated with *Pax7* expression, suggesting that recently activated and fusing satellite cells express *Pax7*. Together, these data demonstrate the specific expression of *Pax7* in quiescent and activated muscle satellite cells (275).

The analysis of *Pax7*$^{-/-}$ skeletal muscles demonstrated the important role for *Pax7* in satellite cell development. Indeed, *Pax7*$^{-/-}$ mice appear normal at birth but fail to grow postnatally, leading to a 50% decrease in body weight by 7 days of age compared with wild-type littermates (199, 275). *Pax7* mutant animals fail to thrive and usually die within 2 wk after birth (199, 275). This runted phenotype is characterized by a decreased skeletal muscle mass resulting from a fiber size decrease rather than a decrease in fiber number (S. Chargé, unpublished observation; Ref. 275). *Pax7*$^{-/-}$ skeletal muscles have a striking absence of satellite cells (275). Under standard derivation and growth conditions, primary cell cultures from mutant skeletal muscles failed to generate myoblasts; instead, mutant cultures were uniformly composed of fibroblasts and adipocytes. Furthermore, morphological analysis of mutant skeletal muscles by transmission electron microscopy confirmed the lack of satellite cells in *Pax7*-deficient musculature. Overall, the data to date suggest a key role for *Pax7* in lineage determination, especially in the specification of myogenic progenitors to the satellite cell lineage. Recent studies have highlighted the multiple functions of the *Pax* genes, implicating Pax proteins in regulating organogenesis and maintaining proliferating, pluripotent stem cell populations (reviewed in Refs. 65, 198). *Pax7* is unequivocally required for satellite cell development. However, whether *Pax7* has a role in the specification or the survival of the satellite cell progenitor pool remains unclear. Understanding the molecular pathways regulated by *Pax7* should prove useful in understanding the early event of satellite cell development.

III. MUSCLE SATELLITE CELLS IN MUSCLE REPAIR

The activation of satellite cells upon muscle injury resulting from mechanical trauma, direct injury to the muscle, or in the course of a disease is well characterized (for review, see Refs. 48, 129, 142). Moreover, when transplanted into regenerating muscle, cultured satellite cells contribute to new myofiber formation as well as to reconstitution of satellite cell population for later rounds of regeneration (41, 124, 143, 194, 290). Furthermore, preliminary experiments performed in our laboratory suggest that in *Pax7*$^{-/-}$ mice, which lack muscle satellite cells, normal skeletal muscle regeneration is dramatically reduced (P. Seale and S. Chargé, unpublished observations). Thus the activation of muscle satellite cells appears an important step in the ability of muscle to regenerate.

A. Activation of Muscle Satellite Cell Upon Injury: Role of MRFs

In the course of muscle regeneration, satellite cells first exit their normal quiescent state to start proliferating. After several rounds of proliferation, the majority of the satellite cells differentiate and fuse to form new myofibers or to repair damaged one. Satellite cell activation is not restricted to the damaged site. Indeed, damage at one end of a muscle fiber will activate satellite cells all along this fiber leading to the proliferation and migration of the satellite cells to the regeneration site (272). However, recruitment of satellite cells from adjacent muscles is seldom observed and requires damage to the connective tissue separating the two muscles (271, 272). After proliferation, quiescent satellite cells are restored underneath the basal lamina for subsequent rounds of regeneration (272). The process of satellite cell activation and differentiation during muscle regeneration is reminiscent of embryonic muscle development. In particular, the critical role of the MRFs is observed in both processes (Figs. 1 and 4).

Upon exposure to signals from the damaged environment, quiescent satellite cells are activated and start proliferating at which stage they are often referred to as myogenic precursor cells (mpc) or adult myoblasts (Fig. 4). At the molecular level, activation of mpc is characterized by the rapid upregulation of two MRFs, *Myf5* and *MyoD* (71, 73, 74, 110, 127, 197, 246, 285, 341, 353). In general, quiescent satellite cells do not have any detectable levels of MRFs. Although a low level of *Myf5-lacZ*

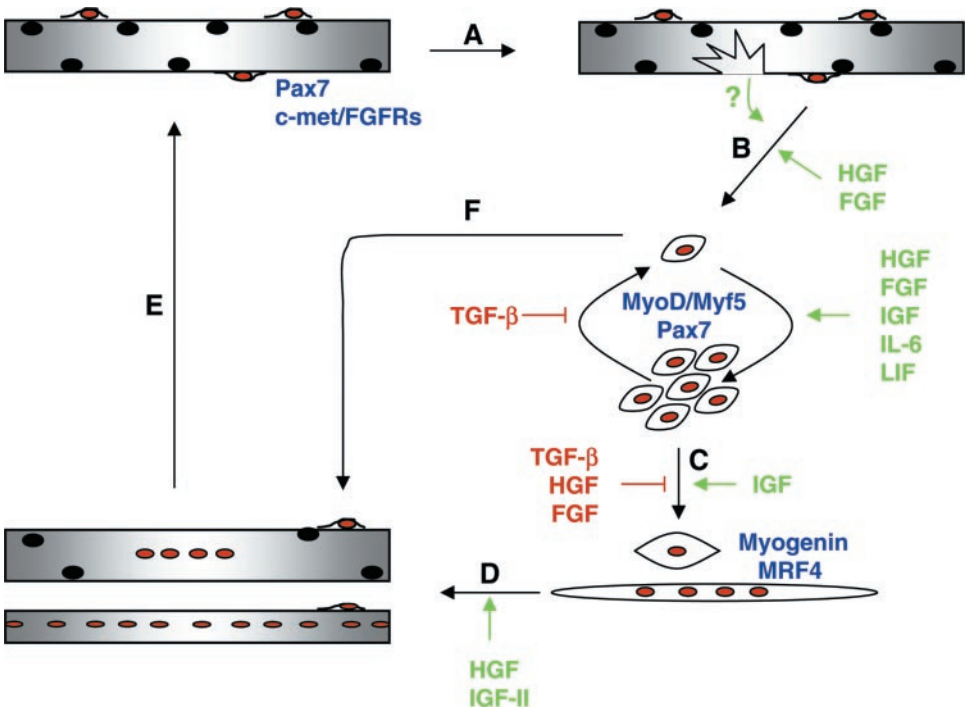

FIG. 4. Schematic representation of the molecular events regulating muscle satellite cell activation during skeletal muscle regeneration. Following damage to the myofiber (*A*), quiescent satellite cells are activated to enter the cell cycle and proliferate, allowing for expansion of the myogenic cell population (*B*). Activated satellite cells are characterized by high expression of the MRFs MyoD and Myf5. The proliferative phase is followed by terminal differentiation (*C*) and fusion of myoblasts to damaged myofibers for repair or to each other for new myofiber formation (*D*). Myoblast terminal differentiation is characterized by the upregulation of the MRFs Myogenin and MRF4. Finally, repaired or new myofibers grow to resemble original myofibers (*E*). During the course of muscle regeneration, a subset of myoblasts reenters the quiescent state to replenish the satellite cell pool for subsequent muscle repair (*F*). In vitro and in vivo experiments have highlighted the possible role for several growth factors among which positive (green arrows) and negative (red lines) regulators are presented. HGF, hepatocyte growth factor; FGF, fibroblast growth factor; IGF, insulin-like growth factor; IL-6, interleukin-6; LIF, leukemia inhibitory factor; TGF-β, transforming growth factor-β family.

expression has been reported in a subset of quiescent satellite cells using knock-in mice, this observation is likely an allele-specific phenomenon (28). Upon satellite cell activation, *MyoD* upregulation appears the earliest within 12 h of activation and is detectable before any sign of cellular division such as proliferative cell antigen nuclear (PCNA) expression (71, 73, 74, 79, 285, 341). A sensitive multiplex expression analysis at the single-cell level suggested that some satellite cells enter the MRF-positive compartment by expressing either *Myf5* or *MyoD*; however, this state is rapidly followed by coexpression of the two (74). Activation of *MyoD* and *Myf5* expression following muscle injury has also been observed in various in vivo models for muscle regeneration and in varying muscle types (32, 71, 110, 127, 246). Of particular interest is the study by Cooper et al. (71), which confirmed the initial upregulation of *Myf5* and/or *MyoD*, followed by the coexpression of these MRFs by using CTX to induce muscle regeneration in *Myf5-nlacZ* mice combined with MyoD immunostaining. These data suggest an important role for *MyoD* in satellite cell differentiation. Supporting this view is the observation by Megeney et al. (210) that $MyoD^{-/-}$ mice have a reduced regenerative capacity characterized by an increase in mpc population and a decrease in regenerated myotubes. Furthermore, $MyoD^{-/-}$ muscles display an increased occurrence of branched myofibers suggestive of chronic or inefficient muscle regeneration in vivo (73). By electron microscopy, $MyoD^{-/-}$ satellite cells are morphologically normal (210). However, in vitro cultures of $MyoD^{-/-}$ satellite cells demonstrate a myogenic cell population with abnormal morphology characterized by a stellate, flattened appearance compared with the compact rounded appearance displayed by normal mouse myoblasts (260). Under low serum conditions, which usually are favorable for myogenic differentiation, $MyoD^{-/-}$ cells continue to proliferate and eventually yield a reduced number of mononucleated differentiated myocytes (73, 260, 342). The increased level of insulin-like growth factor I (IGF-I) expression in $MyoD^{-/-}$ cells is in accordance with the previously reported role for IGF-I in promoting myoblast proliferation (discussed in sect. IV*C*) (107, 260). In normal satellite cells, MyoD may downregulate IGF-I expression to promote myogenic differentiation. In addition, expression of M-cadherin is decreased in $MyoD^{-/-}$ cells (73, 260), and a requirement for M-cadherin has been reported for myoblast differentiation and fusion (150, 356). A recent study has isolated Slug, a zinc-finger protein of the snail family, as a direct downstream target of MyoD (358). Endogenous MyoD was shown to bind to the Slug promoter, and in vitro reporter analysis demonstrated the direct activation of this promoter by MyoD (358). Slug expression is dramatically increased in the late phase (4–10 days post-CTX injection) of muscle regeneration. Moreover, induction of muscle regeneration by CTX injection in $Slug^{-/-}$ muscle reveals a defective regenerative capacity with rare centrally nucleated fibers (i.e., rare regenerating myofibers) and smaller regenerated muscle area. Even though the data are suggestive of a role for Slug in muscle regeneration, the authors are rightly cautious in concluding that a number of developmental abnormalities could give rise to such defect. Indeed, Slug is a transcription factor with

a broad expression pattern and is likely to play regulatory roles in multiple processes (67, 276). Nevertheless, one attractive hypothesis is that the muscle regeneration defect observed in *MyoD*$^{-/-}$ mice stems from the inappropriate expression of Slug. Understanding the downstream targets of Slug may shed some light on the molecular pathways regulating mpc activation. Overall, these data suggest an important role for MyoD in the process of satellite cell differentiation during muscle regeneration. However, further studies are required to determine the downstream targets of MyoD important in the regenerative process. Moreover, it remains unclear whether lack of MyoD during embryonic development or in mature fibers has an effect on muscle regeneration independently of satellite cell activity. A recent study argues against this possibility, wherein antisense nucleotides to inhibit *MyoD* expression during adult muscle regeneration were used successfully and muscle regeneration appeared somewhat delayed (352). In the future, this technique should prove useful in inhibiting individual factors during muscle regeneration in mice that have a similar developmental background.

Identifying a role for *Myf5* in muscle regeneration has been more problematic. Indeed, until recently no viable *Myf5*$^{-/-}$ mice were available (164). Although *Myf5*-deficient mice display a delayed epaxial (deep back muscle) embryonic myogenesis and a normal hypaxial (trunk and limb muscles) embryonic myogenesis, no apparent phenotype in the adult muscle has been reported to date (reviewed in Ref. 156). These data combined with the reciprocal delay in hypaxial myogenesis in *MyoD*-deficient mice and the mutually exclusive expression of *Myf5* and *MyoD* in early stages of embryonic muscle precursor cells have led to the hypothesis that Myf5 and MyoD support distinct myogenic lineages during embryonic muscle development (reviewed in Ref. 156). Similarly, circumstantial evidence suggests that Myf5 and MyoD may play distinct roles during muscle regeneration: Myf5 promotes satellite cell self-renewal (discussed below), whereas MyoD promotes satellite cell progression to terminal differentiation (discussed above). Analysis of viable *Myf5*-deficient mice under regeneration conditions and of *Myf5*-deficient myoblasts in culture should shed some light into these mechanisms.

After the mpc proliferation phase, expression of *Myogenin* and *MRF4* (MRF members) is upregulated in cells beginning their terminal differentiation program (Fig. 4) (73, 74, 110, 127, 197, 285, 341). This is followed by the activation of the cell cycle arrest protein p21 and permanent exit from the cell cycle. The differentiation program is then completed with the activation of muscle-specific proteins, such as MHC, and the fusion of mpc to repair damaged muscle. The observation that MRF4 expression is in myonuclei of newly regenerated myofibers or young myotubes at a time after fusion suggests a distinct role from Myf5, MyoD, and Myogenin, possibly in myofiber maturation (359). Gross defects in embryonic muscle development of mutant mice for Myogenin and MRF4 have impeded further study of these genes in muscle regeneration.

Other factors important in regulating myogenic differentiation, possibly by acting upstreams of the MRFs, have been identified. Mice deficient in myocyte nuclear factor (MNF), a winged helix transcription factor important in regulating myoblasts cell cycle progression, are impaired in their ability to regenerate skeletal muscle (114, 115, and reviewed in 142). Recently, Fernando et al. (102) have demonstrated the requirement for caspase-3/MST1 signaling in initiating myoblasts differentiation in vitro. However, the requirement for caspase-3 activity for normal muscle regeneration remains to be demonstrated. Thus the activation of satellite cells following muscle injury results in the activation of the myogenic program, which allows expansion of the myogenic cell pool necessary for new myonuclei fusion and myofiber formation (Fig. 4).

B. Fusion of Muscle Precursor Cells

During the course of muscle regeneration, akin to during muscle development, mpc are required to specifically fuse to each other to form syncytial muscle fibers. Semi-stable intercellular junction structures that mediate cell-cell adhesion and regulate intracellular cytoskeleton architecture are important in the course of such complex tissue organization. Classical cadherins, which are transmembrane proteins mediating cell-cell interactions in a calcium-dependent manner, are thought to play important roles in these processes (reviewed in Refs. 116, 163). M-cadherin, in particular, has been postulated as an essential molecule for the specific fusion of myoblasts with each other during embryonic myogenesis and muscle regeneration (163, 219, 356). First, the preferential expression of M-cadherin in developing and regenerating skeletal muscles, as well as in skeletal muscle cell lines, is suggestive of a role for this molecule in myoblast fusion. More specifically, although M-cadherin mRNA can be detected in only a small subset of quiescent satellite cells, its protein remains constant in most of these cells. Moreover, M-cadherin expression within satellite cells is markedly induced upon muscle injury, suggesting a possible role for this protein in the muscle repair process (150, 219). Second, in vitro experiments using antagonistic peptides and antisense RNA strategies have demonstrated the essential role played by M-cadherin during myoblast fusion process without affecting the biochemical differentiation of myoblasts, as seen by normal upregulation of muscle-specific genes (175, 356). Finally, *MyoD*$^{-/-}$ satellite cells, which fail to fuse upon injury-induced activation, display a

marked decreased in M-cadherin expression (73, 260). However, the essential role for M-cadherin in myoblast fusion has recently been placed in question by the analysis of *M-cadherin*$^{-/-}$ mice (144). In this careful analysis, M-cadherin-deficient mice did not show any gross developmental defect. In particular, the mutant mice developed normal skeletal musculature and demonstrated normal kinetics of muscle regeneration after CTX injection (144). The authors rightly suggest that such observation may be the result of a compensatory mechanism by other cadherins, such as N-cadherin and R-cadherin, which are present in skeletal muscle and may substitute for M-cadherin function. N-cadherin expression is upregulated in activated satellite cells following injury, but its function in myoblasts fusion remains to be determined (62). Thus, although M-cadherin may have an important role in fusion of myoblasts during muscle regeneration, it is not essential and other cadherins may also play a role during this process.

The role of m-calpain has also been suggested in cytoskeletal reorganization during myoblast fusion. M-calpain belongs to a family of calcium-dependent intracellular nonlysosomal cysteine proteases of relatively unknown functions (reviewed in Ref. 293). M-calpain activity is dramatically increased during fusion of embryonic primary myoblasts (78, 86, 180). In vitro fusion of myoblasts is prevented by calpastatin (a specific inhibitor of m- and μ-calpains) or by decreasing levels of m-calpain using an antisense strategy (20, 311). Conversely, myoblasts fuse earlier and faster after m-calpain injection or artificial decrease of endogenous calpastatin levels using antisense RNA (20, 311). The biological role for m-calpain in myoblast fusion is unclear because substrates for this proteinase during this process are unknown. A potential target of m-calpain is the intermediate filament desmin (90). Interestingly, cytoplasmic intermediate filament proteins such as vimentin, desmin, and nestin have been implicated in myoblast fusion during muscle regeneration (288, 319). Moreover, although the overall muscle formation in *Desmin*$^{-/-}$ mice appears normal, muscle regeneration appears impaired with delayed myoblast fusion (193, 288). Further analyses are required to determine the specific role for m-calpain and/or desmin in satellite cell fusion during regeneration. Recent advances uncovering the interacting extracellular molecules and the intracellular effectors that facilitate myoblast fusion in *Drosophila* should benefit the understanding of the mammalian myoblast fusion process (reviewed in Ref. 93).

