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Soft-Tissue Pathomechanics

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15.1 Soft-Tissue Pathomechanics

Although epidemiological studies have been beneficial in identifying the prevalence of musculoskeletal disorders, the demographics and job types most affected, and types of injuries most sustained, they do not address the soft-tissue injury mechanisms that result in pain, injury, and impaired function. This chapter briefly reviews the relevant and related research in soft-tissue pathomechanics of muscle. Pathomechanics is defined generally as the study of the mechanisms of soft-tissue injury that result from physical loading exposures. The study of muscle pathomechanics focuses on the effects of both short- and long-term static and dynamic muscle contractions intrinsic during physical loading on the functional and cellular changes that lead to injury, pain, and loss of function. Because the internal and external forces involved in any work-related activity act on multiple structures and tissues in the body, multiple systems often are affected. Injury, pain, and loss of function may involve damage to bone, and to soft tissues in the body, such as cartilage, tendon, ligaments, muscle, nerve, or the vasculature. Because the pathomechanics of these tissues may involve different mechanisms that often are

studied independently, separate treatments are warranted. This review will focus specifically on the pathomechanics of contraction-induced muscle injury.

The field of pathomechanics is not new; however, much of relevant basic and applied research has been conducted under more established but related scientific disciplines. For example, much of the research on skeletal muscle pathomechanics comes from the fields of muscle physiology, exercise physiology, and sports medicine where the study of the risk factors and physiological mechanisms associated with contraction-induced muscle injury have long been investigated. It is therefore a goal of this chapter to integrate this vast, diverse body of knowledge into a brief, yet helpful resource for applied ergonomists.

This chapter is organized into three sections. The first section introduces and illustrates the fundamental terms and concepts pertaining to basic muscle anatomy and physiology. The second section reviews the various methodological approaches that have been used with both human and nonhuman subjects. The third section examines the body of research that sheds light on the underlying molecular, cellular, and functional mechanisms associated with muscle injury caused by the major work-related risk factors such as force, repetition, posture, and vibration. The chapter concludes with a discussion of the current knowledge gaps and possible directions for further research.

15.2 Skeletal Muscle Physiology

Occupationally related musculoskeletal disorders have been associated with exposure to excessive physical loads, repetitive movements, awkward postures, and vibration.[24] A number of different tissues, including skeletal muscles, can be injured by exposure to these various factors.[24] To understand how exposure to these factors results in muscle injury, it is necessary to understand the biological and physiological mechanisms that allow skeletal muscles to generate movement, maintain posture, and support loads. The goal of this part of the chapter is to provide a basic description of skeletal muscle physiology and cellular biology.

15.2.1 Muscle Physiology and Anatomy

Individual skeletal muscles are comprised of bundles of muscle cells or myofibers (Figure 15.1a). Each myofiber is surrounded by a collagenous basement membrane (basal lamina) in addition to a cellular membrane called the sarcolemma. Myofibers are similar to other cells in the body, but they have a couple of unique features. First, myofibers contain a modified endoplasmic reticulum called a sarcoplasmic reticulum (SR). The SR functions as a protein processing and distribution organelle, and it regulates the levels of free intracellular calcium (Ca^{2+}) within the myofiber. Second, most of the intracellular space within the myofiber is comprised of the contractile elements or myofibrils (80% of a muscle's volume) (Figure 15.1b). Each myofibril is comprised of thick and thin filaments. Each thick filament (12–18 nm diameter) is composed of several hundred myosin proteins. Within the thick filaments, each myosin protein has a projection or a globular head (Figure 15.1c). These globular heads have binding sites that can interact with and form cross-bridges with the thin filaments, and an ATPase binding site. Thin filaments (5–8 nm diameter) are made of actin molecules that are organized in two strands twisted together to form a helix, that are covered by thread-like tropomyosin molecules and spherical troponin molecules (Figure 15.1d). Thin and thick filaments are organized in a specific pattern, which is repeated down the length of the muscle. It is this patterning that gives skeletal muscle its striated appearance.[237] Each repeated segment of thin and thick fibers forms a sarcomere.[237] The sarcomere is defined as the area between the Z-disk. Each sarcomere is comprised of dark areas (A bands) that contain the thick filaments, and light areas (I bands) that contain the ends of the thin filaments that do not overlap with the thick filaments. At each end of the sarcomere is a three-dimensional structure referred to as a Z-band or Z-disk (Figure 15.1e). Muscle contractions are produced when cross-bridges are formed between overlapping thin and thick fibers in the sarcomere, making the sarcomere the smallest contractile unit in the myofiber.