C. Self-Renewal of Muscle Satellite Cells

Satellite cell self-renewal is a necessary process without which recurrent muscle regeneration would rapidly lead to the depletion of the satellite cell pool. Radiolabel-tracing experiments demonstrated that activated satellite cells contributed to both new myonuclei and satellite cell after muscle damage (124, 131, 143, 268, 272, 287, 347). Moreover, labeling experiments in the growing rat demonstrated the presence of two satellite cell populations (268). One population, representing ~80% of the satellite cells, divided rapidly and was responsible for providing myonuclei to the growing rat. The other population, called "reserve cells," divided more slowly and was suggested to replenish the satellite cell pool (268). These observations are consistent with the idea that a small proportion of satellite cells that has undergone proliferation returns to the quiescent state, thereby replenishing the satellite cell pool (23, 24, 29, 268, 349). Satellite cell self-renewal may result from an asymmetric division generating two distinguishable daughter cells, one committed to myogenic differentiation and one stem cell "self." Alternatively, satellite cells may undergo symmetric division with one daughter cell being able to withdraw from the differentiation program and return to quiescence. Neither hypothesis has been proven wrong. Determining the molecular process involved in this mechanism remains a challenge.

In favor of satellite cell asymmetric division is the recent observation by Conboy and Rando (70) that Numb, a plasma membrane-associated cytoplasmic protein, is asymmetrically segregated within dividing satellite cells in vitro. Segregation of Numb proteins has been associated with cell fate determination in both invertebrate and vertebrate developmental processes including during *Drosophila* myogenesis (54, 58, 169, 252). Numb may influence cell fate by repressing Notch signaling, a pathway which is known to regulate cellular differentiation in different systems and species (88). In cultured satellite cells, activation of Notch-1 appears to promote the proliferation of "primitive" satellite cells (Numb−/Pax3+/Desmin−/Myf5−/MyoD−), whereas its inhibition leads to the commitment of the progenitor cells to the myoblast cell fate (Numb+/Pax3−/Desmin+/Myf5+) and their myogenic differentiation (70). Overall, the data suggest that asymmetric satellite cell division as marked by asymmetric inheritance of Numb may lead to satellite cell self-renewal by causing different patterns of gene expression. However, the specific role for *Pax* genes and Myf5 in this process remains to be substantiated.

Several lines of evidence suggest a role for Myf5 in facilitating satellite cell self-renewal. The increased proliferation and decreased differentiation phenotype of *MyoD*$^{-/-}$ cells is consistent with the notion that *MyoD*-deficient cells represent an intermediate stage between quiescent satellite cells and mpc (73, 260, 342). That observation and the demonstration that upon activation satellite cells express either Myf5 alone or MyoD alone, prior to coexpressing both MRFs and initiating terminal differentiation, have led to the hypothesis that Myf5+/MyoD− cells represent a developmental stage during which satel-

lite cells undergo self-renewal. Interestingly, when human satellite cells or C_2C_{12} murine cell line are induced to differentiate, a small population of undifferentiated Myf5+/MyoD− cells persists (24, 349). Moreover, these cells retain the capacity to self-renew and to give rise to differentiation-competent progeny (24, 349). However, expression of *Myf5* in quiescent satellite cells is controversial (28, 74), suggesting that return to the quiescent state may require downregulation of *Myf5*. Thus satellite cell symmetric division followed by dedifferentiation of one of the daughter cells remains a possibility, especially in light of recent findings that mammalian myotubes are capable of dedifferentiation in vitro. This process may involve the activation of the transcriptional inhibitor *Msx1* (discussed in sect. VI).

Furthermore, renewal of the satellite cell pool may not rely exclusively on the satellite cell compartment. Indeed, adult stem cells, other than satellite cells, capable of myogenic differentiation and of contributing to the satellite cell pool following transplantation have been described (discussed in sect. V). The observation that in the absence of *Pax7* the number of hematopoietic precursors in muscle-derived stem cells is increased at the expense of myogenic precursors suggests a role for *Pax7* in this process, by *Pax7* promoting the determination of satellite cells and restricting alternative developmental pathways in multipotent stem cells (275).

Whatever the cellular mechanism(s) for satellite cell self-renewal, this does not appear to compensate for the chronic loss of myonuclei throughout a lifetime as reflected by the reduction in satellite cell number with aging (see sect. II*B* and Table 3), nor does it compensate for the depletion of the satellite cell pool resulting from continuous activation of muscle repair in dystrophic muscles (42, 235, 329, 340). Exhaustion of the mitotic potential of satellite cells, or replicative senescence, may be responsible for the decrease in the satellite cell pool with age (85, 273, 329). For example, in DMD patients, where satellite cell proliferation is accentuated, telomere lengths (an indicator of the cell replicative age) are prematurely reduced compared with normal human senescence (84). Moreover, proliferative potential of satellite cells is reduced with age or after repeated rounds of regeneration in animal models (240, 270, 273, 340). Alternatively, the inability to sustain a constant satellite cell number may reflect an alteration in the aging environment rather than intrinsic defect in cellular capacity (26, 42, 50, 59, 60). Furthermore, the observation that multiple rounds of acute muscle regeneration do not deplete the satellite cell pool suggests that satellite cell self-renewal after a short but acute loss of myonuclei (i.e., muscle injury) is more efficient than after chronic covert myonuclei turnover (i.e., life time of day-to-day wear and tear). Thus different mechanisms for satellite cell self-renewal may be at play during varying muscle injuries and at varying ages.

D. Multipotentiality of Muscle Satellite Cells

It has been established for several years now that the commitment of skeletal myocytes is reversible under appropriate tissue culture conditions. Primary myoblasts from newborn mice and C_2C_{12} can differentiate into osteogenic or adipogenic cells after in vitro treatment with bone morphogenetic proteins (BMP2) or adipogenic inducers (thiazollidinedione or fatty acids), respectively (162, 310). However, it is only recently that similar multipotential properties have been demonstrated for the adult muscle satellite cell (14, 323). Until these reports, the adult muscle satellite cell was generally considered a stem cell committed to the myogenic lineage. Work from our laboratory and others demonstrated the BMP induced osteogenic and adipogenic conversions of isolated adult murine satellite cells (14, 323). The osteogenic differentiation of primary myoblasts is characterized by a transient coexpression of myogenic markers (such as *MyoD*, *Myf5*, and *Pax7*) and osteogenic markers (such as alkaline phosphatase), suggesting a direct transdifferentiation from the myogenic lineage to the osteogenic lineage, rather than the passage through a common noncommitted progenitor. In vitro culture of single myofibers suggests the spontaneous conversion of satellite cells to the osteogenic and adipogenic lineages is a rare phenomenon. Satellite cells on freshly isolated muscle fibers do not express the myogenic markers *Myf5* and *MyoD*, suggesting that quiescent satellite cells are more plastic and may enter nonmyogenic pathways more readily (74, 197, 285, 341). Supporting this view is the finding that Msx1, a homeobox protein involved in C_2C_{12} dedifferentiation (see sect. VI), is expressed in quiescent satellite cell but downregulated upon satellite cell activation (73). Myoblasts from $MyoD^{-/-}$ mice do not display increased propensity to osteogenic and/or adipogenic differentiations, suggesting that *MyoD* alone may not suppress differentiation of myoblasts to these lineages (14). Overall, these data demonstrate the in vitro ability of satellite cells to differentiate into osteogenic or adipogenic lineages. However, satellite cell differentiation potential appears to be restricted to the mesenchymal range of cell lineages as demonstrated by their inability to undergo hematopoietic conversion (16).

Several in vivo observations have suggested the existence of mesenchymal progenitors within skeletal muscles. For example, ectopic bone formation within skeletal muscle has been described in some human diseases (reviewed in Ref. 282). Furthermore, accumulation of adipose tissue has been widely reported in skeletal muscle undergoing degeneration such as in DMD patients or the related *mdx* mice model and in other models of muscle regeneration (91, 235, 261). Overall, the data support the hypothesis that muscle satellite cells may be involved in the formation of adipogenic and osteogenic tissues under certain in vivo circumstances. Aberrant activation of sat-

ellite cells during muscle regeneration may lead to such reversal of lineage commitment at the expense of effective muscle regeneration. However, the hypothesis remains to be proven in vivo.

IV. ROLE OF SECRETED FACTORS IN THE REGULATION OF MUSCLE REGENERATION

Skeletal muscle regeneration is a highly orchestrated process that involves the activation of adult muscle satellite cells to proliferate and differentiate (Fig. 4). Activation of satellite cells requires the timely, controlled upregulation of muscle transcription factors and muscle specific genes (discussed in sect. III). This process is regulated through mechanisms involving cell-cell and cell-matrix interactions as well as extracellular secreted factors. Muscle injuries have been shown to cause the release of biologically active molecules into the extracellular space. For example, extracts from crushed muscle, but not from intact muscle, contain mitogens for satellite cells (34, 63). Different stimuli have been proposed as initiators of satellite cell activation; extract from the injured fibers, molecules released by the invading macrophages, and soluble factors from connective tissue have all been proposed (96, 191, 299; for review, see Ref. 126). In vitro studies have implicated an extensive number of trophic factors, including members of FGF and transforming growth factor-β (TGF-β) families, IGF, hepatocyte growth factor (HGF), tumor necrosis factor-α (TNF-α), interleukin-6 (IL-6) family of cytokines, neural-derived factors, nitric oxide, and ATP, in maintaining a balance between growth and differentiation of satellite cells to restore a normal muscle architecture (reviewed in Refs. 142, 296). These studies have contributed to our knowledge on the effect of trophic factors, singly or in combination, on the proliferative and differentiative capacity of satellite cells in vitro. Nevertheless, the physiological functions in skeletal muscle regeneration in vivo have been shown for relatively few of these factors.

A. HGF

Scatter factor/HGF was originally isolated from sera of partially hepatectomized rats and found to have mitogenic activity on hepatocyte primary cultures (224, 225). HGF is now considered as one of the most important growth factors involved in organ regeneration through its mitogenic and motogenic properties (reviewed in Ref. 354). Of particular interest, HGF is a key regulator of satellite cell activity during muscle regeneration. The first association of HGF with skeletal muscle regeneration was reported by Jennische et al. (152) who detected HGF transcripts in regenerating muscles. It is now commonly accepted that HGF transcripts and protein levels are increased during the early phase of muscle regeneration, and this increase is in proportion to the degree of injury (300, 306, 308). Furthermore, HGF isolated from extracts of injured muscles was shown to be the primary muscle factor capable of inducing quiescent satellite cell activation, on the basis of immunoneutralization experiments (306). In vitro, HGF can stimulate quiescent satellite cells to enter the cell cycle and thus increase mpc proliferation, as well as it can inhibit mpc differentiation (6, 111, 214, 306). The role for HGF signaling in mpc proliferation is supported by the demonstration that forced expression of an activated form of c-*met*, the HGF receptor, in C_2C_{12} results in their morphological transformation and subsequent inhibition of differentiation (8). Furthermore, injection of HGF protein to injured muscle blocks the repair process, while increasing mpc number by approximately threefold, confirming the mitogenic role of HGF on satellite cells in vivo (214, 306). HGF's role in muscle regeneration is most important during the early phase of the repair process as demonstrated by the decrease in HGF immunostaining with time after injury and the inability of exogenous HGF injection to affect muscle regeneration when performed at later stages of muscle regeneration (214, 306). Moreover, HGF may play a role in promoting satellite cell migration to the site of injury, via activation of the Ras-Ral pathway, as demonstrated by the in vitro chemotactic activity of this factor on satellite cells and C_2C_{12} (36, 301). Taken together, these data demonstrate the pleiotropic role that HGF probably plays during the early stages of muscle regeneration. HGF appears to increase the mpc population by means of mitogenic and chemotactic activities, possibly resulting in an optimal myoblast density whereupon fusion can commence.

HGF appears to act directly on muscle satellite cells as suggested by its receptor *c-met* expression in quiescent and activated satellite cells (74, 306). Furthermore, the presence of HGF transcripts in newly regenerated myotubes and in satellite cells suggests that HGF activity is mediated through paracrine/autocrine mechanisms (8, 111, 281). HGF may also be released from the muscle extracellular matrix through damage to the basal lamina. Indeed, in vitro and in vivo data have suggested that release of nitric oxide synthase (NOS) from the basal lamina after myofiber stretch or damage leads to the production of nitric oxide (NO), which in turn may activate the release of HGF from its extracellular binding to heparan sulfate proteoglycans (HSPGs) (9, 307). However, the recent finding that a rapid (within an hour) upregulation of HGF transcripts and protein is observed in the spleen following muscle injury in the rat suggests that HGF from intact organs may also have an important role in muscle regeneration (300). Thus HGF activation of quiescent satellite cell may be the result of endocrine and paracrine/autocrine regulatory mechanisms.

B. FGFs

In addition to HGF, several FGFs have been described as potent activators of mpc proliferation and as inhibitors of mpc differentiation, suggesting a role for these factors in the expansion of the mpc compartment (5, 92, 122, 161, 188, 203, 281, 336, 343, 345). However, their role in muscle regeneration remains unclear. Of particular interest is FGF-6. FGF-6 expression is muscle specific and is upregulated during muscle regeneration (87, 109, 161). However, contradictory results have been obtained from the study of FGF-6-deficient mice. Floss et al. (109) have shown the reduced muscle regeneration capacity of these mice after crush injury or when interbred with *mdx* mice. This regenerative deficit was characterized by a decrease in the number of MyoD and myogenin-positive cells and an increase in collagen deposition. In contrast, Fiore and colleagues (105, 106) observed no regenerative defects in response to either NTX injection, crush injury, or *mdx* interbreeding in mice carrying a similar targeted null mutation in FGF-6. This discrepancy is not well understood; similar protocols for inducing muscle injury and similar targeting constructs for deleting FGF-6 were used. As in many other cases of knockout analyses which fail to provide a detectable abnormal phenotype, redundant factors may be involved. Other FGFs, and in particular FGF-4, are potential redundant factors to FGF-6. Interbreeding *FGF-6*$^{-/-}$ mice with other FGF knockouts could be informative. FGF-2 is also a likely candidate for regulating satellite cell activity during regeneration. Indeed, FGF-2 is particularly potent on myoblast activation in vitro. Moreover, FGF-2 is present in basement membrane surrounding developing and mature myotubes (72). The injection of a neutralizing antibody against FGF-2 into the muscle at the time of lesion reduced the number and diameter of regenerating myofibers, suggesting a delay in proliferation and/or fusion of activated satellite cells (189). Moreover, injection of FGF-2 in *mdx* mice appeared to improve satellite cell proliferation and muscle regeneration (188). On the contrary, the injection of FGF-2 at various doses and different time schedules after muscle injury in mice did not affect muscle repair, suggesting that FGF levels in some regeneration models are not a limiting factor of muscle repair (218). Nevertheless, even though FGFs may not play a critical role in activating satellite cells during muscle repair, their role in muscle regeneration may reside in the revascularization process during regeneration through their recognized angiogenic properties (190).

Four receptors for FGF, FGFR 1–4, have been identified, each having differing affinity for individual FGFs. FGFR-1 and -4 are the most prominent transcripts in satellite cells. Moreover, FGFR-1 expression is upregulated dramatically during the early phase of satellite cell activation in vitro in response to FGF-1 exposure, and this effect is further potentiated by addition of HGF (280). However, FGFR-1 is not specific to muscle, since it is also expressed in fibroblasts. The availability of these receptors plays a crucial role in myogenesis as demonstrated by the increased myoblast proliferation and decreased differentiation upon expression of a full-length FGFR-1 and the converse decrease in proliferation and increase in differentiation upon expression of a truncated FGFR-1 in myoblasts in vitro (263). Thus FGFs released from muscle and nonmuscle cells appear to act directly to activate satellite cell proliferation and inhibit their differentiation.

FGFRs and c-Met are transmembrane receptor tyrosine kinases. Upon FGF or HGF binding, the receptor dimers autophosphorylate and activate complex downstream signaling events that remain poorly understood. FGFs and HGF are dependent on heparan sulfate to facilitate receptor activation and intracellular signal transduction (reviewed in Ref. 248). Cell surface HSPGs are found almost ubiquitously on the surface and in the extracellular matrix of mammalian cells. In particular, Syndecans, a family of cell-surface transmembrane HSPGs, have been implicated in FGF signaling. The recent identification of Syndecan-3 and Syndecan-4 on quiescent and activated satellite cells in vivo and in vitro is supportive of a role for FGFs and/or HGF in the initial activation of quiescent satellite cells (72). Indeed, all components necessary for activation through these growth factors (i.e., receptors and HSPGs) are present on quiescent satellite cells. Moreover, in the same study, the use of sodium chlorate to reduce protein sulfation and soluble heparin to restore FGF but not HGF activity in freshly isolated satellite cells suggests that FGFs rather than HGF are more likely candidates for regulating the early event of satellite cell activation (72).

C. IGFs

The role of IGF-I and -II in regulating growth and development of various tissues has been known for many years. More recently, the paracrine/autocrine regulation of these hormones and their activity during skeletal muscle development and repair have become apparent. In vitro, IGF-I and IGF-II are able to alter MRFs expression and promote both the proliferation and the differentiation/fusion of myoblasts (5, 69, 89, 97, 108, 122, 203, 321). Increasing the levels of IGF-I within muscle cells by various in vivo and in vitro methods leads to increases in muscle mass due to an augmentation of muscle protein and DNA content (1, 26, 60, 69, 222). The hypertrophic effects of IGF-I are attributed to both the activation of satellite cell to proliferate providing more myonuclei and to the "true hypertrophy" (i.e., increase cytoplasmic-to-DNA volume ratio) through increased protein synthesis within existing myofibers (21, 27, 221, 277, 278).