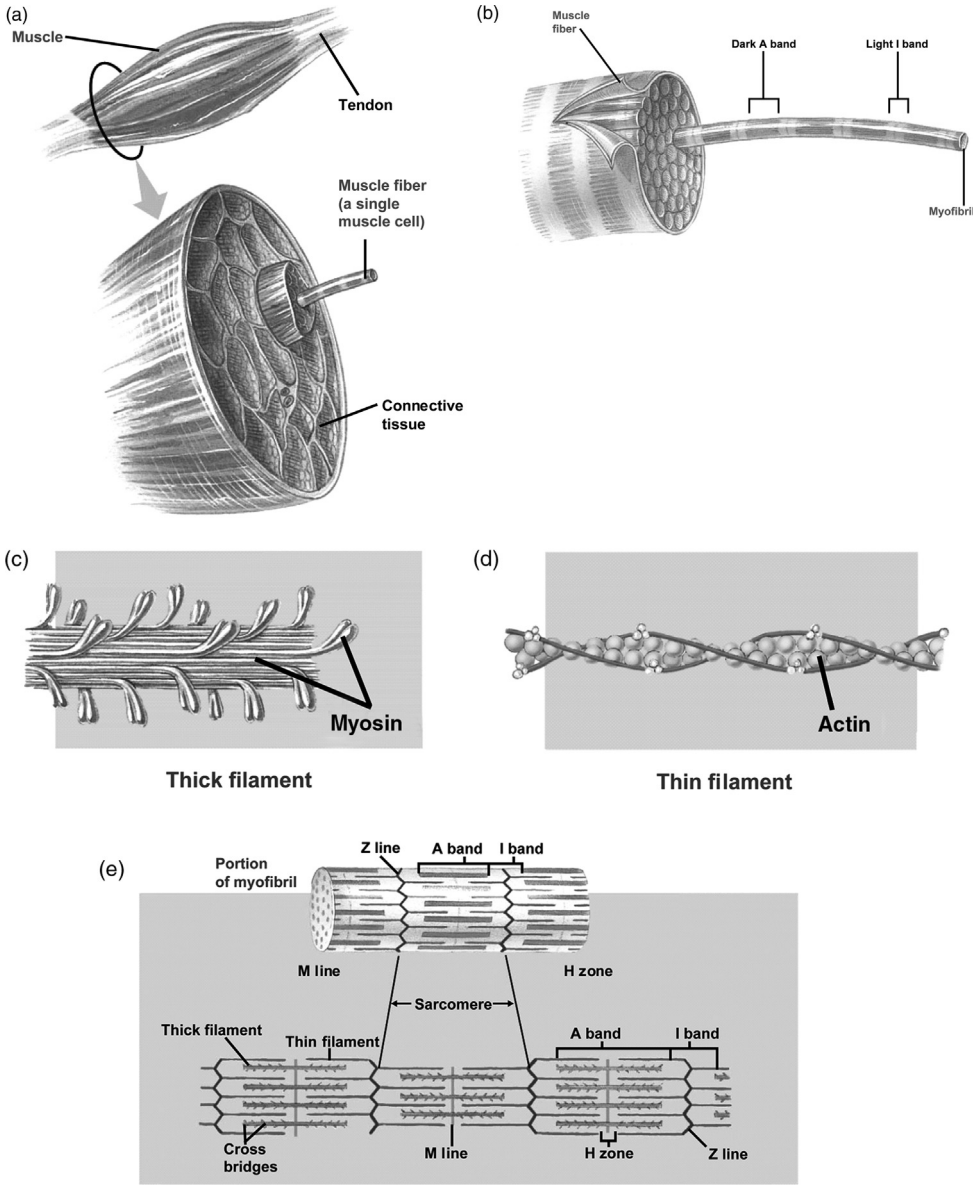

FIGURE 15.1 (a) Cross-sectional view of whole muscle and the attached tendon. (b) Enlarged view of myofibrils within a muscle fiber. (c) View of the thick filament (Myosin). (d) View of the thin filament (Actin). (e) Cytoskeletal components of a myofibril showing cross-bridge arrangement. a–e (Copyright Brooks/Cole — Thomson Learning.)