IGF-I and IGF-II levels are upregulated in skeletal muscle undergoing regeneration (19, 95, 174, 192). IGF-I and IGF-II appear to ameliorate the aging muscle phenotype and the dystrophic muscle phenotype in *mdx* mice (25, 222, 286). As in normal muscle, this process is likely due to an effect of IGF on promoting satellite cell proliferation and differentiation as suggested by the hyperplasic (increased fiber number) and hypertrophic phenotype in transgenic *mdx* mice expressing mIGF-I (*mdx:mIGF*$^{+/+}$) (25). IGF-I may also improve muscle regeneration via promoting cell survival. IGF-I promotes myogenic cell survival in vitro, primarily through the phosphatidylinositol 3-kinase pathway and Akt activation (182, 183; reviewed in Ref. 3). Increased level of apoptosis has been observed in *mdx* muscles, but these levels are too low to allow any reliable comparison between *mdx* and *mdx:mIGF*$^{+/+}$ muscles (25, 262, 295). Increased levels of phosphorylated Akt in *mdx:mIGF*$^{+/+}$ suggest that the cell survival pathway is activated in this model and may contribute to the increased regenerative capacity (25). However, further work is necessary to confirm this hypothesis, since Akt is also involved in myogenic differentiation. IGFs may also be implicated in promoting reinnervation during muscle repair, since motor neurons also respond to these growth factors (56, 322). Thus, although the role for IGFs in promoting muscle repair is evident, the exact mechanisms through which these growth factors act remain to be defined. IGFs most certainly promote muscle repair by signaling to both the satellite cells and the myofibers. Whether distinct roles are played by different IGFs is possible, since IGF-II appears to be upregulated later during the process of muscle regeneration. Additional studies identifying the expression of IGF receptors and levels of IGF binding proteins (IGFBPs), which are known to inhibit IGFs functions, would be useful in understanding the role for these factors in muscle repair.

D. TGF-β Family

TGF-β growth factors are important cytokines regulating cell growth. TGF-β docks at a pair of receptors on the cell membrane which activate phosphorylation of SMAD proteins, resulting in their translocation to the nucleus where they trigger the activation of target genes, depending on the state of the cell (reviewed in Ref. 64). TGF-β family members have long been recognized as modulators of myoblast activity, inhibiting both proliferation and differentiation (4, 5, 122, 189, 190). The roles of TGF-β1, -β2, and -β3 during muscle regeneration are complex, involving myoblast fusion, regulation of an immune response, and motor neuron survival (reviewed in Ref. 207).

More recently, myostatin (MSTN) or growth and differentiation factor-8 (GDF-8) was identified in a screen for new members of the TGF-β family (208). The inhibitory effect of MSTN on muscle growth is demonstrated by the hypertrophic (increased fiber size) and hyperplasic (increased fiber number) muscle phenotype displayed by *MSTN*$^{-/-}$ mice (208). This property of MSTN was further demonstrated in several murine models (302, 360) and extended to other species by the identification of mutations within the *MSTN* gene in various European cattle breeds which exhibit a "double muscling" phenotype (123, 158, 160, 209). However, the mechanism of MSTN function remains unclear (reviewed in Ref. 279). A recent study suggests that MSTN signaling may be achieved by binding to activin type II receptors, particularly Act RIIB (186). In vitro studies of chicken myoblasts suggest that MSTN expression is highest during differentiation and fusion (170). Furthermore, recent in vitro experiments demonstrated that at high levels, MSTN inhibits proliferation and affects differentiation of C_2C_{12}, via cyclin-Cdk2 inactivation of retinoblastoma protein (Rb) (253, 254, 309, 313). In vivo studies analyzing MSTN expression in normal muscles suggest that endogenous MSTN expression is strongly associated with fast contracting muscles, which contain a low number of satellite cells, rather than muscle mass (52, 330). Furthermore, rodents subjected to hindlimb unloading, which suppresses satellite cell proliferation, demonstrate increased levels of MSTN (52). Taken together, these findings suggest that MSTN may regulate muscle mass by functioning as an inhibitor of satellite cell proliferation.

During muscle regeneration, MSTN appears to have a similar role as in muscle growth, although the number of studies is limited. First, some factor(s) capable of inducing MSTN expression in fibroblasts is present in regenerating skeletal muscle extract, suggesting that MSTN expression is regulated during muscle regeneration (346). Confirming this idea, several studies have shown temporal and spatial regulation of *MSTN* expression after muscle injury. Kirk et al. (168), using immunostaining after NTX injection, showed high levels of MSTN within necrotic fibers and connective tissues at a time when degeneration is high and satellite cell activation is low, supporting the concept that MSTN may act as an inhibitor of satellite cell proliferation while the degeneration process takes place. However, high levels of MSTN at this early time of muscle regeneration could also suggest that MSTN acts as a chemoattractant for phagocytes and inflammatory cells, a role that has been ascribed to the related TGF-β1. Later during the regenerating process, the same study showed that MSTN levels are low in the mononucleated cells located in regenerating areas where activated satellite cells are most abundant, and low in nascent and newly regenerated myotubes. Finally, MSTN levels rise back to normal in more mature muscle fiber. These findings are supported by a similar study performed on total protein and mRNA MSTN expression after NTX injection (211). However, in situ hybridization analysis performed

48 h after bupivacaine injection demonstrates high levels of MSTN within myogenic and nonmyogenic cells within the regenerating area (346). The discrepancy between MSTN mRNA and protein localization between these two studies may reflect differential activities in myotoxins used, or reflect the secretory properties of the molecule, or posttranslational regulation. MSTN proteins with different molecular masses have been isolated in different rat muscles (330), suggesting that posttranslational modifications of MSTN occur in different muscle types. In any case, more careful analyses of localizing MSTN mRNA and protein are necessary to identify MSTN role in muscle regeneration.

Further evidence for a role of MSTN during muscle regeneration comes from the analysis of murine models of muscular dystrophy. Indeed, MSTN mRNA is greatly reduced in limb muscles undergoing muscle regeneration such as in mice models for muscular dystrophy *mdx* and *Gsg*$^{-/-}$ (γ-sarcoglycan null mice) (316, 318, 360). The authors postulated that *MSTN* downregulation may play a role in the fiber hypertrophy observed following the degenerative process in the *mdx* muscle, which consequently maintains the muscle mass, leading to the functional rescue of *mdx* muscle. To support this hypothesis, a very recent study showed the improved muscle dystrophic phenotype in *mdx:MSTN*$^{-/-}$ mice (324). This function of MSTN could be adapted to alleviate symptoms associated with muscle mass loss during muscular dystrophies. In conclusion, the kinetics of MSTN expression during muscle regeneration support the view that MSTN may regulate muscle regeneration by acting as an inhibitor of myoblast proliferation and regulator of fiber growth similarly to its assigned function in muscle development. However, further experiments are required to determine whether MSTN acts directly and specifically on satellite cell regulation during regeneration.

E. IL-6 Family of Cytokines

Compelling in vitro and in vivo evidence supports a role for leukemia inhibitory factor (LIF) and IL-6 in muscle regeneration. In vitro, LIF stimulates myoblast growth via direct receptor-mediated mechanisms without affecting myoblast differentiation and fusion (17, 44, 294, 320). The in vitro effect of LIF on myoblast proliferation appears to involve activation of the JAK2-STAT3 signaling pathway (294). In vivo, administration of LIF to the site of muscle injury or in *mdx* muscle results in increased rate of muscle regeneration, characterized by increased myoblast proliferation and increased regenerated myofiber number and size (18, 22, 177, 178, 334). Furthermore, using a muscle crush model, Kurek et al. (178) showed that muscle regeneration in *LIF*$^{-/-}$ mice is significantly reduced compared with control littermates, and this deficiency is rescued by injection of LIF. After muscle injury, LIF transcripts have been shown to increase in both human and murine models (22, 159, 177, 250). LIF expression upon injury appears ubiquitous, with mRNA transcripts detected in resident nonmuscle cells, such as macrophages and Schwann cells, as well as in muscle cells (159, 179). Although IL-6 has homology to LIF and has similar expression pattern to LIF following muscle injury and similar in vitro effects on myoblasts, in vivo administration of IL-6 does not appear to affect the muscle repair process (159, 177, 178). Recently, Wallenius et al. (327) described IL-6-deficient mice that develop a mature-onset obesity, but no muscle phenotype has been described so far.

V. CONTRIBUTION OF OTHER STEM CELLS TO THE MUSCLE REPAIR PROCESS

Until recently, the muscle satellite cell was presumed to be the sole source of myonuclei in muscle repair (Fig. 5). However, recent findings have demonstrated the presence of multipotential stem cells in various adult tissues and challenged the widely held view that tissue-specific stem cells are predetermined to a specific tissue lineage. In fact, adult stem cells isolated from various tissues appear to differentiate in vitro and in vivo into multiple lineages depending on environmental cues. Progenitor cells isolated from bone marrow (BM) (37, 103, 132, 181), the adult musculature (16, 132, 151, 245, 317), the neuronal compartment (66, 112), and various mesenchymal tissues (350, 351) can differentiate into the myogenic lineage. In particular, BM and muscle adult stem cells have been shown to differentiate into muscle cells in vitro and to contribute to muscle regeneration in vivo (Fig. 5) (for review, see Refs. 119, 129, 274).

A. Nonmuscle Resident Stem Cells

A striking demonstration that nonmuscle stem cells participate in muscle regeneration was presented in seminal work by Ferrari et al. (103). In this study, donor BM from transgenic mice expressing the bacterial *lacZ* reporter gene under a muscle-specific promoter (ML3CF-*nlacZ* transgene) was intramuscularly or intravenously transplanted into severe combined immunodeficient *scid/bg* host mice (mouse strain combining characteristics of *scid* animals which lack functional B and T cells and *beige* animals which have intrinsically low natural killer cell activity) (103, 173). Donor-derived myogenic cells (LacZ+) were unambiguously identified within the host musculature following either type of transplants. However, the frequency at which this event occurred was very low compared with myoblast incorporation and necessitated the induction of extensive muscle regeneration

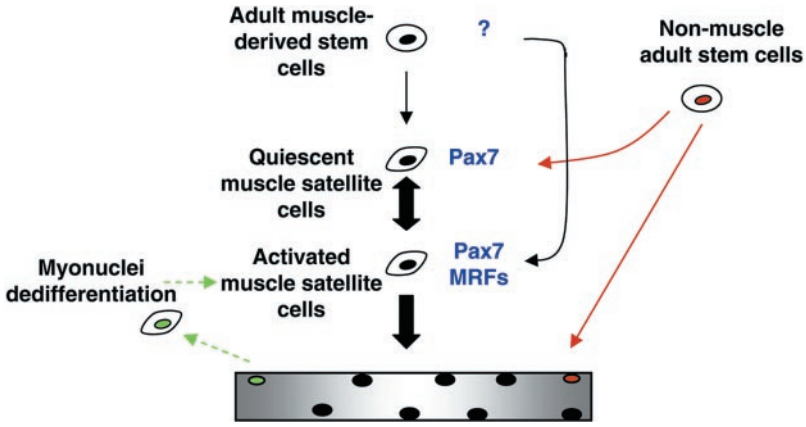

FIG. 5. Possible sources of myonuclei during skeletal muscle regeneration. Mammalian skeletal muscle regeneration involves the activation of the quiescent muscle satellite cell population to proliferate, differentiate, and fuse to provide new myonuclei for muscle repair (large black arrows). In this process, the transcription factor *Pax7* is required for muscle satellite cell specification/survival, whereas MRFs are essential in satellite cell proliferation and differentiation. Recently, in vitro and in vivo studies have demonstrated the presence of multipotential stem cells in adult muscles (adult muscle-derived stem cells) capable of myogenic commitment. Adult muscle-derived stem cells contribute to both muscle satellite cell pool and myonuclei, albeit at very low frequency (small black arrows). Similarly, stem cells capable of myogenic commitment can be isolated from other adult tissues (bone marrow stem cells, neuronal stem cells, and various mesenchymal stem cells) (red arrows). Finally, although dedifferentiation of myonuclei is the primary source of new myonuclei during amphibian regeneration, the ability of mammalian differentiated myonuclei to reenter the cell cycle in the course of muscle repair remains a hypothesis (green dashed arrows).

(103). Similarly, the rare incorporation of donor-derived BM cells into both heart and skeletal muscles of *mdx* mice was observed in other studies (37, 132). Gussoni et al. (132) demonstrated that myogenic activity was found in an enriched population of BM-derived stem cells called BM side population (SP), which contains hematopoietic stem cells. The isolation of a highly enriched population of SP cells by fluorescence-activated cell sorting (FACS) relies on the unique property of these cells to actively exclude dyes like Hoechst 33342 due to high expression of *mdr* (multidrug resistant) genes (120). These BM SP cells are Sca-1 (stem cell antigen-1)+, cKit+, CD43+, CD45+, linage marker (B220, Mac-1, Gr-1, CD4, CD5, CD8) −low/− and CD34− (132). Following tail vein injection, BM SP cells are capable of contributing to myofiber nuclei and restoring dystrophin expression to regenerating *mdx* muscle, albeit at similarly low levels since only 1–4% of total myofibers expressed dystrophin with 10–40% of which containing detectable fused donor-derived nuclei at 8–12 wk postinjection (132). Recently, LaBarge and Blau (181) unequivocally demonstrated that BM-derived cells not only contribute to regenerating myofibers but also to the muscle satellite cell pool. In this careful study, syngeneic mice received whole body irradiation followed by transplantation via tail vein injection of donor GFP(+) BM-derived cells. Two to six months after transplantation, GFP(+) cells expressing satellite cell markers were identified at the correct anatomical location for satellite cells. Moreover, clonal progenies of GFP(+) satellite cell isolated from recipient muscles expressed satellite cell markers underwent myogenic differentiation when exposed to low-mitogen media in vitro and contributed to new fiber formation when injected in tibialis anterior muscles of *scid* recipient mice. Together, these data demonstrate that following a BM transplant, BM-derived cells can adopt characteristics of muscle satellite cells. Furthermore, depletion of endogenous muscle satellite cell population by localized hindlimb irradiation potentiated the incorporation of GFP(+) BM-derived cells into the satellite cell compartment and exercise-induced muscle damage increased the frequency at which GFP(+) BM-derived cells contributed to new fiber formation (181). Thus, under certain conditions, the frequency of conversion of BM-derived cells to myogenic lineage can be increased.

These observations made in murine models recapitulate what is occurring in human muscular dystrophy patients as suggested by the study of a DMD patient who received a BM transplant at an early age (130). In this case study, fused donor nuclei were detected in the patient musculature. However, as in the mouse model, these did not appear to significantly increase the number of dystrophin-positive myofibers. These findings highlight the inefficient and slow incorporation of such BM-derived cells into the musculature, compared with myoblasts, making them at present an ineffective therapeutic intervention (103, 143, 332). It appears therefore that further studies are necessary to establish the optimal cellular and environmental conditions to promote recruitment and myogenic conversion of nonmuscle stem cells for therapeutic use. Moreover, these findings suggest a minor role, if any, for nonmuscle stem cells in normal skeletal muscle repair.

B. Muscle Resident Stem Cells

Similarly to BM, an enriched population of adult stem cells can be isolated from skeletal muscles by FACS analysis on the basis of Hoechst 33342 staining (16, 132). This muscle stem cell population, also called muscle SP (mSP), is capable of reconstituting the entire hematopoietic repertoire after intravenous injection into lethally irradiated mice, albeit less effectively than BM SP cells (132). More importantly for this review, mSP can commit to myogenic conversion in vivo, as demonstrated by the presence of 3–9% of dystrophin-expressing myofibers, of which 3–9% contain donor-derived myonuclei, following intravenous injections (132). The presence of mSP in *Pax7*$^{-/-}$ musculature, which lack satellite cells, suggests that mSP cells and satellite cells represent distinct cellular populations (275). This hypothesis is substantiated by the observation that mSP do not express the satellite cell markers *Myf5-nlacZ*, *Pax7*, or *desmin* (16). Furthermore, the increased propensity of *Pax7*$^{-/-}$ mSP cells to form hematopoeitic colonies in vitro and the competence of normal mSP to give rise to satellite cells within the recipient muscle suggest that mSP cells may represent satellite cell progenitors (16, 132, 275). The in vitro conversion of mSP to the myogenic lineage requires coculture with myoblasts, suggesting that the process is subject to regulation via cell-mediated inductive interactions (16). Thus there is strong evidence for the presence of progenitor cells with myogenic potential other than satellite cells within skeletal muscles. Although mSP represent a separate cellular population than satellite cell population, they may represent satellite cell progenitors and/or myogenic progenitors capable of direct myogenic fusion (Fig. 5).

The SP phenotype results from the efficient efflux of Hoechst dye and therefore provides a mixed cellular population with little information on cellular characteristics or origin. Therefore, it is becoming increasingly important to characterize muscle-derived stem cells on the basis of cell surface markers. Of interest are those that have been previously used to define murine BM SP, primarily Sca-1, CD34, and CD45. Most of the mSP cells (92%) express Sca-1, whereas only 16% express CD45 consisting of both Sca-1+ (9.2%) and Sca-1− (6.8%) (16, 132). CD45+, but not CD45−, cells are responsible for the majority of the in vivo and in vitro hematopoietic activity derived from murine muscle (16, 206). A fraction of both CD45+ (9%) and CD45− (5%) mSP cells undergo myogenic conversion when cocultured with myoblasts in vitro (16). However, myogenic potential appears greater in CD45− than CD45+ muscle-derived cells after intramuscular injection (206). The myogenic conversion of Sca-1+/CD45+ cells, a population generally associated with hematopoietic differentiation potential, suggests that reversal of fate determination or transdifferentiation is possible albeit at a low rate.