15.2.2 Cellular Processes Initiating Muscle Contractions

To initiate a contraction, myofibers must receive stimulation from motor neurons located in the ventral horn of the spinal cord. Motor neurons are considered the final common pathway where skeletal muscle activity can be governed only by input from these neurons. When activated, an action potential is propagated down the nerve axon and terminates on the neuromuscular junction (NMJ). The NMJ is a space where the action potential cannot cross from the nerve axon to the muscle fibers it innervates. Thus, a chemical messenger is used to transmit the signal from the nerve axon to the muscle fibers. As the signal is transmitted down the nerve axon, voltage-gated channels open to release Ca^{2+} into

the terminal button of the NMJ. This facilitates the release of the chemical messenger Acetylcholine (ACH) that crosses the space to the motor end plate. This causes an ionic shift, which results in the propagation of the action potential down the basement membrane of the muscle fiber and then down the T-tubules of the muscle cell (Figure 15.2). The action potential activates the voltage-gated dihydropyridine receptors in the T-tubule. This change in the T-tubules triggers the opening of Ca^{2+} release channels (ryanodine receptors) on the SR. Ca^{2+} leaves the SR through the ryanodine receptors, enters the cytoplasm, and binds to troponin, one of the proteins on the thin filaments.

Troponin has three polypeptide units; one binds to tropomyosin, one binds to actin, and a third one binds to Ca^{2+}. Under resting conditions, tropomyosin is bound to actin and it blocks the myosin-binding site on the actin protein, preventing the formation of cross-bridges (Figure 15.3). However, when free Ca^{2+} rises in the cytoplasm of a myofiber, it binds to troponin and tropomyosin is pulled away from the myosin-binding site on actin, leaving it open for cross-bridge formation. Once cross-bridges are formed, the ATPase located on the myosin head increases its activity and hydrolyzes ATP. This causes the cross-bridge to break, and Ca^{2+} then dissociates from its binding site on tropomyosin. When Ca^{2+} is removed, tropomyosin slides back into the blocking position and the muscle relaxes. Thus, troponin and tropomyosin are referred to as regulatory proteins in muscle contraction (Figure 15.3).

15.2.3 Force Generation and Transmission in Skeletal Muscle

One of the main functions of skeletal muscle is to generate and transmit force. Force, or muscle tension, is directly related to the number of actin and myosin cross-bridges that are formed and the frequency of stimulation. A single action potential results in a single muscle contraction referred to as "twitch." As the frequency of stimulation increases, the resultant twitch tension (Figure 15.4a) increases with increasing stimulation frequency (Figure 15.4b) until a force plateau results (Figure 15.4c). Force is produced at each attached cross-bridge, so the total force development is proportional to the number of attached cross-bridges. The number of cross-bridges that can be formed depends upon the degree of overlap between the thin and thick filaments (Figure 15.5). When a sarcomere is overstretched or compressed,