Another interesting cell population is the Sca-1+/CD34+ population isolated from murine muscle (151, 245, 317). CD34 is a sialylated transmembrane glycoprotein that is expressed in myeloid progenitors and endothelial cells (101, 172, 220). A muscle culture system of successive preplating combined with FACS sorting was shown to facilitate the enrichment and purification of Sca-1+/CD34+ cells of both myogenic and hematopoietic differentiation potential (317). When injected into the arterial circulation of the hindlimb of *mdx* mice, the late-passage (late-adhering) Sca-1+/CD34+ cells adhere to the endothelium (317). Moreover, after muscle regeneration involving vascular damage, Sca-1+/CD34+ cells migrate from blood vessels to incorporate into regenerating myofibers at a relatively high percentage (12% of the fibers) (317). These data suggest that bipotential stem cells present within the vasculature may be activated to enter the myogenic lineage when the muscle receives extensive damage. However, the data do not preclude the possibility of the muscle-derived Sca-1+/CD34+ cells representing progenitors of satellite cells. Another population of highly proliferative, late-adhering, Sca-1+/CD34+ cells (MDSC) obtained from preplating enrichment technique also appear to have myogenic potential and high regenerative capacity in vivo (245). MDSC have unique characteristics usually associated with noncommitted progenitor cells such as high self-renewal capacity, high proliferation capacity, and multipotential capacity (245). However, MDSC are c-Kit−/CD45− eliminating their potential hematopoietic origin. Moreover, several observations suggest that they are progenitors of satellite cells. First, the MDSC have a high potential for myogenic conversion in vitro and in vivo. Second, they can spontaneously express myogenic markers, MyoD and desmin. Finally, they have similar phenotypic characteristics (Sca-1+/m-cadherin−) to a subpopulation of cells that have been identified as satellite cells in situ (245). Another in vivo study confirmed the enhanced regenerative capacity of a CD34+, late-adhering population of muscle-derived stem cells compared with the CD34− cell population (151). Thus it appears that CD34 plays an important role in identifying myogenic progenitors. However, the identification of a subpopulation of mouse satellite cells expressing CD34 suggests that CD34 antigen is also expressed in myogenic cells and therefore highlights the need for multiple markers for identification of muscle-derived stem cells other than satellite cells (28, 185).

The data available to date support the notion that muscle-derived progenitors, other than muscle satellite cells, are capable of myogenic differentiation and of integration into the regenerating musculature in vivo. Understanding the signaling and molecular pathways involved in this commitment is becoming an important question in the fields of muscle regeneration and cellular biology. Further studies are required to determine whether multiple types of muscle-derived stem cells exist and whether

they display differential ability to regenerate muscle. To this aim, rigorous phenotypic characterizations of donor population including clonal analyses before transplanting will be necessary.

VI. CONTRIBUTION OF DEGENERATING FIBER NUCLEI TO NEW MYOFIBER FORMATION

The muscle degeneration process is commonly thought to induce proteolysis and degradation of the sarcoplasmic contents and DNA of the injured fibers or the injured segment of the fiber. The newly regenerated fibers are produced by the differentiation and fusion of activated satellite cells and possibly other mononuclear stem cells as discussed earlier. However, another possibility is the "recycling" of myonuclei, as suggested by early electron microscopic observations of sequestered myonuclei during regeneration (discussed in Ref. 129). The subsequent demonstration that such events are critical during amphibian regeneration combined with more recent reports that mammalian myonuclei are capable of reverting their differentiated state have reinstated this possibility.

A. The Amphibian Versus Mammalian Regenerative Process

It has long been established that in urodele amphibians, such as the newt, the repair process in response to tissue injury or amputation is highly competent, since it can regenerate entire adult limbs and various other structures (reviewed in Ref. 45). However, the more intriguing aspect of this regenerative phenomenon is that it involves dedifferentiation of postmitotic cells, which reenter the cell cycle to proliferate and differentiate, generating new tissues without the necessary activation of a "reserve" stem cell (176, 195). Newt myonuclei cell-cycle reentry is induced by phosphorylation of Rb in the presence of thrombin and serum (305). Using in vivo fluorescent labeling of single muscle fibers, Echeverri et al. (94) recently demonstrated the in vivo dedifferentiation of urodele muscle upon amputation. The same study estimated that up to 29% of nondermal-derived cells in the blastema (or growth zone) come from dedifferentiation of mature muscle fibers. Thus it is generally accepted that, in these organisms, the cellular terminal differentiation is reversible when appropriate signals are present within the tissue. Moreover, mononucleated cells generated by dedifferentiation can contribute to the blastema and the regenerative process (reviewed in Ref. 45).

In comparison, mammalian regenerative capacity is more limited resulting in significant scar tissue formation. This discrepancy in regenerative capacity between the two systems has been mainly attributed to the lack of the cellular machinery necessary to undergo a similar dedifferentiation process within mammalian cells. Indeed, contrary to newt muscle, mouse myocytes were shown to be incapable of cell cycle reentry when triggered by the active newt serum-derived factor (305). However, this finding has been challenged by a recent study demonstrating the presence of an active molecule (most likely a protein or a protein complex) within a crude newt limb regeneration extract which induces in vitro dedifferentiation of C_2C_{12} myotubes (204).

B. In Vitro Mammalian Cell Dedifferentiation

The first report challenging mammalian myonuclei dedifferentiation came from an in vitro study on a small microtubule-binding molecule called myoseverin (257). When exposed to myoseverin, C_2C_{12} multinucleated myotubes underwent fission to form viable mononucleated fragments capable of DNA synthesis and proliferation after removal of the molecule and exposure to growth medium (239, 257). Expression profile analysis suggests that myoseverin does not affect most cell-cycle specific transcripts but rather induces changes in the expression of genes normally associated with tissue injury such as growth factors and genes involved in extracellular matrix remodeling. Myoseverin did not downregulate the expression of MRFs, suggesting that commitment of the cells to the myogenic lineage was not affected. Overall, these data suggest that disassembly of the cytoskeleton by myoseverin promotes mammalian myotube cytokinesis and myonuclei cell cycle reentry possibly resulting in the reversal of terminal differentiation of murine muscle.

In the same year, another in vitro study demonstrated the capacity of C_2C_{12} myotubes to reenter the cell cycle using ectopic expression of the homeobox *Msx1* transcription factor (227). Using C_2C_{12} combined with a conditional tetracycline promoter regulating *Msx1* cDNA expression, Odelberg et al. (227) confirmed previous findings that exogenous expression of *Msx1* inhibits muscle terminal differentiation (314). The failure of myoblasts to differentiate under low serum conditions was attributed to the reduced level of nuclear protein MyoD (314). Furthermore, overexpression of *Msx1* in terminally differentiated myotubes resulted in the downregulation of MyoD, myogenin, and MRF4 and the cell cycle inhibitor p21 in 20–50% of the myotubes (227). Importantly, a low number of myotubes underwent cytokinesis, producing smaller multinucleated myotubes and proliferating mononucleated cells. Clonal analysis of the newly derived mononucleated cells demonstrated their ability to redifferentiate into cells expressing chondrogenic, adipogenic, myogenic, and osteogenic markers under appropriate culture conditions. Overall, these findings suggest that following appropriate in vitro stimulation mammalian myotubes are capable of dedifferentiation.

C. Role of *Msx* Genes in Mammalian Muscle Regeneration

The role for *Msx* genes during mammalian muscle regeneration is suggested by the observation that *Msx1* appears to be involved in the repression of muscle differentiation in chick and mouse muscle progenitor cells during migration of these cells to the limb bud. This inhibition seems to involve direct protein-protein interaction of *Msx1* with *Pax3* (31). In both chick and mouse migrating limb muscle precursors, expression of *Msx1* and *Pax3* overlaps (31, 145). In addition, *Msx1* suppresses *MyoD* gene expression in vitro through the binding of *Msx1* to the regulatory region of *MyoD* gene (338). Ectopic expression of *Msx1* in the forelimb and somites of chicken embryos inhibits *MyoD* expression and muscle differentiation (31). As well as playing an important role during mammalian muscle development, *Msx1* expression correlates with regenerative processes in various systems such as in the digit tip of newborn and fetal mice (251), in the blastema of urodele limb (53, 80, 171, 284), and zebrafish fin (243). *Msx1* transcripts were also detected in a PCR-based analysis of isolated murine satellite cells (73). Moreover, *Msx1* gene expression is upregulated by FGFs (47, 215, 243), a factor important in mammalian skeletal muscle regeneration. Overall, these data are suggestive of a role for *Msx1* in mammalian muscle regeneration. Whether this role is to prevent myogenic differentiation to allow expansion of a myogenic or stem cell pool or whether it is in the induction of myonuclei dedifferentiation remains to be determined.

D. Conclusion

Overall, these data suggest that myogenic mammalian cells are capable of dedifferentiation at least under tissue culture conditions and that *Msx1* and myoseverin can contribute to this process. It raises the old question of whether myonuclei dedifferentiation during mammalian muscle regeneration is a source of mononuclear cells for new fiber formation (Fig. 5). The development of new techniques enabling the in vivo labeling of individual myofibers may one day allow researchers to identify this process in vivo. Nevertheless, understanding the signaling events leading to the dedifferentiation of myotubes may lead to the development of new therapeutic strategies in the context of mammalian muscle regeneration.

VII. PERSPECTIVES

The muscle satellite cell population is the principal cellular component of mammalian skeletal muscle regeneration. Although compelling evidence suggests satellite cells are stem cells capable of self-renewal, this is yet to be directly addressed. Advances in identifying satellite cell molecular markers will prove useful in further characterizing this cell population. Non-muscle-derived and muscle-derived stem cells, other than satellite cells, are capable of myogenic differentiation and integrate into the regenerating musculature in vivo. However, the relationship between these stem cell populations and satellite cells has not been defined. Understanding the signaling and molecular pathways involved in recruitment and myogenic commitment of these progenitors is an important question in the fields of muscle regeneration and cellular biology. Furthermore, by focusing on donor population with enhanced regeneration abilities, advances in clinical cellular therapies for muscular dystrophies should be observed in a near future.

We thank Atsushi Asakura, Josée Coulombe, Jonathan Schertzer, Patrick Seale, and Dave Wilkinson for critical reading of the manuscript and Jeff Ishibashi, Claire Palmer, and Patrick Seale for help with figures.

M. A. Rudnicki holds the Canada Research Chair in Molecular Genetics and is a Howard Hughes Medical Institute International Scholar. This work was supported by grants from the Muscular Dystrophy Association, the National Institutes of Health, the Canadian Institutes of Health Research, and the Canada Research Chair Program (to M. A. Rudnicki).

Address for reprint requests and other correspondence: M. A. Rudnicki, Ottawa Health Research Institute, 501 Smyth Rd., Ottawa, Ontario K1H 8L6, Canada (E-mail: mrudnicki@ohri.ca).

REFERENCES

1. **Adams GR and McCue SA.** Localized infusion of IGF-I results in skeletal muscle hypertrophy in rats. *J Appl Physiol* 84: 1716–1722, 1998.
2. **Alderton JM and Steinhardt RA.** How calcium influx through calcium leak channels is responsible for the elevated levels of calcium-dependent proteolysis in dystrophic myotubes. *Trends Cardiovasc Med* 10: 268–272, 2000.
3. **Alessi DR and Cohen P.** Mechanism of activation and function of protein kinase B. *Curr Opin Genet Dev* 8: 55–62, 1998.
4. **Allen RE and Boxhorn LK.** Inhibition of skeletal muscle satellite cell differentiation by transforming growth factor-beta. *J Cell Physiol* 133: 567–572, 1987.
5. **Allen RE and Boxhorn LK.** Regulation of skeletal muscle satellite cell proliferation and differentiation by transforming growth factor-beta, insulin-like growth factor I, and fibroblast growth factor. *J Cell Physiol* 138: 311–315, 1989.
6. **Allen RE, Sheehan SM, Taylor RG, Kendall TL, and Rice GM.** Hepatocyte growth factor activates quiescent skeletal muscle satellite cells in vitro. *J Cell Physiol* 165: 307–312, 1995.
7. **Almekinders LC and Gilbert JA.** Healing of experimental muscle strains and the effects of nonsteroidal anti-inflammatory medication. *Am J Sports Med* 14: 303–308, 1986.
8. **Anastasi S, Giordano S, Sthandier O, Gambarotta G, Maione R, Comoglio P, and Amati P.** A natural hepatocyte growth factor/scatter factor autocrine loop in myoblast cells and the effect of the constitutive Met kinase activation on myogenic differentiation. *J Cell Biol* 137: 1057–1068, 1997.
9. **Anderson JE.** A role for nitric oxide in muscle repair: nitric oxide-mediated activation of muscle satellite cells. *Mol Biol Cell* 11: 1859–1874, 2000.
10. **Anderson JE, Mitchell CM, McGeachie JK, and Grounds MD.** The time course of basic fibroblast growth factor expression in

crush-injured skeletal muscles of SJL/J and BALB/c mice. *Exp Cell Res* 216: 325–334, 1995.
11. **Armand O, Boutineau AM, Mauger A, Pautou MP, and Kieny M.** Origin of satellite cells in avian skeletal muscles. *Arch Anat Microsc Morphol Exp* 72: 163–181, 1983.
12. **Armstrong RB.** Initial events in exercise-induced muscular injury. *Med Sci Sports Exercise* 22: 429–435, 1990.
13. **Armstrong RB, Warren GL, and Warren JA.** Mechanisms of exercise-induced muscle fibre injury. *Sports Med* 12: 184–207, 1991.
14. **Asakura A, Komaki M, and Rudnicki M.** Muscle satellite cells are multipotential stem cells that exhibit myogenic, osteogenic, and adipogenic differentiation. *Differentiation* 68: 245–253, 2001.
15. **Asakura A and Rudnicki MA.** Cellular and molecular mechanisms regulating skeletal muscle development. In: *Mouse Development*. Orlando, FL: Academic, 2002, p. 253–278.
16. **Asakura A, Seale P, Girgis-Gabardo A, and Rudnicki MA.** Myogenic specification of side population cells in skeletal muscle. *J Cell Biol* 159: 123–134, 2002.
17. **Austin L, Bower J, Kurek J, and Vakakis N.** Effects of leukaemia inhibitory factor and other cytokines on murine and human myoblast proliferation. *J Neurol Sci* 112: 185–191, 1992.
18. **Austin L, Bower JJ, Bennett TM, Lynch GS, Kapsa R, White JD, Barnard W, Gregorevic P, and Byrne E.** Leukemia inhibitory factor ameliorates muscle fiber degeneration in the mdx mouse. *Muscle Nerve* 23: 1700–1705, 2000.
19. **Bakay M, Zhao P, Chen J, and Hoffman EP.** A web-accessible complete transcriptome of normal human and DMD muscle. *Neuromuscul Disorders* 12 *Suppl* 1: S125–S141, 2002.
20. **Balcerzak D, Poussard S, Brustis JJ, Elamrani N, Soriano M, Cottin P, and Ducastaing A.** An antisense oligodeoxyribonucleotide to m-calpain mRNA inhibits myoblast fusion. *J Cell Sci* 108: 2077–2082, 1995.
21. **Bark TH, McNurlan MA, Lang CH, and Garlick PJ.** Increased protein synthesis after acute IGF-I or insulin infusion is localized to muscle in mice. *Am J Physiol Endocrinol Metab* 275: E118–E123, 1998.
22. **Barnard W, Bower J, Brown MA, Murphy M, and Austin L.** Leukemia inhibitory factor (LIF) infusion stimulates skeletal muscle regeneration after injury: injured muscle expresses lif mRNA. *J Neurol Sci* 123: 108–113, 1994.
23. **Baroffio A, Bochaton-Piallat ML, Gabbiani G, and Bader CR.** Heterogeneity in the progeny of single human muscle satellite cells. *Differentiation* 59: 259–268, 1995.
24. **Baroffio A, Hamann M, Bernheim L, Bochaton-Piallat ML, Gabbiani G, and Bader CR.** Identification of self-renewing myoblasts in the progeny of single human muscle satellite cells. *Differentiation* 60: 47–57, 1996.
25. **Barton ER, Morris L, Musaro A, Rosenthal N, and Sweeney HL.** Muscle-specific expression of insulin-like growth factor I counters muscle decline in mdx mice. *J Cell Biol* 157: 137–148, 2002.
26. **Barton-Davis ER, Shoturma DI, Musaro A, Rosenthal N, and Sweeney HL.** Viral mediated expression of insulin-like growth factor I blocks the aging-related loss of skeletal muscle function. *Proc Natl Acad Sci USA* 95: 15603–15607, 1998.
27. **Barton-Davis ER, Shoturma DI, and Sweeney HL.** Contribution of satellite cells to IGF-I induced hypertrophy of skeletal muscle. *Acta Physiol Scand* 167: 301–305, 1999.
28. **Beauchamp JR, Heslop L, Yu DS, Tajbakhsh S, Kelly RG, Wernig A, Buckingham ME, Partridge TA, and Zammit PS.** Expression of CD34 and Myf5 defines the majority of quiescent adult skeletal muscle satellite cells. *J Cell Biol* 151: 1221–1234, 2000.
29. **Beauchamp JR, Morgan JE, Pagel CN, and Partridge TA.** Dynamics of myoblast transplantation reveal a discrete minority of precursors with stem cell-like properties as the myogenic source. *J Cell Biol* 144: 1113–1122, 1999.
30. **Belcastro AN, Shewchuk LD, and Raj DA.** Exercise-induced muscle injury: a calpain hypothesis. *Mol Cell Biochem* 179: 135–145, 1998.
31. **Bendall AJ, Ding J, Hu G, Shen MM, and Abate-Shen C.** Msx1 antagonizes the myogenic activity of Pax3 in migrating limb muscle precursors. *Development* 126: 4965–4976, 1999.
32. **Bhagwati S, Ghatpande A, Shafiq SA, and Leung B.** In situ hybridization analysis for expression of myogenic regulatory factors in regenerating muscle of mdx mouse. *J Neuropath Exp Neurol* 55: 509–514, 1996.
33. **Bischoff R.** Proliferation of muscle satellite cells on intact myofibers in culture. *Dev Biol* 115: 129–139, 1986.
34. **Bischoff R.** A satellite cell mitogen from crushed adult muscle. *Dev Biol* 115: 140–147, 1986.
35. **Bischoff R.** The satellite cell and muscle regneration. In: *Myology*. New York: McGraw-Hill, 1994, p. 97–118.
36. **Bischoff R.** Chemotaxis of skeletal muscle satellite cells. *Dev Dyn* 208: 505–515, 1997.
37. **Bittner RE, Schofer C, Weipoltshammer K, Ivanova S, Streubel B, Hauser E, Freilinger M, Hoger H, Elbe-Burger A, and Wachtler F.** Recruitment of bone-marrow-derived cells by skeletal and cardiac muscle in adult dystrophic mdx mice. *Anat Embryol* 199: 391–396, 1999.
38. **Blaivas M and Carlson BM.** Muscle fiber branching—difference between grafts in old and young rats. *Mech Ageing Dev* 60: 43–53, 1991.
39. **Blake DJ, Weir A, Newey SE, and Davies KE.** Function and genetics of dystrophin and dystrophin-related proteins in muscle. *Physiol Rev* 82: 291–329, 2002.
40. **Blau HM, Pavlath GK, Hardeman EC, Chiu CP, Silberstein L, Webster SG, Miller SC, and Webster C.** Plasticity of the differentiated state. *Science* 230: 758–766, 1985.
41. **Blaveri K, Heslop L, Yu DS, Rosenblatt JD, Gross JG, Partridge TA, and Morgan JE.** Patterns of repair of dystrophic mouse muscle: studies on isolated fibers. *Dev Dyn* 216: 244–256, 1999.
42. **Bockhold KJ, Rosenblatt JD, and Partridge TA.** Aging normal and dystrophic mouse muscle: analysis of myogenicity in cultures of living single fibers. *Muscle Nerve* 21: 173–183, 1998.
43. **Bourke DL and Ontell M.** Branched myofibers in long-term whole muscle transplants: a quantitative study. *Anat Rec* 209: 281–288, 1984.
44. **Bower J, Vakakis N, Nicola NA, and Austin L.** Specific binding of leukemia inhibitory factor to murine myoblasts in culture. *J Cell Physiol* 164: 93–98, 1995.
45. **Brockes JP and Kumar A.** Plasticity and reprogramming of differentiated cells in amphibian regeneration. *Nat Rev Mol Cell Biol* 3: 566–574, 2002.
46. **Brussee V, Tardif F, and Tremblay JP.** Muscle fibers of mdx mice are more vulnerable to exercise than those of normal mice. *Neuromuscular Disorders* 7: 487–492, 1997.
47. **Bushdid PB, Chen CL, Brantley DM, Yull F, Raghow R, Kerr LD, and Barnett JV.** NF-kappaB mediates FGF signal regulation of msx-1 expression. *Dev Biol* 237: 107–115, 2001.
48. **Campion DR.** The muscle satellite cell: a review. *Int Rev Cytol* 87: 225–251, 1984.
49. **Campion DR, Richardson RL, Reagan JO, and Kraeling RR.** Changes in the satellite cell population during postnatal growth of pig skeletal muscle. *J Anim Sci* 52: 1014–1018, 1981.
50. **Carlson BM and Faulkner JA.** Muscle transplantation between young and old rats: age of host determines recovery. *Am J Physiol Cell Physiol* 256: C1262–C1266, 1989.
51. **Carlson BM and Gutmann E.** Regneration in free grafts of normal and denervated muscles in the rat: morphology and histochemistry. *Anat Rec* 183: 47–62, 1975.
52. **Carlson CJ, Booth FW, and Gordon SE.** Skeletal muscle myostatin mRNA expression is fiber-type specific and increases during hindlimb unloading. *Am J Physiol Regul Integr Comp Physiol* 277: R601–R606, 1999.
53. **Carlson MR, Bryant SV, and Gardiner DM.** Expression of Msx-2 during development, regeneration, and wound healing in axolotl limbs. *J Exp Zool* 282: 715–723, 1998.
54. **Carmena A, Murugasu-Oei B, Menon D, Jimenez F, and Chia W.** Inscuteable and numb mediate asymmetric muscle progenitor cell divisions during *Drosophila* myogenesis. *Genes Dev* 12: 304–315, 1998.
55. **Carnwath JW and Shotton DM.** Muscular dystrophy in the mdx mouse: histopathology of the soleus and extensor digitorum longus muscles. *J Neurol Sci* 80: 39–54, 1987.
56. **Caroni P and Grandes P.** Nerve sprouting in innervated adult

skeletal muscle induced by exposure to elevated levels of insulin-like growth factors. *J Cell Biol* 110: 1307–1317, 1990.
57. **Carpenter S and Karpati G.** Major general pathological reactions and their consequences on skeletal muscle cells. In: *Pathology of Skeletal Muscle*. London: Churchill Livingstone, 1984, p. 84–101.
58. **Cayouette M and Raff M.** Asymmetric segregation of Numb: a mechanism for neural specification from *Drosophila* to mammals. *Nat Neurosci* 5: 1265–1269, 2002.
59. **Chakravarthy MV, Abraha TW, Schwartz RJ, Fiorotto ML, and Booth FW.** Insulin-like growth factor-I extends in vitro replicative life span of skeletal muscle satellite cells by enhancing G_1/S cell cycle progression via the activation of phosphatidylinositol 3′-kinase/Akt signaling pathway. *J Biol Chem* 275: 35942–35952, 2000.
60. **Chakravarthy MV, Davis BS, and Booth FW.** IGF-I restores satellite cell proliferative potential in immobilized old skeletal muscle. *J Appl Physiol* 89: 1365–1379, 2000.
61. **Charge SB, Brack AS, and Hughes SM.** Aging-related satellite cell differentiation defect occurs prematurely after Ski-induced muscle hypertrophy. *Am J Physiol Cell Physiol* 283: C1228–C1241, 2002.
62. **Charlton CA, Mohler WA, Radice GL, Hynes RO, and Blau HM.** Fusion competence of myoblasts rendered genetically null for N-cadherin in culture. *J Cell Biol* 138: 331–336, 1997.
63. **Chen G and Quinn LS.** Partial characterization of skeletal myoblast mitogens in mouse crushed muscle extract. *J Cell Physiol* 153: 563–574, 1992.
64. **Chen W, Fu X, and Sheng Z.** Review of current progress in the structure and function of Smad proteins. *Chin Med J* 115: 446–450, 2002.
65. **Chi N and Epstein JA.** Getting your Pax straight: Pax proteins in development and disease. *Trends Genet* 18: 41–47, 2002.
66. **Clarke DL, Johansson CB, Wilbertz J, Veress B, Nilsson E, Karlstrom H, Lendahl U, and Frisen J.** Generalized potential of adult neural stem cells. *Science* 288: 1660–1663, 2000.
67. **Cohen ME, Yin M, Paznekas WA, Schertzer M, Wood S, and Jabs EW.** Human SLUG gene organization, expression, and chromosome map location on 8q. *Genomics* 51: 468–471, 1998.
68. **Cohn R, Henry M, Michele D, Barresi R, Saito F, Moore S, Flanagan J, Skwarchuk M, Robbins M, Mendell J, Williamson R, and Campbell K.** Disruption of dag1 in differentiated skeletal muscle reveals a role for dystroglycan in muscle regeneration. *Cell* 110: 639, 2002.
69. **Coleman ME, Demayo F, Yin KC, Lee HM, Geske R, Montgomery C, and Schwartz RJ.** Myogenic vector expression of insulin-like growth factor I stimulates muscle cell differentiation and myofiber hypertrophy in transgenic mice. *J Biol Chem* 270: 12109–12116, 1995.
70. **Conboy IM and Rando TA.** The regulation of notch signaling controls satellite cell activation and cell fate determination in postnatal myogenesis. *Dev Cell* 3: 397–409, 2002.
71. **Cooper RN, Tajbakhsh S, Mouly V, Cossu G, Buckingham M, and Butler-Browne GS.** In vivo satellite cell activation via Myf5 and MyoD in regenerating mouse skeletal muscle. *J Cell Sci* 112: 2895–2901, 1999.
72. **Cornelison DD, Filla MS, Stanley HM, Rapraeger AC, and Olwin BB.** Syndecan-3 and syndecan-4 specifically mark skeletal muscle satellite cells and are implicated in satellite cell maintenance and muscle regeneration. *Dev Biol* 239: 79–94, 2001.
73. **Cornelison DD, Olwin BB, Rudnicki MA, and Wold BJ.** MyoD(−/−) satellite cells in single-fiber culture are differentiation defective and MRF4 deficient. *Dev Biol* 224: 122–137, 2000.
74. **Cornelison DDW and Wold BJ.** Single-cell analysis of regulatory gene expression in quiescent and activated mouse skeletal muscle satellite cells. *Dev Biol* 191: 270–283, 1997.
75. **Cossu G, Cicinelli P, Fieri C, Coletta M, and Molinaro M.** Emergence of TPA-resistant "satellite" cells during muscle histogenesis of human limb. *Exp Cell Res* 160: 403–411, 1985.
76. **Cossu G and Molinaro M.** Cell heterogeneity in the myogenic lineage. *Curr Top Dev Biol* 23: 185–208, 1987.
77. **Cossu G, Molinaro M, and Pacifici M.** Differential response of satellite cells and embryonic myoblasts to a tumor promoter. *Dev Biol* 98: 520–524, 1983.
78. **Cottin P, Brustis JJ, Poussard S, Elamrani N, Broncard S, and Ducastaing A.** Ca(2+)-dependent proteinases (calpains) and muscle cell differentiation. *Biochim Biophys Acta* 1223: 170–178, 1994.
79. **Coulton GR, Morgan JE, Partridge TA, and Sloper JC.** The mdx mouse skeletal muscle myopathy. I. A histological, morphometric and biochemical investigation. *Neuropathol Appl Neurobiol* 14: 53–70, 1988.
80. **Crews L, Gates PB, Brown R, Joliot A, Foley C, Brockes JP, and Gann AA.** Expression and activity of the newt Msx-1 gene in relation to limb regeneration. *Proc R Soc Lond B Biol Sci* 259: 161–171, 1995.
81. **D'Albis A, Couteaux R, Janmot C, Roulet A, and Mira JC.** Regeneration after cardiotoxin injury of innervated and denervated slow and fast muscles of mammals. Myosin isoform analysis. *Eur J Biochem* 174: 103–110, 1988.
82. **Darr KC and Schultz E.** Exercise-induced satellite cell activation in growing and mature skeletal muscle. *J Appl Physiol* 63: 1816–1821, 1987.
83. **De Angelis L, Berghella L, Coletta M, Lattanzi L, Zanchi M, Cusella-De Angelis MG, Ponzetto C, and Cossu G.** Skeletal myogenic progenitors originating from embryonic dorsal aorta coexpress endothelial and myogenic markers and contribute to postnatal muscle growth and regeneration. *J Cell Biol* 147: 869–878, 1999.
84. **Decary S, Hamida CB, Mouly V, Barbet JP, Hentati F, and Butler-Browne GS.** Shorter telomeres in dystrophic muscle consistent with extensive regeneration in young children. *Neuromuscular Disorders* 10: 113–120, 2000.
85. **Decary S, Mouly V, Hamida CB, Sautet A, Barbet JP, and Butler-Browne GS.** Replicative potential and telomere length in human skeletal muscle: implications for satellite cell-mediated gene therapy. *Hum Gene Ther* 8: 1429–1438, 1997.
86. **Dedieu S, Dourdin N, Dargelos E, Poussard S, Veschambre P, Cottin P, and Brustis JJ.** Calpain and myogenesis: development of a convenient cell culture model. *Biol Cell* 94: 65–76, 2002.
87. **Delapeyriere O, Ollendorff V, Planche J, Ott MO, Pizette S, Coulier F, and Birnbaum D.** Expression of the Fgf6 gene is restricted to developing skeletal muscle in the mouse embryo. *Development* 118: 601–611, 1993.
88. **Delfini M, Hirsinger E, Pourquie O, and Duprez D.** Delta 1-activated notch inhibits muscle differentiation without affecting Myf5 and Pax3 expression in chick limb myogenesis. *Development* 127: 5213–5224, 2000.
89. **Doumit ME, Cook DR, and Merkel RA.** Fibroblast growth factor, epidermal growth factor, insulin-like growth factors, and platelet-derived growth factor-BB stimulate proliferation of clonally derived porcine myogenic satellite cells. *J Cell Physiol* 157: 326–332, 1993.
90. **Dourdin N, Balcerzak D, Brustis JJ, Poussard S, Cottin P, and Ducastaing A.** Potential m-calpain substrates during myoblast fusion. *Exp Cell Res* 246: 433–442, 1999.
91. **Dulor JP, Cambon B, Vigneron P, Reyne Y, Nougues J, Casteilla L, and Bacou F.** Expression of specific white adipose tissue genes in denervation-induced skeletal muscle fatty degeneration. *FEBS Lett* 439: 89–92, 1998.
92. **Dusterhoft S and Pette D.** Evidence that acidic fibroblast growth factor promotes maturation of rat satellite-cell-derived myotubes in vitro. *Differentiation* 65: 161–169, 1999.
93. **Dworak HA and Sink H.** Myoblast fusion in *Drosophila*. *Bioessays* 24: 591–601, 2002.
94. **Echeverri K, Clarke JD, and Tanaka EM.** In vivo imaging indicates muscle fiber dedifferentiation is a major contributor to the regenerating tail blastema. *Dev Biol* 236: 151–164, 2001.
95. **Edwall D, Schalling M, Jennische E, and Norstedt G.** Induction of insulin-like growth factor I messenger ribonucleic acid during regeneration of rat skeletal muscle. *Endocrinology* 124: 820–825, 1989.
96. **El Fahime E, Mills P, Lafreniere JF, Torrente Y, and Tremblay JP.** The urokinase plasminogen activator: an interesting way to improve myoblast migration following their transplantation. *Exp Cell Res* 280: 169–178, 2002.
97. **Engert JC, Berglund EB, and Rosenthal N.** Proliferation pre-

cedes differentiation in IGF-I-stimulated myogenesis. *J Cell Biol* 135: 431–440, 1996.
98. **Faulkner JA, Brooks SV, and Opiteck JA.** Injury to skeletal muscle fibers during contractions: conditions of occurrence and prevention. *Phys Ther* 73: 911–921, 1993.
99. **Fehr HG, Lotzerich H, and Michna H.** The influence of physical exercise on peritoneal macrophage functions: histochemical and phagocytic studies. *Int J Sports Med* 9: 77–81, 1988.
100. **Feldman JL and Stockdale FE.** Temporal appearance of satellite cells during myogenesis. *Dev Biol* 153: 217–226, 1992.
101. **Fennie C, Cheng J, Dowbenko D, Young P, and Lasky LA.** Cd34+ endothelial cell lines derived from murine yolk sac induce the proliferation and differentiation of yolk sac CD34+ hematopoietic progenitors. *Blood* 86: 4454–4467, 1995.
102. **Fernando P, Kelly JF, Balazsi K, Slack RS, and Megeney LA.** Caspase 3 activity is required for skeletal muscle differentiation. *Proc Natl Acad Sci USA* 99: 11025–11030, 2002.
103. **Ferrari G, Cussela-De Angelis G, Coletta M, Paolucci E, Stornaiuolo A, Cossu G, and Mavilio F.** Muscle regeneration by bone marrow-derived myogenic progenitors. *Science* 279: 1528–1530, 1998.
104. **Fielding RA, Manfredi TJ, Ding W, Fiatarone MA, Evans WJ, and Cannon JG.** Acute phase response in exercise. III. Neutrophil and IL-1 beta accumulation in skeletal muscle. *Am J Physiol Regul Integr Comp Physiol* 265: R166–R172, 1993.
105. **Fiore F, Planche J, Gibier P, Sebille A, Delapeyriere O, and Birnbaum D.** Apparent normal phenotype of Fgf6−/− mice. *Int J Dev Biol* 41: 639–642, 1997.
106. **Fiore F, Sebille A, and Birnbaum D.** Skeletal muscle regeneration is not impaired in Fgf6 −/− mutant mice. *Biochem Biophys Res Commun* 272: 138–143, 2000.
107. **Florini JR, Ewton DZ, and Coolican SA.** Growth hormone and the insulin-like growth factor system in myogenesis. *Endocr Rev* 17: 481–517, 1996.
108. **Florini JR, Ewton DZ, and Roof SL.** Insulin-like growth factor-I stimulates terminal myogenic differentiation by induction of myogenin gene expression. *Mol Endocrinol* 5: 718–724, 1991.
109. **Floss T, Arnold HH, and Braun T.** A role for FGF-6 in skeletal muscle regeneration. *Genes Dev* 11: 2040–2051, 1997.
110. **Fuchtbauer EM and Westphal H.** MyoD and myogenin are coexpressed in regenerating skeletal muscle of the mouse. *Dev Dyn* 193: 34–39, 1992.
111. **Gal-Levi R, Leshem Y, Aoki S, Nakamura T, and Halevy O.** Hepatocyte growth factor plays a dual role in regulating skeletal muscle satellite cell proliferation and differentiation. *Biochim Biophys Acta* 1402: 39–51, 1998.
112. **Galli R, Borello U, Gritti A, Minasi MG, Bjornson C, Coletta M, Mora M, De Angelis MG, Fiocco R, Cossu G, and Vescovi AL.** Skeletal myogenic potential of human and mouse neural stem cells. *Nat Neurosci* 3: 986–991, 2000.
113. **Gamble HJ, Fenton J, and Allsopp G.** Electron microscope observations on human fetal striated muscle. *J Anat* 126: 567–589, 1978.
114. **Garry DJ, Meeson A, Elterman J, Zhao Y, Yang P, Bassel-Duby R, and Williams RS.** Myogenic stem cell function is impaired in mice lacking the forkhead/winged helix protein MNF. *Proc Natl Acad Sci USA* 97: 5416–5421, 2000.
115. **Garry DJ, Yang Q, Bassel-Duby R, and Williams RS.** Persistent expression of MNF identifies myogenic stem cells in postnatal muscles. *Dev Biol* 188: 280–294, 1997.
116. **Geiger B and Ayalon O.** Cadherins. *Annu Rev Cell Biol.* 8: 307–332, 1992.
117. **Gibson MC and Schultz E.** The distribution of satellite cells and their relationship to specific fiber types in soleus and extensor digitorum longus muscles. *Anat Rec* 202: 329–337, 1982.
118. **Gibson MC and Schultz E.** Age-related differences in absolute numbers of skeletal muscle satellite cells. *Muscle Nerve* 6: 574–580, 1983.
119. **Goldring K, Partridge T, and Watt D.** Muscle stem cells. *J Pathol* 197: 457–467, 2002.
120. **Goodell MA, Brose K, Paradis G, Conner AS, and Mulligan RC.** Isolation and functional properties of murine hematopoietic stem cells that are replicating in vivo. *J Exp Med* 183: 1797–1806, 1996.
121. **Goulding MD, Chalepakis G, Deutsch U, Erselius JR, and Gruss P.** Pax-3, a novel murine DNA binding protein expressed during early neurogenesis. *EMBO J* 10: 1135–1147, 1991.
122. **Greene EA and Allen RE.** Growth factor regulation of bovine satellite cell growth in vitro. *J Anim Sci* 69: 146–152, 1991.
123. **Grobet L, Martin LJ, Poncelet D, Pirottin D, Brouwers B, Riquet J, Schoeberlein A, Dunner S, Menissier F, Massabanda J, Fries R, Hanset R, and Georges M.** A deletion in the bovine myostatin gene causes the double-muscled phenotype in cattle. *Nat Genet* 17: 71–74, 1997.
124. **Gross JG and Morgan JE.** Muscle precursor cells injected into irradiated mdx mouse muscle persist after serial injury. *Muscle Nerve* 22: 174–185, 1999.
125. **Grounds MD.** Phagocytosis of necrotic muscle in muscle isografts is influenced by the strain, age, and sex of host mice. *J Pathol* 153: 71–82, 1987.
126. **Grounds MD.** Muscle regeneration: molecular aspects and therapeutic implications. *Curr Opin Neurol* 12: 535–543, 1999.
127. **Grounds MD, Garrett KL, Lai MC, Wright WE, and Beilharz MW.** Identification of skeletal muscle precursor cells in vivo by use of MyoD1 and myogenin probes. *Cell Tissue Res* 267: 99–104, 1992.
128. **Grounds MD and McGeachie JK.** A comparison of muscle precursor replication in crush-injured skeletal muscle of Swiss and BALBc mice. *Cell Tissue Res* 255: 385–391, 1989.
129. **Grounds MD, White JD, Rosenthal N, and Bogoyevitch MA.** The role of stem cells in skeletal and cardiac muscle repair. *J Histochem Cytochem* 50: 589–610, 2002.
130. **Gussoni E, Bennett RR, Muskiewicz KR, Meyerrose T, Nolta JA, Gilgoff I, Stein J, Chan YM, Lidov HG, Bonnemann CG, Von Moers A, Morris GE, Den Dunnen JT, Chamberlain JS, Kunkel LM, and Weinberg K.** Long-term persistence of donor nuclei in a Duchenne muscular dystrophy patient receiving bone marrow transplantation. *J Clin Invest* 110: 807–814, 2002.
131. **Gussoni E, Blau HM, and Kunkel LM.** The fate of individual myoblasts after transplantation into muscles of DMD patients. *Nat Med* 3: 970–977, 1997.
132. **Gussoni E, Soneoka Y, Strickland CD, Buzney EA, Khan MK, Flint AF, Kunkel LM, and Mulligan RC.** Dystrophin expression in the mdx mouse restored by stem cell transplantation. *Nature* 401: 390–394, 1999.
133. **Hall-Craggs EC.** Rapid degeneration and regeneration of a whole skeletal muscle following treatment with bupivacaine (Marcain). *Exp Neurol* 43: 349–358, 1974.
134. **Hall-Craggs EC and Seyan HS.** Histochemical changes in innervated and denervated skeletal muscle fibers following treatment with bupivacaine (marcain). *Exp Neurol* 46: 345–354, 1975.
135. **Hamer PW, McGeachie JM, Davies MJ, and Grounds MD.** Evans Blue dye as an in vivo marker of myofibre damage: optimising parameters for detecting initial myofibre membrane permeability. *J Anat* 200: 69–79, 2002.
136. **Hansen-Smith FM.** Development and innervation of soleplates in the freely grafted extensor digitorum longus (EDL) muscle in the rat. *Anat Rec* 207: 55–67, 1983.
137. **Hansen-Smith FM and Carlson BM.** Cellular responses to free grafting of the extensor digitorum longus muscle of the rat. *J Neurol Sci* 41: 149–173, 1979.
138. **Harris JB and Johnson MA.** Further observations on the pathological responses of rat skeletal muscle to toxins isolated from the venom of the Australian tiger snake, *Notechis scutatus scutatus*. *Clin Exp Pharmacol Physiol* 5: 587–600, 1978.
139. **Harris JB, Johnson MA, and Karlsson E.** Pathological responses of rat skeletal muscle to a single subcutaneous injection of a toxin isolated from the venom of the Australian tiger snake *Notechis scutatus scutatus*. *Clin Chem Pharm Physiol* 2: 383–404, 1975.
140. **Hartley RS, Bandman E, and Yablonka-Reuveni Z.** Skeletal muscle satellite cells appear during late chicken embryogenesis. *Dev Biol* 153: 206–216, 1992.
141. **Hasty P, Bradley A, Morris JH, Edmondson DG, Venuti JM, Olson EN, and Klein WH.** Muscle deficiency and neonatal death