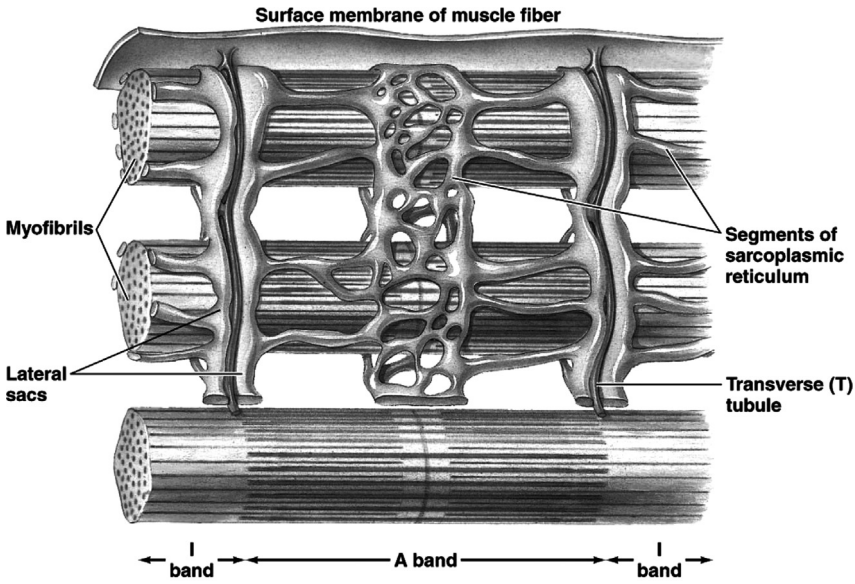

FIGURE 15.2 The T-tubules and SR in relationship to the myofibrils. (Copyright Brooks/Cole — Thomson Learning, 2001.)

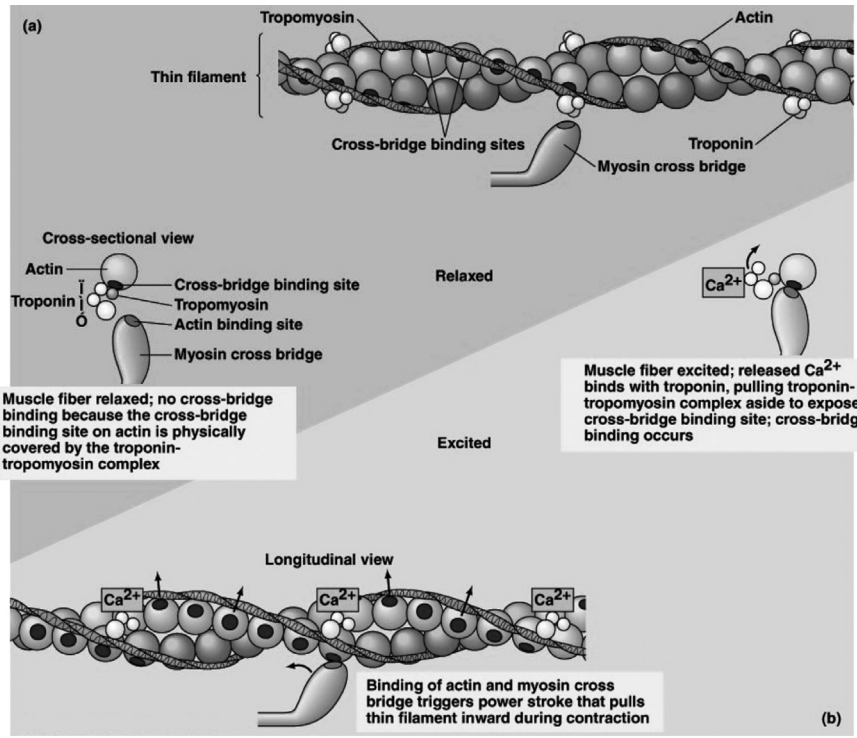

FIGURE 15.3 The role of Ca^{2+} in activating the cross-bridges. (Copyright Brooks/Cole — Thomson Learning, 2001.)

the area over which thin and thick filaments overlap is reduced, and thus there is a decrease in the number of cross-bridges that can be formed resulting in a reduction in force (Figure 15.5). Thus, maximal force is generated when sarcomeres are at a length that produces the optimal overlap between thin and thick fibers.