in mice with a targeted mutation in the myogenin gene. *Nature* 364: 501–506, 1993.
142. **Hawke TJ and Garry DJ.** Myogenic satellite cells: physiology to molecular biology. *J Appl Physiol* 91: 534–551, 2001.
143. **Heslop L, Beauchamp JR, Tajbakhsh S, Buckingham ME, Partridge TA, and Zammit PS.** Transplanted primary neonatal myoblasts can give rise to functional satellite cells as identified using the Myf5nlacZl+ mouse. *Gene Ther* 8: 778–783, 2001.
144. **Hollnagel A, Grund C, Franke WW, and Arnold HH.** The cell adhesion molecule M-cadherin is not essential for muscle development and regeneration. *Mol Cell Biol* 22: 4760–4770, 2002.
145. **Houzelstein D, Auda-Boucher G, Cheraud Y, Rouaud T, Blanc I, Tajbakhsh S, Buckingham ME, Fontaine-Perus J, and Robert B.** The homeobox gene *Msx1* is expressed in a subset of somites, and in muscle progenitor cells migrating into the forelimb. *Development* 126: 2689–2701, 1999.
146. **Hubank M and Schatz DG.** Identifying differences in mRNA expression by representational difference analysis of cDNA. *Nucleic Acids Res* 22: 5640–5648, 1994.
147. **Huxley AF.** Cross-bridge action: present views, prospects, and unknowns. *J Biomech* 33: 1189–1195, 2000.
148. **Illa I, Leon-Monzon M, and Dalakas MC.** Regenerating and denervated human muscle fibers and satellite cells express neural cell adhesion molecule recognized by monoclonal antibodies to natural killer cells. *Ann Neurol* 31: 46–52, 1992.
149. **Irintchev A and Wernig A.** Muscle damage and repair in voluntarily running mice: strain and muscle differences. *Cell Tissue Res* 249: 509–521, 1987.
150. **Irintchev A, Zeschnigk M, Starzinski-Powitz A, and Wernig A.** Expression pattern of M-cadherin in normal, denervated, and regenerating mouse muscles. *Dev Dyn* 199: 326–337, 1994.
151. **Jankowski RJ, Deasy BM, Cao B, Gates C, and Huard J.** The role of CD34 expression and cellular fusion in the regeneration capacity of myogenic progenitor cells. *J Cell Sci* 115: 4361–4374, 2002.
152. **Jennische E, Ekberg S, and Matejka GL.** Expression of hepatocyte growth factor in growing and regenerating rat skeletal muscle. *Am J Physiol Cell Physiol* 265: C122–C128, 1993.
153. **Jesse TL, Lachance R, Iademarco MF, and Dean DC.** Interferon regulatory factor-2 is a transcriptional activator in muscle where it regulates expression of vascular cell adhesion molecule-1. *J Cell Biol* 140: 1265–1276, 1998.
154. **Jostes B, Walther C, and Gruss P.** The murine paired box gene, Pax7, is expressed specifically during the development of the nervous and muscular system. *Mech Dev* 33: 27–37, 1990.
155. **Kablar B, Asakura A, Krastel K, Ying C, May LL, Goldhamer DJ, and Rudnicki MA.** MyoD and Myf-5 define the specification of musculature of distinct embryonic origin. *Biochem Cell Biol* 76: 1079–1091, 1998.
156. **Kablar B and Rudnicki MA.** Skeletal muscle development in the mouse embryo. *Histol Histopathol* 15: 649–656, 2000.
157. **Kahn EB and Simpson SB Jr.** Satellite cells in mature, uninjured skeletal muscle of the lizard tail. *Dev Biol* 37: 219–223, 1974.
158. **Kambadur R, Sharma M, Smith TP, and Bass JJ.** Mutations in myostatin (GDF8) in double-muscled Belgian Blue and Piedmontese cattle. *Genome Res* 7: 910–916, 1997.
159. **Kami K and Senba E.** Localization of leukemia inhibitory factor and interleukin-6 messenger ribonucleic acids in regenerating rat skeletal muscle. *Muscle Nerve* 21: 819–822, 1998.
160. **Karim L, Coppieters W, Grobet L, Valentini A, and Georges M.** Convenient genotyping of six myostatin mutations causing double-muscling in cattle using a multiplex oligonucleotide ligation assay. *Anim Genet* 31: 396–399, 2000.
161. **Kastner S, Elias MC, Rivera AJ, and Yablonka-Reuveni Z.** Gene expression patterns of the fibroblast growth factors and their receptors during myogenesis of rat satellite cells. *J Histochem Cytochem* 48: 1079–1096, 2000.
162. **Katagiri T, Yamaguchi A, Komaki M, Abe E, Takahashi N, Ikeda T, Rosen V, Wozney JM, Fujisawa-Sehara A, and Suda T.** Bone morphogenetic protein-2 converts the differentiation pathway of C_2C_{12} myoblasts into the osteoblast lineage. *J Cell Biol* 127: 1755–1766, 1994.
163. **Kaufmann U, Martin B, Link D, Witt K, Zeitler R, Reinhard S, and Starzinski-Powitz A.** M-cadherin and its sisters in development of striated muscle. *Cell Tissue Res* 296: 191–198, 1999.
164. **Kaul A, Koster M, Neuhaus H, and Braun T.** Myf-5 revisited: loss of early myotome formation does not lead to a rib phenotype in homozygous Myf-5 mutant mice. *Cell* 102: 17–19, 2000.
165. **Kay PH, Harmon D, Fletcher S, Robertson T, Ziman M, and Papadimitriou JM.** Pax7 includes two polymorphic homeoboxes which contain rearrangements associated with differences in the ability to regenerate damaged skeletal muscle in adult mice. *Int J Biochem Cell Biol* 30: 261–269, 1998.
166. **Kay PH, Marlow SA, Mitchell CA, and Papadimitriou JM.** Studies on the evolution and function of different forms of the mouse myogenic gene Myo-D1 and upstream flanking region. *Gene* 124: 215–222, 1993.
167. **Kay PH, Mitchell CA, Akkari A, and Papadimitriou JM.** Association of an unusual form of a Pax7-like gene with increased efficiency of skeletal muscle regeneration. *Gene* 163: 171–177, 1995.
168. **Kirk S, Oldham J, Kambadur R, Sharma M, Dobbie P, and Bass J.** Myostatin regulation during skeletal muscle regeneration. *J Cell Physiol* 184: 356–363, 2000.
169. **Knoblich JA, Jan LY, and Jan YN.** Asymmetric segregation of Numb and Prospero during cell division. *Nature* 377: 624–627, 1995.
170. **Kocamis H, McFarland DC, and Killefer J.** Temporal expression of growth factor genes during myogenesis of satellite cells derived from the biceps femoris and pectoralis major muscles of the chicken. *J Cell Physiol* 186: 146–152, 2001.
171. **Koshiba K, Kuroiwa A, Yamamoto H, Tamura K, and Ide H.** Expression of *Msx* genes in regenerating and developing limbs of axolotl. *J Exp Zool* 282: 703–714, 1998.
172. **Krause DS, Ito T, Fackler MJ, Smith OM, Collector MI, Sharkis SJ, and May WS.** Characterization of murine CD34, a marker for hematopoietic progenitor and stem cells. *Blood* 84: 691–701, 1994.
173. **Krensky AM.** Scid mouse models: more than furry flasks. *Nat Biotechnol* 15: 720–721, 1997.
174. **Krishan K and Dhoot GK.** Changes in some troponin and insulin-like growth factor messenger ribonucleic acids in regenerating and denervated skeletal muscles. *J Muscle Res Cell Motil* 17: 513–521, 1996.
175. **Kuch C, Winnekendonk D, Butz S, Unvericht U, Kemler R, and Starzinski-Powitz A.** M-cadherin-mediated cell adhesion and complex formation with the catenins in myogenic mouse cells. *Exp Cell Res* 232: 331–338, 1997.
176. **Kumar A, Velloso CP, Imokawa Y, and Brockes JP.** Plasticity of retrovirus-labelled myotubes in the newt limb regeneration blastema. *Dev Biol* 218: 125–136, 2000.
177. **Kurek J, Bower J, Romanella M, and Austin L.** Leukaemia inhibitory factor treatment stimulates muscle regeneration in the mdx mouse. *Neurosci Lett* 212: 167–170, 1996.
178. **Kurek JB, Bower JJ, Romanella M, Koentgen F, Murphy M, and Austin L.** The role of leukemia inhibitory factor in skeletal muscle regeneration. *Muscle Nerve* 20: 815–822, 1997.
179. **Kurek JB, Nouri S, Kannourakis G, Murphy M, and Austin L.** Leukemia inhibitory factor and interleukin-6 are produced by diseased and regenerating skeletal muscle. *Muscle Nerve* 19: 1291–1301, 1996.
180. **Kwak KB, Chung SS, Kim OM, Kang MS, Ha DB, and Chung CH.** Increase in the level of m-calpain correlates with the elevated cleavage of filamin during myogenic differentiation of embryonic muscle cells. *Biochim Biophys Acta* 1175: 243–249, 1993.
181. **Labarge MA and Blau HM.** Biological progression from adult bone marrow to mononucleate muscle stem cell to multinucleate muscle fiber in response to injury. *Cell* 111: 589–601, 2002.
182. **Lawlor MA, Feng X, Everding DR, Sieger K, Stewart CE, and Rotwein P.** Dual control of muscle cell survival by distinct growth factor-regulated signaling pathways. *Mol Cell Biol* 20: 3256–3265, 2000.
183. **Lawlor MA and Rotwein P.** Insulin-like growth factor-mediated muscle cell survival: central roles for Akt and cyclin-dependent kinase inhibitor p21. *Mol Cell Biol* 20: 8983–8995, 2000.
184. **Leblanc AD, Jaweed M, and Evans H.** Evaluation of muscle