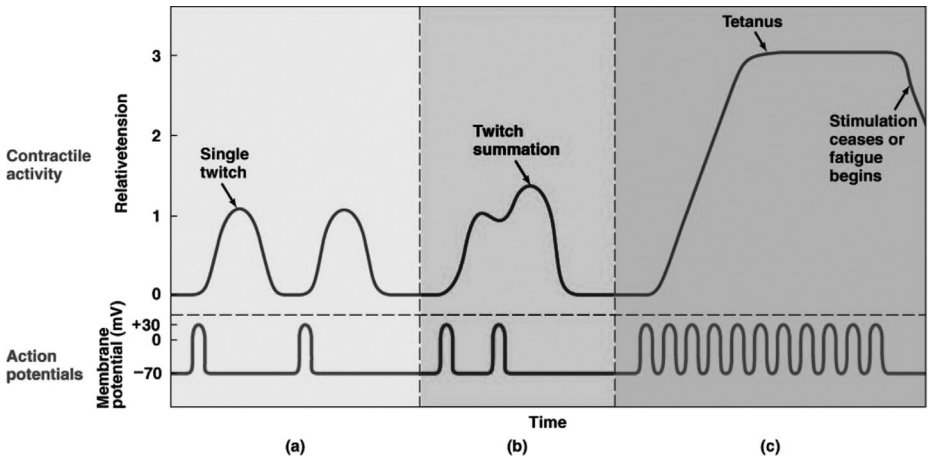

FIGURE 15.4 Muscle twitch, twitch summation, and tetanus. (a) If a muscle fiber is restimulated after complete relaxation, the second response is the same as the initial response. (b) If the muscle fiber is restimulated before complete relaxation takes place, the second twitch is added to the first twitch. (c) If the muscle fiber is stimulated rapidly such that it does not have the opportunity to relax, a maximal contraction or tetanus occurs. (Copyright Brooks/Cole — Thomson Learning, 2001.)

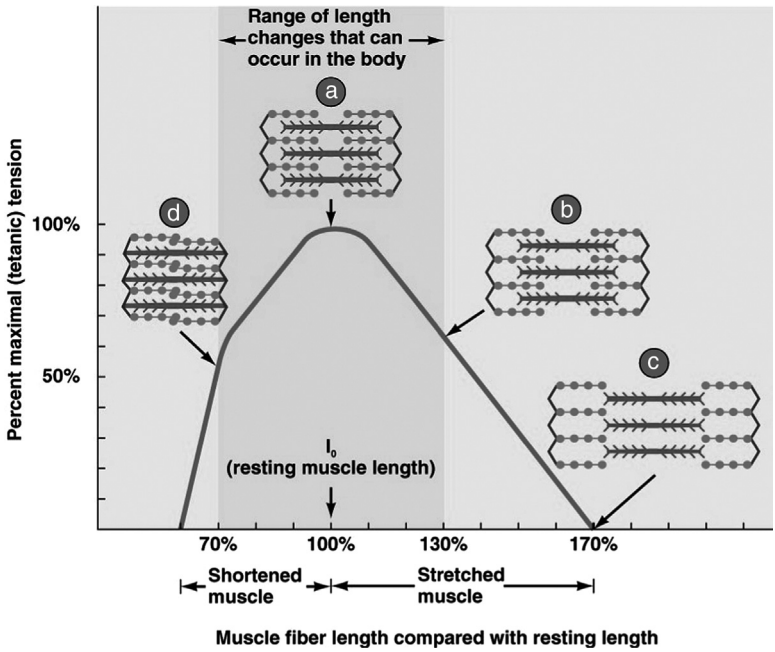

FIGURE 15.5 Length–tension relationship of muscle. At point A optimal overlap of thick and thin filaments results in maximal tension developed. This is referred to as the normal resting length in the body (l_o). As the muscle lengthens (point B), less cross-bridges are attached, which results in a decrease in tension. Further increases in length correspond with less cross-bridge attachment and further declines in tension (Point C). The response from point A to point C is usually referred to as the descending limb of the length–tension relationship. If shortening occurs at less than l_o, fewer filament-binding sites are exposed to filament cross-bridges, thus tension decreases (Point D). (Copyright Brooks/Cole — Thomson Learning, 2001.)

Force is generated at the cross-bridges, but it is transmitted longitudinally and radially along myofibrils. The longitudinal transmission of force occurs down the thick myosin filament to the Z-disk, and on to the next serial set of myofibrils. Two proteins titin and nebulin, maintain length registry of the sarcomere and aid in axial transmission of contractile forces. The actions of titin and nebulin maintain registry of the A-band with the Z-disk, which is important for sarcomere integrity. Nebulin maintains length registry of the thin filaments[49,306] by interacting with tropomyosin and troponin to form a lateral network with actin to regulate thin filament length. Titin functions as a two part spring to transmit force from the thick filaments to the corresponding Z-disk.

Radial forces are transmitted via lateral stabilization of adjacent myofibrils. The protein responsible for maintaining lateral registry of adjacent myofibrils at the Z-line is desmin.[160] The Z-disk structure is thought to be three-dimensional in nature and comprised of the proteins desmin, actin, and α-actinin. The radial enclosure of these three proteins also extends longitudinally along the myofibrils to provide both radial and longitudinal stability.[207] These proteins are thought to be anchored to the Z-disk via intermediate filament-associated proteins (IFAP). The cytoskeletal lattice extends radially from the Z-disk to the sarcolemma via the transmembrane proteins. The transmembrane proteins are thought to anchor the myofilaments to the sarcolemma via focal adhesions.[205] These adhesions or "costameres" are made up of a variety of transmembrane proteins. The basement membrane is then attached to the sarcolemma via the dystroglycan complex.[205,207] Radial transmission of forces occurs through structural proteins located inside and outside of the sarcomeric region via the intermediate filament network, and to the sarcolemma via the transmembrane proteins.[207] Capability of radial force transmission is necessary for redundancy in case of fiber injury. Thus, force can be transmitted

in any direction in relation to the axis of the muscle fibers via endosarcomeric and exosarcomeric protein lattices.

15.2.4 Types of Muscle Contractions

There are three primary types of muscle contractions. These contraction types are distinguished by how the muscle length changes during the contraction.[13,48] Isometric contractions are defined as muscle activity where tension is generated without a change in length. This is also referred to as a static contraction where muscle is generating tension, but does not result in a change in length, and thus, there is no segmental (about a single joint) or whole body motion. Shortening contractions (often referred to as concentric contractions[13] are defined as the muscle generating tension while getting shorter. Concentric contractions usually generate segmental or whole body motion. Lengthening (or eccentric) muscle contractions are defined as the muscle generating tension while the muscle is lengthening. Lengthening contractions are usually used to absorb work or energy, thereby applying braking to segmental or whole body motion.

During concentric muscle actions, the tension varies as a function of shortening velocity where tension decreases as shortening velocity increases. The "force–velocity" relation is hyperbolic and also depicts that maximum shortening velocity occurs at zero load and zero velocity occurs at maximum isometric force.[295] During isometric muscle contractions, it is well understood that force varies as a function of muscle length. It has been shown that muscle tension is lowest at very short and very long muscle lengths and develops higher tension in the intermediate lengths.[68,103] This is due to the degree of sarcomere overlap in the cross-bridge. Thus, the length–tension curve has an ascending and descending limb as length increases (see Figure 15.5). The ascending limb, defined as the increase in force with increase in length, is due to more actin-binding sites being available to bind with the myosin filaments. As tension plateaus, this is thought to be due to all the actin-binding sites being bound to the myosin filaments. The descending limb, defined as the decrease in muscle tension with increasing length, is due to less actin-binding sites being available as the actin filaments are pulled out of register with the myosin filaments. Thus, the length–tension relationship of muscle is due to myofilament overlap in the sarcomere (as shown in Figure 15.5).

It is now well known that muscle can generate more tension during eccentric muscle actions than during concentric or isometric contractions. This was first reported in a study involving human muscles under volitional control.[238] It is also interesting that while muscles generate more tension during eccentric muscle actions than concentric muscle actions, EMG activity is less in muscles during stretch than during shortening at the same tension. During maximal effort, the EMG signature remains constant and force varies due to the length–tension relation of that specific muscle or muscle group, however force during volitional eccentric activity never exceeded 140% of maximal shortening forces.[148] In animal studies that employ electrical stimulation to activate the muscle of interest, forces of 180% of maximum isometric force are typical.[284] High eccentric forces in humans with spastic paresis have been attained to levels similar to those seen in animal studies.[142] In addition, if muscles in humans are stimulated by external electrical stimulators, as in the case of spinal cord injured patients, the external forces generated during eccentric muscle actions are nearly 200% of the forces generated concentrically using the same electrical stimulation paradigm.[275] Thus, exogenous electrical stimulation overrides the inhibitory influences that moderate muscle output force.

Stretch-shortening cycles (SSC) are a type of muscle action that incorporates both concentric and eccentric muscle actions. In most sports-related activities, it involves a prior stretch before shortening to enhance the shortening phase of the movement. Activities that typically use SSCs are jumping, walking, running, and movement in and around obstacles. In occupational-related activities, it is mostly related to reciprocal lifting and lowering activities and repetitive lift and carry tasks. It is an excellent model to study physiological muscle function.[149] It also allows for simultaneous study of concentric and eccentric muscle function and their synergism.