injury using magnetic resonance imaging. *Clin J Sport Med* 3: 26–30, 1993.
185. **Lee JY, Qu-Petersen Z, Cao B, Kimura S, Jankowski R, Cummins J, Usas A, Gates C, Robbins P, Wernig A, and Huard J.** Clonal isolation of muscle-derived cells capable of enhancing muscle regeneration and bone healing. *J Cell Biol* 150: 1085–1100, 2000.
186. **Lee SJ and McPherron AC.** Regulation of myostatin activity and muscle growth. *Proc Natl Acad Sci USA* 98: 9306–9311, 2001.
187. **Lefaucheur J and Sebille A.** The cellular events of injured muscle regeneration depend on the nature of the injury. *Neuromuscular Disorders* 5: 501–509, 1995.
188. **Lefaucheur J and Sebille A.** Basic fibroblast growth factor promotes in vivo muscle regeneration in murine muscular dystrophy. *Neurosci Lett* 202: 121–124, 1995.
189. **Lefaucheur J and Sebille A.** Muscle regeneration following injury can be modified in vivo by immune neutralization of basic fibroblast growth factor, transforming growth factor beta 1 or insulin-like growth factor I. *J Neuroimmunol* 57: 85–91, 1995.
190. **Lefaucheur JP, Gjata B, Lafont H, and Sebille A.** Angiogenic and inflammatory responses following skeletal muscle injury are altered by immune neutralization of endogenous basic fibroblast growth factor, insulin-like growth factor-1 and transforming growth factor-beta 1. *J Neuroimmunol* 70: 37–44, 1996.
191. **Lescaudron L, Peltekian E, Fontaine-Perus J, Paulin D, Zampieri M, Garcia L, and Parrish E.** Blood borne macrophages are essential for the triggering of muscle regeneration following muscle transplant. *Neuromuscular Disorders* 9: 72–80, 1999.
192. **Levinovitz A, Jennische E, Oldfors A, Edwall D, and Norstedt G.** Activation of insulin-like growth factor II expression during skeletal muscle regeneration in the rat: correlation with myotube formation. *Mol Endocrinol* 6: 1227–1234, 1992.
193. **Li Z, Mericskay M, Agbulut O, Butler-Browne G, Carlsson L, Thornell LE, Babinet C, and Paulin D.** Desmin is essential for the tensile strength and integrity of myofibrils but not for myogenic commitment, differentiation, and fusion of skeletal muscle. *J Cell Biol* 139: 129–144, 1997.
194. **Lipton BH and Schultz E.** Developmental fate of skeletal muscle satellite cells. *Science* 205: 1292–1294, 1979.
195. **Lo DC, Allen F, and Brockes JP.** Reversal of muscle differentiation during urodele limb regeneration. *Proc Natl Acad Sci USA* 90: 7230–7234, 1993.
196. **Lotzerich H, Fehr HG, and Appell HJ.** Potentiation of cytostatic but not cytolytic activity of murine macrophages after running stress. *Int J Sports Med* 11: 61–65, 1990.
197. **Maley MA, Fan Y, Beilharz MW, and Grounds MD.** Intrinsic differences in MyoD and myogenin expression between primary cultures of SJL/J and BALB/C skeletal muscle. *Exp Cell Res* 211: 99–107, 1994.
198. **Mansouri A, Goudreau G, and Gruss P.** Pax genes and their role in organogenesis. *Cancer Res* 59: 1707s–1710s, 1999.
199. **Mansouri A, Stoykova A, Torres M, and Gruss P.** Dysgenesis of cephalic neural crest derivatives in Pax7−/− mutant mice. *Development* 122: 831–838, 1996.
200. **Maroto M, Reshef R, Munsterberg AE, Koester S, Goulding M, and Lassar AB.** Ectopic Pax-3 activates MyoD and Myf-5 expression in embryonic mesoderm and neural tissue. *Cell* 89: 139–148, 1997.
201. **Matsuda R, Nishikawa A, and Tanaka H.** Visualization of dystrophic muscle fibers in mdx mouse by vital staining with Evans blue: evidence of apoptosis in dystrophin-deficient muscle. *J Biochem* 118: 959–964, 1995.
202. **Mauro A.** Satellite cells of skeletal muscle fibres. *J Biophys Biochem Cytol* 9: 493–495, 1961.
203. **McFarland DC, Pesall JE, and Gilkerson KK.** The influence of growth factors on turkey embryonic myoblasts and satellite cells in vitro. *Gen Comp Endocrinol* 89: 415–424, 1993.
204. **McGann CJ, Odelberg SJ, and Keating MT.** Mammalian myotube dedifferentiation induced by newt regeneration extract. *Proc Natl Acad Sci USA* 98: 13699–13704, 2001.
205. **McGeachie JK and Grounds MD.** Retarded myogenic cell replication in regenerating skeletal muscles of old mice: an autoradiographic study in young and old BALBc and SJL/J mice. *Cell Tissue Res* 280: 277–282, 1995.
206. **McKinney-Freeman SL, Jackson KA, Camargo FD, Ferrari G, Mavilio F, and Goodell MA.** Muscle-derived hematopoietic stem cells are hematopoietic in origin. *Proc Natl Acad Sci USA* 99: 1341–1346, 2002.
207. **McLennan IS and Koishi K.** The transforming growth factor-betas: multifaceted regulators of the development and maintenance of skeletal muscles, motoneurons and Schwann cells. *Int J Dev Biol* 46: 559–567, 2002.
208. **McPherron AC, Lawler AM, and Lee SJ.** Regulation of skeletal muscle mass in mice by a new TGF-beta superfamily member. *Nature* 387: 83–90, 1997.
209. **McPherron AC and Lee SJ.** Double muscling in cattle due to mutations in the myostatin gene. *Proc Natl Acad Sci USA* 94: 12457–12461, 1997.
210. **Megeney LA, Kablar B, Garrett K, Anderson JE, and Rudnicki MA.** MyoD is required for myogenic stem cell function in adult skeletal muscle. *Genes Dev* 10: 1173–1183, 1996.
211. **Mendler L, Zador E, Ver Heyen M, Dux L, and Wuytack F.** Myostatin levels in regenerating rat muscles and in myogenic cell cultures. *J Muscle Res Cell Motil* 21: 551–563, 2000.
212. **Merly F, Lescaudron L, Rouaud T, Crossin F, and Gardahaut MF.** Macrophages enhance muscle satellite cell proliferation and delay their differentiation. *Muscle Nerve* 22: 724–732, 1999.
213. **Miller JB, Schaefer L, and Dominov JA.** Seeking muscle stem cells. *Curr Top Dev Biol* 43: 191–219, 1999.
214. **Miller KJ, Thaloor D, Matteson S, and Pavlath GK.** Hepatocyte growth factor affects satellite cell activation and differentiation in regenerating skeletal muscle. *Am J Physiol Cell Physiol* 278: C174–C181, 2000.
215. **Mina M, Wang YH, Ivanisevic AM, Upholt WB, and Rodgers B.** Region- and stage-specific effects of FGFs and BMPs in chick mandibular morphogenesis. *Dev Dyn* 223: 333–352, 2002.
216. **Minasi MG, Riminucci M, De Angelis L, Borello U, Berarducci B, Innocenzi A, Caprioli A, Sirabella D, Baiocchi M, De Maria R, Boratto R, Jaffredo T, Broccoli V, Bianco P, and Cossu G.** The meso-angioblast: a multipotent, self-renewing cell that originates from the dorsal aorta and differentiates into most mesodermal tissues. *Development* 129: 2773–2783, 2002.
217. **Mitchell CA, McGeachie JK, and Grounds MD.** Cellular differences in the regeneration of murine skeletal muscle: a quantitative histological study in SJL/J and BALB/c mice. *Cell Tissue Res* 269: 159–166, 1992.
218. **Mitchell CA, McGeachie JK, and Grounds MD.** The exogenous administration of basic fibroblast growth factor to regenerating skeletal muscle in mice does not enhance the process of regeneration. *Growth Factors* 13: 37–55, 1996.
219. **Moore R and Walsh FS.** The cell adhesion molecule M-cadherin is specifically expressed in developing and regenerating, but not denervated skeletal muscle. *Development* 117: 1409–1420, 1993.
220. **Morel F, Szilvassy SJ, Travis M, Chen B, and Galy A.** Primitive hematopoietic cells in murine bone marrow express the CD34 antigen. *Blood* 88: 3774–3784, 1996.
221. **Musaro A, McCullagh KJ, Naya FJ, Olson EN, and Rosenthal N.** IGF-1 induces skeletal myocyte hypertrophy through calcineurin in association with GATA-2 and NF-ATc1. *Nature* 400: 581–585, 1999.
222. **Musaro A, McCullagh K, Paul A, Houghton L, Dobrowolny G, Molinaro M, Barton ER, Sweeney HL, and Rosenthal N.** Localized IGF-1 transgene expression sustains hypertrophy and regeneration in senescent skeletal muscle. *Nat Genet* 27: 195–200, 2001.
223. **Nabeshima Y, Hanaoka K, Hayasaka M, Esumi E, Li S, and Nonaka I.** Myogenin gene disruption results in perinatal lethality because of severe muscle defect. *Nature* 364: 532–535, 1993.
224. **Nakamura T, Nishizawa T, Hagiya M, Seki T, Shimonishi M, Sugimura A, Tashiro K, and Shimizu S.** Molecular cloning and expression of human hepatocyte growth factor. *Nature* 342: 440–443, 1989.
225. **Nakamura T, Teramoto H, and Ichihara A.** Purification and characterization of a growth factor from rat platelets for mature parenchymal hepatocytes in primary cultures. *Proc Natl Acad Sci USA* 83: 6489–6493, 1986.
226. **Nicholson GA, Gardner-Medwin D, Pennington RJ, and Walton JN.** Carrier detection in Duchenne muscular dystrophy: as-

sessment of the effect of age on detection-rate with serum-creatine-kinase-activity. *Lancet* 1: 692–694, 1979.
227. **Odelberg SJ, Kollhoff A, and Keating MT.** Dedifferentiation of mammalian myotubes induced by msx1. *Cell* 103: 1099–1109, 2000.
228. **Ordahl CP.** Myogenic shape-shifters. *J Cell Biol* 147: 695–698, 1999.
229. **Ordahl CP, Williams BA, and Denetclaw W.** Determination and morphogenesis in myogenic progenitor cells: an experimental embryological approach. *Curr Top Dev Biol* 48: 319–367, 2000.
230. **Orimo S, Hiyamuta E, Arahata K, and Sugita H.** Analysis of inflammatory cells and complement C3 in bupivacaine-induced myonecrosis. *Muscle Nerve* 14: 515–520, 1991.
231. **Palacio J, Galdiz JB, Alvarez FJ, Orozco-Levi M, Lloreta J, and Gea J.** Procion orange tracer dye technique vs. identification of intrafibrillar fibronectin in the assessment of sarcolemmal damage. *Eur J Clin Invest* 32: 443–447, 2002.
232. **Palmer CM and Rudnicki MA.** The myogenic regulatory factors. In: *Advances in Developmental Biology and Biochemistry*. New York: Elsevier Science, 2001, p. 1–32.
233. **Pardanaud L and Dieterlen-Lievre F.** Ontogeny of the endothelial system in the avian model. *Adv Exp Med Biol* 476: 67–78, 2000.
234. **Pastoret C and Sebille A.** Further aspects of muscular dystrophy in mdx mice. *Neuromuscular Disorders* 3: 471–475, 1993.
235. **Pastoret C and Sebille A.** mdx mice show progressive weakness and muscle deterioration with age. *J Neurol Sci* 129: 97–105, 1995.
236. **Patapoutian A, Yoon JK, Miner JH, Wang S, Stark K, and Wold B.** Disruption of the mouse MRF4 gene identifies multiple waves of myogenesis in the myotome. *Development* 121: 3347–3358, 1995.
237. **Pavlath GK, Thaloor D, Rando TA, Cheong M, English AW, and Zheng B.** Heterogeneity among muscle precursor cells in adult skeletal muscles with differing regenerative capacities. *Dev Dyn* 212: 495–508, 1998.
238. **Percy ME, Chang LS, Murphy EG, Oss I, Verellen-Dumoulin C, and Thompson MW.** Serum creatine kinase and pyruvate kinase in Duchenne muscular dystrophy carrier detection. *Muscle Nerve* 2: 329–339, 1979.
239. **Perez OD, Chang YT, Rosania G, Sutherlin D, and Schultz PG.** Inhibition and reversal of myogenic differentiation by purine-based microtubule assembly inhibitors. *Chem Biol* 9: 475–483, 2002.
240. **Peterson CA.** Cell culture systems as tools for studying age-related changes in skeletal muscle. *J Gerontol* 50A: 142–144, 1995.
241. **Pietsch J.** The effects of colchicine on regeneration of mouse skeletal muscle. *Anat Rec* 139: 167–172, 1961.
242. **Popiela H.** Muscle satellite cells in urodele amphibians: faciliatated identification of satellite cells using ruthenium red staining. *J Exp Zool* 198: 57–64, 1976.
243. **Poss KD, Shen J, and Keating MT.** Induction of lef1 during zebrafish fin regeneration. *Dev Dyn* 219: 282–286, 2000.
244. **Quinlan JG, Lyden SP, Cambier DM, Johnson SR, Michaels SE, and Denman DL.** Radiation inhibition of mdx mouse muscle regeneration: dose and age factors. *Muscle Nerve* 18: 201–206, 1995.
245. **Qu-Petersen Z, Deasy B, Jankowski R, Ikezawa M, Cummins J, Pruchnic R, Mytinger J, Cao B, Gates C, Wernig A, and Huard J.** Identification of a novel population of muscle stem cells in mice: potential for muscle regeneration. *J Cell Biol* 157: 851–864, 2002.
246. **Rantanen J, Hurme T, Lukka R, Heino J, and Kalimo H.** Satellite cell proliferation and the expression of myogenin and desmin in regenerating skeletal muscle: evidence for two different populations of satellite cells. *Lab Invest* 72: 341–347, 1995.
247. **Rappolee DA and Werb Z.** Macrophage-derived growth factors. *Curr Top Microbiol Immunol* 181: 87–140, 1992.
248. **Rapraeger AC.** Syndecan-regulated receptor signaling. *J Cell Biol* 149: 995–998, 2000.
249. **Rawls A, Morris JH, Rudnicki M, Braun T, Arnold HH, Klein WH, and Olson EN.** Myogenin's functions do not overlap with those of MyoD or Myf-5 during mouse embryogenesis. *Dev Biol* 172: 37–50, 1995.
250. **Reardon KA, Kapsa RM, Davis J, Kornberg AJ, Austin L, Choong P, and Byrne E.** Increased levels of leukemia inhibitory factor mRNA in muscular dystrophy and human muscle trauma. *Muscle Nerve* 23: 962–966, 2000.
251. **Reginelli AD, Wang YQ, Sassoon D, and Muneoka K.** Digit tip regeneration correlates with regions of Msx1 (Hox 7) expression in fetal and newborn mice. *Development* 121: 1065–1076, 1995.
252. **Rhyu MS, Jan LY, and Jan YN.** Asymmetric distribution of numb protein during division of the sensory organ precursor cell confers distinct fates to daughter cells. *Cell* 76: 477–491, 1994.
253. **Rios R, Carneiro I, Arce VM, and Devesa J.** Myostatin regulates cell survival during C_2C_{12} myogenesis. *Biochem Biophys Res Commun* 280: 561–566, 2001.
254. **Rios R, Carneiro I, Arce VM, and Devesa J.** Myostatin is an inhibitor of myogenic differentiation. *Am J Physiol Cell Physiol* 282: C993–C999, 2002.
255. **Roberts P, McGeachie JK, and Grounds MD.** The host environment determines strain-specific differences in the timing of skeletal muscle regeneration: cross-transplantation studies between SJL/J and BALB/c mice. *J Anat* 191: 585–594, 1997.
256. **Robertson TA, Maley MA, Grounds MD, and Papadimitriou JM.** The role of macrophages in skeletal muscle regeneration with particular reference to chemotaxis. *Exp Cell Res* 207: 321–331, 1993.
257. **Rosania GR, Chang YT, Perez O, Sutherlin D, Dong H, Lockhart DJ, and Schultz PG.** Myoseverin, a microtubule-binding molecule with novel cellular effects. *Nat Biotechnol* 18: 304–308, 2000.
258. **Rosenblatt JD, Lunt AI, Parry DJ, and Partridge TA.** Culturing satellite cells from living single muscle fiber explants. *In Vitro Cell Dev Biol* 31: 773–779, 1995.
259. **Rudnicki MA, Schnegelsberg PN, Stead RH, Braun T, Arnold HH, and Jaenisch R.** MyoD or Myf-5 is required for the formation of skeletal muscle. *Cell* 75: 1351–1359, 1993.
260. **Sabourin LA, Girgis-Gabardo A, Seale P, Asakura A, and Rudnicki MA.** Reduced differentiation potential of primary MyoD−/− myogenic cells derived from adult skeletal muscle. *J Cell Biol* 144: 631–643, 1999.
261. **Sancesario G, Massa R, Anzil AP, and Bernardi G.** Active muscle length reduction progressively damages soleus in hindlimb-suspended rabbits. *Muscle Nerve* 15: 1002–1015, 1992.
262. **Sandri M, Minetti C, Pedemonte M, and Carraro U.** Apoptotic myonuclei in human Duchenne muscular dystrophy. *Lab Invest* 78: 1005–1016, 1998.
263. **Scata KA, Bernard DW, Fox J, and Swain JL.** FGF receptor availability regulates skeletal myogenesis. *Exp Cell Res* 250: 10–21, 1999.
264. **Schiaffino S, Bormioli SP, and Aloisi M.** Fibre branching and formation of new fibres during compensatory muscle hypertrophy. In: *Muscle Regeneration*. New York: Raven, 1979, p. 177–188.
265. **Schmalbruch H and Hellhammer U.** The number of nuclei in adult rat muscles with special reference to satellite cells. *Anat Rec* 189: 169–175, 1977.
266. **Schmalbruch H and Lewis DM.** Dynamics of nuclei of muscle fibers and connective tissue cells in normal and denervated rat muscles. *Muscle Nerve* 23: 617–626, 2000.
267. **Schubert W, Zimmermann K, Cramer M, and Starzinski-Powitz A.** Lymphocyte antigen Leu-19 as a molecular marker of regeneration in human skeletal muscle. *Proc Natl Acad Sci USA* 86: 307–311, 1989.
268. **Schultz E.** Satellite cell proliferative compartments in growing skeletal muscles. *Dev Biol* 175: 84–94, 1996.
269. **Schultz E, Gibson MC, and Champion T.** Satellite cells are mitotically quiescent in mature mouse muscle: an EM and radioautographic study. *J Exp Zool* 206: 451–456, 1978.
270. **Schultz E and Jaryszak DL.** Effects of skeletal muscle regeneration on the proliferation potential of satellite cells. *Mech Ageing Dev* 30: 63–72, 1985.
271. **Schultz E, Jaryszak DL, Gibson MC, and Albright DJ.** Absence of exogenous satellite cell contribution to regeneration of frozen skeletal muscle. *J Muscle Res Cell Motil* 7: 361–367, 1986.
272. **Schultz E, Jaryszak DL, and Valliere CR.** Response of satellite cells to focal skeletal muscle injury. *Muscle Nerve* 8: 217–222, 1985.
273. **Schultz E and Lipton BH.** Skeletal muscle satellite cells: changes in proliferation potential as a function of age. *Mech Ageing Dev* 20: 377–383, 1982.
274. **Seale P and Rudnicki MA.** A new look at the origin, function, and "stem-cell" status of muscle satellite cells. *Dev Biol* 218: 115–124, 2000.
275. **Seale P, Sabourin LA, Girgis-Gabardo A, Mansouri A, Gruss**