15.2.5 Musculotendon Actuator

Muscle and tendon have typically been studied in isolation although they function synergistically. Their integrative function has been defined as the musculotendon actuator.[311] While physiologists have long recognized that muscle and tendon act in a synergistic fashion, they have studied those tissues in isolation to better understand the function of muscle and tendon separately. The musculo tendon actuators interact with body segments to produce movement and the dynamics of movement are dependent upon the contraction dynamics of the actuator. This system also functions as a feedback loop where the dynamics of body segments affect the force output of the actuator via the length and velocity of the actuator.[311]

Tendon compliance affects the contraction dynamics of the muscle. In actuators with highly compliant tendons, a length change of the actuator would be mostly realized by the length change in the tendon, with very little concomitant length change in muscle. Compliance of an actuator is defined by the ratio of tendon slack length to muscle fiber length. Muscle length changes will be commensurate with length perturbations of the musculotendon actuator if the actuator is stiff. However in compliant actuators, muscle length changes will not follow exogenous length perturbations. This is quite relevant since changes in muscle length are rarely measured directly, indeed in human studies of muscle function the kinematics of the musculotendon are measured. The assertion that changes in musclotendon length are representative of muscle length changes may be incorrect, particularly for actuators with highly compliant tendons.[311] The active and passive force–length relation of muscles that have been published to-date may be erroneous due to the fact that muscle fibers are at different lengths in the active versus passive state even though the actuator is at the same length. This is due to differing amounts of tendon stretch, which are caused by different forces exerted by the muscle fibers. Thus, the stretch of the tendon must be accounted for to accurately represent muscle stretch.[118] The muscle–tendon interface (at the aponeurosis) exists in a state of dynamic equilibria, where force transients are equalized via stretch of the tendon and muscle activation and muscle length change. The dynamic equilibria are also governed by the response time of the tendon and muscle, which are often different. Actuator compliance varies depending on the muscle group and animal species. In humans, actuator compliance appears to be highest on the plantar flexor group and lowest on proximal groups, such as biceps and triceps.[31,128,293] In summary, one must think in terms of the musculotendon when investigating *in vivo* muscle function or reviewing scientific studies of *in vivo* function. While most studies refer specifically to muscle function, the measurements are typically made on the musculotendon group. Thus one must be cognizant of the influence of tendon mechanics on muscle function and the musculotendon unit.

15.3 Contraction-Induced Injury Models

15.3.1 Human Studies

In the past four decades, there has been substantial inquiry into the physiological effects of eccentric exercise on physiological responses, such as oxygen consumption,[29,140,143,145,147] muscle metabolites,[32,33,208] and performance via bicycle ergometry or dynamometry[28,78,115,144,146,150,151] just to name a few. However, only in the past two decades there has been an increasing amount of interest in the area of skeletal muscle injury. Formal inquiry in the area of muscle injury was initiated by Hough in 1902[126] where it was noted that delayed onset muscle soreness resulted days after the exposure and probably was due to microtears in the muscle tissue. It is interesting that there was not much scientific inquiry in the area of muscle injury until the early 1980s when Friden et al.[90] were one of the first to provide evidence of muscle fiber damage after exercise. Fortunately, the study of muscle injury has rapidly increased since then.

It has been well documented that muscle injury primarily results from lengthening contractions but not concentric or isometric contractions.[53,58,253] Thus, studies of muscle damage have employed eccentric-biased segmental exercises using either isolated muscle groups, such as the elbow

flexors,[193–197,221,222,231] knee flexors,[173,174,294] hamstring,[179] pectoralis and anterior deltoids (chest press),[245] and calf and biceps.[136] Whole body movements such as downhill stepping exercise,[187,189] downhill running,[199] or cycling[175] have also been used to create muscle injuries. The amount of acute resistance for most human studies ranged from 10 to 180 repetitions for the segmental exercise studies, and as little as 20 min of downhill stepping exercise[187] to produce muscle soreness and evidence of myofiber injury. Thus, even a low number of repetitions or short exposure to lengthening contractions can result in a strain injury.