P, and Rudnicki MA. Pax7 is required for the specification of myogenic satellite cells. *Cell* 102: 777–786, 2000.
276. **Sefton M, Sanchez S, and Nieto MA.** Conserved and divergent roles for members of the Snail family of transcription factors in the chick and mouse embryo. *Development* 125: 3111–3121, 1998.
277. **Semsarian C, Sutrave P, Richmond DR, and Graham RM.** Insulin-like growth factor (IGF-I) induces myotube hypertrophy associated with an increase in anaerobic glycolysis in a clonal skeletal-muscle cell model. *Biochem J* 339: 443–451, 1999.
278. **Semsarian C, Wu MJ, Ju YK, Marciniec T, Yeoh T, Allen DG, Harvey RP, and Graham RM.** Skeletal muscle hypertrophy is mediated by a Ca^{2+}-dependent calcineurin signalling pathway. *Nature* 400: 576–581, 1999.
279. **Sharma M, Langley B, Bass J, and Kambadur R.** Myostatin in muscle growth and repair. *Exercise Sports Sci Rev* 29: 155–158, 2001.
280. **Sheehan SM and Allen RE.** Skeletal muscle satellite cell proliferation in response to members of the fibroblast growth factor family and hepatocyte growth factor. *J Cell Physiol* 181: 499–506, 1999.
281. **Sheehan SM, Tatsumi R, Temm-Grove CJ, and Allen RE.** HGF is an autocrine growth factor for skeletal muscle satellite cells in vitro. *Muscle Nerve* 23: 239–245, 2000.
282. **Shore EM, Glaser DL, and Gannon FH.** Osteogenic induction in hereditary disorders of heterotopic ossification. *Clin Orthop* 374: 303–316, 2000.
283. **Sicinski P, Geng Y, Ryder-Cook AS, Barnard EA, Darlison MG, and Barnard PJ.** The molecular basis of muscular dystrophy in the mdx mouse: a point mutation. *Science* 244: 1578–1580, 1989.
284. **Simon HG, Nelson C, Goff D, Laufer E, Morgan BA, and Tabin C.** Differential expression of myogenic regulatory genes and Msx-1 during dedifferentiation and redifferentiation of regenerating amphibian limbs. *Dev Dyn* 202: 1–12, 1995.
285. **Smith CK, Janney MJ, and Allen RE.** Temporal expression of myogenic regulatory genes during activation, proliferation and differentiation of rat skeletal muscle satellite cells. *J Cell Physiol* 159: 379–385, 1994.
286. **Smith J, Goldsmith C, Ward A, and Ledieu R.** IGF-II ameliorates the dystrophic phenotype and coordinately down-regulates programmed cell death. *Cell Death Differ* 7: 1109–1118, 2000.
287. **Smith J and Schofield PN.** Stable integration of an mdx skeletal muscle cell line into dystrophic (mdx) skeletal muscle: evidence for stem cell status. *Cell Growth Differ* 8: 927–934, 1997.
288. **Smythe GM, Davies MJ, Paulin D, and Grounds MD.** Absence of desmin slightly prolongs myoblast proliferation and delays fusion in vivo in regenerating grafts of skeletal muscle. *Cell Tissue Res* 304: 287–294, 2001.
289. **Snow MH.** Myogenic cell formation in regenerating rat skeletal muscle injured by mincing. II. An autoradiographic study. *Anat Rec* 188: 201–217, 1977.
290. **Snow MH.** An autoradiographic study of satellite cell differentiation into regenerating myotubes following transplantation of muscles in young rats. *Cell Tissue Res* 186: 535–540, 1978.
291. **Snow MH.** A quantitative ultrastructure analysis of satellite cells in denervated fast and slow muscles of the mouse. *Anat Rec* 207: 593–604, 1983.
292. **Sorichter S, Mair J, Koller A, Muller E, Kremser C, Judmaier W, Haid C, Calzolari C, and Puschendorf B.** Creatine kinase, myosin heavy chains and magnetic resonance imaging after eccentric exercise. *J Sports Sci* 19: 687–691, 2001.
293. **Sorimachi H, Ishiura S, and Suzuki K.** Structure and physiological function of calpains. *Biochem J* 328: 721–732, 1997.
294. **Spangenburg EE and Booth FW.** Multiple signaling pathways mediate LIF-induced skeletal muscle satellite cell proliferation. *Am J Physiol Cell Physiol* 283: C204–C211, 2002.
295. **Spencer MJ, Walsh CM, Dorshkind KA, Rodriguez EM, and Tidball JG.** Myonuclear apoptosis in dystrophic mdx muscle occurs by perforin-mediated cytotoxicity. *J Clin Invest* 99: 2745–2751, 1997.
296. **Stamler JS and Meissner G.** Physiology of nitric oxide in skeletal muscle. *Physiol Rev* 81: 209–237, 2001.
297. **Straub V, Duclos F, Venzke DP, Lee JC, Cutshall S, Leveille CJ, and Campbell KP.** Molecular pathogenesis of muscle degeneration in the delta-sarcoglycan-deficient hamster. *Am J Pathol* 153: 1623–1630, 1998.
298. **Straub V, Rafael JA, Chamberlain JS, and Campbell KP.** Animal models for muscular dystrophy show different patterns of sarcolemmal disruption. *J Cell Biol* 139: 375–385, 1997.
299. **Suelves M, Lopez-Alemany R, Lluis F, Aniorte G, Serrano E, Parra M, Carmeliet P, and Munoz-Canoves P.** Plasmin activity is required for myogenesis in vitro and skeletal muscle regeneration in vivo. *Blood* 99: 2835–2844, 2002.
300. **Suzuki S, Yamanouchi K, Soeta C, Katakai Y, Harada R, Naito K, and Tojo H.** Skeletal muscle injury induces hepatocyte growth factor expression in spleen. *Biochem Biophys Res Commun* 292: 709–714, 2002.
301. **Suzuki J, Yamazaki Y, Li G, Kaziro Y, and Koide H.** Involvement of Ras and Ral in chemotactic migration of skeletal myoblasts. *Mol Cell Biol* 20: 4658–4665, 2000.
302. **Szabo G, Dallmann G, Muller G, Patthy L, Soller M, and Varga L.** A deletion in the myostatin gene causes the compact (Cmpt) hypermuscular mutation in mice. *Mamm Genome* 9: 671–672, 1998.
303. **Tajbakhsh S, Rocancourt D, Cossu G, and Buckingham M.** Redefining the genetic hierarchies controlling skeletal myogenesis: Pax-3 and Myf-5 act upstream of MyoD. *Cell* 89: 127–138, 1997.
304. **Tanabe Y, Esaki K, and Nomura T.** Skeletal muscle pathology in X chromosome-linked muscular dystrophy (*mdx*) mouse. *Acta Neuropathol* 69: 91–95, 1986.
305. **Tanaka EM, Drechsel DN, and Brockes JP.** Thrombin regulates S-phase re-entry by cultured newt myotubes. *Curr Biol* 9: 792–799, 1999.
306. **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.
307. **Tatsumi R, Hattori A, Ikeuchi Y, Anderson JE, and Allen RE.** Release of hepatocyte growth factor from mechanically stretched skeletal muscle satellite cells and role of pH and nitric oxide. *Mol Biol Cell* 13: 2909–2918, 2002.
308. **Tatsumi R, Sheehan SM, Iwasaki H, Hattori A, and Allen RE.** Mechanical stretch induces activation of skeletal muscle satellite cells in vitro. *Exp Cell Res* 267: 107–114, 2001.
309. **Taylor WE, Bhasin S, Artaza J, Byhower F, Azam M, Willard DH Jr, Kull FC Jr, and Gonzalez-Cadavid N.** Myostatin inhibits cell proliferation and protein synthesis in C_2C_{12} muscle cells. *Am J Physiol Endocrinol Metab* 280: E221–E228, 2001.
310. **Teboul L, Gaillard D, Staccini L, Inadera H, Amri EZ, and Grimaldi PA.** Thiazolidinediones and fatty acids convert myogenic cells into adipose-like cells. *J Biol Chem* 270: 28183–28187, 1995.
311. **Temm-Grove CJ, Wert D, Thompson VF, Allen RE, and Goll DE.** Microinjection of calpastatin inhibits fusion in myoblasts. *Exp Cell Res* 247: 293–303, 1999.
312. **Tennyson VM, Brzin M, and Kremzner LT.** Acetylcholinesterase activity in the myotube and muscle satellite cell of the fetal rabbit. An electron microscopic-cytochemical and biochemical study. *J Histochem Cytochem* 21: 634–652, 1973.
313. **Thomas M, Langley B, Berry C, Sharma M, Kirk S, Bass J, and Kambadur R.** Myostatin, a negative regulator of muscle growth, functions by inhibiting myoblast proliferation. *J Biol Chem* 275: 40235–40243, 2000.
314. **Thompson-Jaeger S and Raghow R.** Exogenous expression of Msx1 renders myoblasts refractory to differentiation into myotubes and elicits enhanced biosynthesis of four unique mRNAs. *Mol Cell Biochem* 208: 63–69, 2000.
315. **Tidball JG.** Inflammatory cell response to acute muscle injury. *Med Sci Sports Exercise* 27: 1022–1032, 1995.
316. **Tkatchenko AV, Le Cam G, Leger JJ, and Dechesne CA.** Large-scale analysis of differential gene expression in the hindlimb muscles and diaphragm of mdx mouse. *Biochim Biophys Acta* 1500: 17–30, 2000.
317. **Torrente Y, Tremblay JP, Pisati F, Belicchi M, Rossi B, Sironi M, Fortunato F, El Fahime M, D'Angelo MG, Caron NJ, Constantin G, Paulin D, Scarlato G, and Bresolin N.** Intra-arterial injection of muscle-derived CD34(+)Sca-1(+) stem cells restores dystrophin in *mdx* mice. *J Cell Biol* 152: 335–348, 2001.
318. **Tseng BS, Zhao P, Pattison JS, Gordon SE, Granchelli JA, Madsen RW, Folk LC, Hoffman EP, and Booth FW.** Regenerated mdx mouse skeletal muscle shows differential mRNA expression. *J Appl Physiol* 93: 537–545, 2002.

319. **Vaittinen S, Lukka R, Sahlgren C, Hurme T, Rantanen J, Lendahl U, Eriksson JE, and Kalimo H.** The expression of intermediate filament protein nestin as related to vimentin and desmin in regenerating skeletal muscle. *J Neuropathol Exp Neurol* 60: 588–597, 2001.
320. **Vakakis N, Bower J, and Austin L.** In vitro myoblast to myotube transformations in the presence of leukemia inhibitory factor. *Neurochem Int* 27: 329–335, 1995.
321. **Vandenburgh HH, Karlisch P, Shansky J, and Feldstein R.** Insulin and IGF-I induce pronounced hypertrophy of skeletal myofibers in tissue culture. *Am J Physiol Cell Physiol* 260: C475–C484, 1991.
322. **Vergani L, Di Giulio AM, Losa M, Rossoni G, Muller EE, and Gorio A.** Systemic administration of insulin-like growth factor decreases motor neuron cell death and promotes muscle reinnervation. *J Neurosci Res* 54: 840–847, 1998.
323. **Wada MR, Inagawa-Ogashiwa M, Shimizu S, Yasumoto S, and Hashimoto N.** Generation of different fates from multipotent muscle stem cells. *Development* 129: 2987–2995, 2002.
324. **Wagner KR, McPherron AC, Winik N, and Lee SJ.** Loss of myostatin attenuates severity of muscular dystrophy in mdx mice. *Ann Neurol* 52: 832–836, 2002.
325. **Wakeford S, Watt DJ, and Partridge TA.** X-irradiation improves mdx mouse muscle as a model of myofiber loss in DMD. *Muscle Nerve* 14: 42–50, 1991.
326. **Walker BE.** An investigation of skeletal muscle regeneration with radioautography. *Anat Rec* 136: 350, 1960.
327. **Wallenius V, Wallenius K, Ahren B, Rudling M, Carlsten H, Dickson SL, Ohlsson C, and Jansson JO.** Interleukin-6-deficient mice develop mature-onset obesity. *Nat Med* 8: 75–79, 2002.
328. **Watchko JF, O'Day TL, and Hoffman EP.** Functional characteristics of dystrophic skeletal muscle: insights from animal models. *J Appl Physiol* 93: 407–417, 2002.
329. **Webster C and Blau HM.** Accelerated age-related decline in replicative life-span of Duchenne muscular dystrophy myoblasts: implications for cell and gene therapy. *Somat Cell Mol Genet* 16: 557–565, 1990.
330. **Wehling M, Cai B, and Tidball JG.** Modulation of myostatin expression during modified muscle use. *FASEB J* 14: 103–110, 2000.
331. **Weller B, Karpati G, Lehnert S, and Carpenter S.** Major alteration of the pathological phenotype in gamma irradiated mdx soleus muscles. *J Neuropathol Exp Neurol* 50: 419–431, 1991.
332. **Wernig A, Zweyer M, and Irintchev A.** Function of skeletal muscle tissue formed after myoblast transplantation into irradiated mouse muscles. *J Physiol* 522: 333–345, 2000.
333. **Whalen RG, Harris JB, Butler-Browne GS, and Sesodia S.** Expression of myosin isoforms during notexin-induced regeneration of rat soleus muscles. *Dev Biol* 141: 24–40, 1990.
334. **White JD, Bower JJ, Kurek JB, and Austin L.** Leukemia inhibitory factor enhances regeneration in skeletal muscles after myoblast transplantation. *Muscle Nerve* 24: 695–697, 2001.
335. **Wigmore PM and Evans DJ.** Molecular and cellular mechanisms involved in the generation of fiber diversity during myogenesis. *Int Rev Cytol* 216: 175–232, 2002.
336. **Wilkie RS, O'Neill IE, Butterwith SC, Duclos MJ, and Goddard C.** Regulation of chick muscle satellite cells by fibroblast growth factors: interaction with insulin-like growth factor-I and heparin. *Growth Regul* 5: 18–27, 1995.
337. **Wokke JH, Van Den Oord CJ, Leppink GJ, and Jennekens FG.** Perisynaptic satellite cells in human external intercostal muscle: a quantitative and qualitative study. *Anat Rec* 223: 174–180, 1989.
338. **Woloshin P, Song K, Degnin C, Killary AM, Goldhamer DJ, Sassoon D, and Thayer MJ.** Msx1 inhibits myoD expression in fibroblast × 10T1/2 cell hybrids. *Cell* 82: 611–620, 1995.
339. **Woods JA, Davis JM, Mayer EP, Ghaffar A, and Pate RR.** Exercise increases inflammatory macrophage antitumor cytotoxicity. *J Appl Physiol* 75: 879–886, 1993.
340. **Wright WE.** Myoblast senescence in muscular dystrophy. *Exp Cell Res* 157: 343–354, 1985.
341. **Yablonka-Reuveni Z and Rivera AJ.** Temporal expression of regulatory and structural muscle proteins during myogenesis of satellite cells on isolated adult rat fibers. *Dev Biol* 164: 588–603, 1994.
342. **Yablonka-Reuveni Z, Rudnicki MA, Rivera AJ, Primig M, Anderson JE, and Natanson P.** The transition from proliferation to differentiation is delayed in satellite cells from mice lacking MyoD. *Dev Biol* 210: 440–455, 1999.
343. **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.
344. **Yaffe D and Saxel O.** Serial passaging and differentiation of myogenic cells isolated from dystrophic mouse muscle. *Nature* 270: 725–727, 1977.
345. **Yamada S, Buffinger N, Dimario J, and Strohman RC.** Fibroblast growth factor is stored in fiber extracellular matrix and plays a role in regulating muscle hypertrophy. *Med Sci Sports Exercise* 21: S173–S180, 1989.
346. **Yamanouchi K, Soeta C, Naito K, and Tojo H.** Expression of myostatin gene in regenerating skeletal muscle of the rat and its localization. *Biochem Biophys Res Commun* 270: 510–516, 2000.
347. **Yao SN and Kurachi K.** Implanted myoblasts not only fuse with myofibers but also survive as muscle precursor cells. *J Cell Sci* 105: 957–963, 1993.
348. **Yoon JK, Olson EN, Arnold HH, and Wold BJ.** Different MRF4 knockout alleles differentially disrupt Myf-5 expression: *cis*-regulatory interactions at the MRF4/Myf-5 locus. *Dev Biol* 188: 349–362, 1997.
349. **Yoshida N, Yoshida S, Koishi K, Masuda K, and Nabeshima Y.** Cell heterogeneity upon myogenic differentiation: down-regulation of MyoD and Myf-5 generates "reserve cells." *J Cell Sci* 111: 769–779, 1998.
350. **Young HE, Duplaa C, Young TM, Floyd JA, Reeves ML, Davis KH, Mancini GJ, Eaton ME, Hill JD, Thomas K, Austin T, Edwards C, Cuzzourt J, Parikh A, Groom J, Hudson J, and Black AC Jr.** Clonogenic analysis reveals reserve stem cells in postnatal mammals. I. Pluripotent mesenchymal stem cells. *Anat Rec* 263: 350–360, 2001.
351. **Young HE, Steele TA, Bray RA, Hudson J, Floyd JA, Hawkins K, Thomas K, Austin T, Edwards C, Cuzzourt J, Duenzl M, Lucas PA, and Black AC Jr.** Human reserve pluripotent mesenchymal stem cells are present in the connective tissues of skeletal muscle and dermis derived from fetal, adult, and geriatric donors. *Anat Rec* 264: 51–62, 2001.
352. **Zador E, Bottka S, and Wuytack F.** Antisense inhibition of myoD expression in regenerating rat soleus muscle is followed by an increase in the mRNA levels of myoD, myf-5 and myogenin and by a retarded regeneration. *Biochim Biophys Acta* 1590: 52–63, 2002.
353. **Zammit PS, Heslop L, Hudon V, Rosenblatt JD, Tajbakhsh S, Buckingham ME, Beauchamp JR, and Partridge TA.** Kinetics of myoblast proliferation show that resident satellite cells are competent to fully regenerate skeletal muscle fibers. *Exp Cell Res* 281: 39–49, 2002.
354. **Zarnegar R and Michalopoulos GK.** The many faces of hepatocyte growth factor: from hepatopoiesis to hematopoiesis. *J Cell Biol* 129: 1177–1180, 1995.
355. **Zatz M, Rapaport D, Vainzof M, Passos-Bueno MR, Bortolini ER, Pavanello Rde C, and Peres CA.** Serum creatine-kinase (CK) and pyruvate-kinase (PK) activities in Duchenne (DMD) compared with Becker (BMD) muscular dystrophy. *J Neurol Sci* 102: 190–196, 1991.
356. **Zeschnigk M, Kozian D, Kuch C, Schmoll M, and Starzinski-Powitz A.** Involvement of M-cadherin in terminal differentiation of skeletal muscle cells. *J Cell Sci* 108: 2973–2981, 1995.
357. **Zhang W, Behringer RR, and Olson EN.** Inactivation of the myogenic bHLH gene MRF4 results in up-regulation of myogenin and rib anomalies. *Genes Dev* 9: 1388–1399, 1995.
358. **Zhao P, Iezzi S, Carver E, Dressman D, Gridley T, Sartorelli V, and Hoffman EP.** Slug is a novel downstream target of MyoD. Temporal profiling in muscle regeneration. *J Biol Chem* 277: 30091–30101, 2002.
359. **Zhou Z and Bornemann A.** Mrf4 protein expression in regenerating rat muscle. *J Muscle Res Cell Motil* 22: 311–316, 2001.
360. **Zhu X, Hadhazy M, Wehling M, Tidball JG, and McNally EM.** Dominant negative myostatin produces hypertrophy without hyperplasia in muscle. *FEBS Lett* 474: 71–75, 2000.