It is interesting to note that exposures that involve both whole body movements and segmental loading result in muscle damage, loss of force, and delayed onset muscle soreness. Different types of information can be attained from these two types of exposures. Whole body exercise typically involves closed kinetic chain movements where the level of exercise (treadmill or cycling speed, metabolic load, angle of inclination) is controlled but muscle forces or torques, velocity, range of motion, and number of repetitions are not typically measured. In contrast, most segmental exercise models are open kinetic chain movements involving isolated loading of the limbs, and are administered by either isokinetic or isotonic dynamometry (computer-controlled strength testing equipment that operates in either constant velocity or constant torque mode), or by isoinertial loads (using either free weight or weight attached to an apparatus). Joint torques or forces, as well as velocity, range of motion, and number of repetitions are measured.

Prolonged strength loss, as measured by maximal isometric force, is considered to be the best method to quantify the degree and time course of muscle injury and recovery after exposure to damaging lengthening contractions.[289] It is also the primary means to quantify muscle function in humans where muscle function is defined as the ability of the muscle group of interest to generate force over a prescribed range of motion, or fixed length at a given level of muscle activation.[289] Other measures, such as biochemical markers in the blood and urine, and level of histological damage as obtained from biopsy, are not well associated temporally with functional performance.[289]

Muscle injury studies with humans have been very beneficial in elucidating the type and intensity of exercise and muscle actions that produce injury. Those studies have also been beneficial for examining the resultant myofiber changes after injurious exposure and the recovery time after injury. The corresponding levels of pain perception and muscle soreness after exposure was also consonant with the degree of performance deficit.[58] However, though many studies of muscle injury with humans have been conducted in controlled laboratory settings, some experimental questions cannot be fully addressed using human subjects. Confounding factors such as lifestyle, level of psychological stress, pre-existing disease states such as diabetes and hypertension, and genetic polymorphisms are difficult to control. In addition, key issues such as the amount of fiber strain and muscle biomechanics necessary for injury, the amount and site of resultant muscle injury from longitudinal and cross-sectional tissue analysis, the effect of structural protein knockouts and inflammatory mediator blockade on muscle injury and repair, and biochemical analyses of muscle tissue, is either difficult or impossible with human subjects.

Animal models have been developed that reduce or eliminate these confounding factors as well as provide for control of the degree of motivation and muscle activation strategies. In depth physiological questions about the pathways involved in muscle injury and repair, the site and extent of injury, the role of structural proteins in muscle force transmission and injurious response, and biomechanical loading signature necessary for injury can be more easily addressed in animal models. Animal models that have been developed allow for more controlled study from the level of isolated muscle fibers to fully intact muscle groups that contain intact neural and vascular supplies.

15.3.2 Animal Models of Muscle Performance and Injury

Animal models provide a good platform to study the physiological responses to injurious and noninjurious muscle contractions and the biological aspects underlying muscle injury and adaptation. The majority of animal models used to investigate skeletal muscle performance and injury have used

rodents. The majority of studies of acute injury in animals have focused on the temporal force response of muscle to a single exposure (1 to 1800 repetitions) of high force isometric (muscle generating force at a fixed length), concentric contractions, or lengthening contractions.[13,35,36,39,72,74,75,129,161,163–165]

The similarities between the micro-architecture of rodent and human skeletal muscle and the ability to precisely control the biomechanics of contractile activity in rodents through various *in vitro*, *in situ*, or *in vivo* methods, as well as the ability to investigate in depth physiological questions such as injury pathways, are major advantages using animal preparations. Each type of animal model has inherent advantages and disadvantages for the study of muscle injury. The models that have been used to investigate muscle pathomechanics are described here.

15.3.2.1 *In Vitro* Models

In vitro models use muscle that has been excised from an animal to study muscle function. The muscle group or muscle fibers are placed in a sealed physiological bath and activated by plate electrodes attached to an electrical stimulator. The resultant muscle tension is measured at the ends of the muscle by ergometry using strain gage force transducers. Length changes also are produced via the attached ergometer and invoked after maximal activation of the whole muscle or isolated muscle fibers. The target muscles studied were mouse soleus,[167,259,286,290,283] mouse extensor digitorum longus (EDL),[36,210] toad sartorius,[258,260] rat soleus,[171,172,284,285,309] and mouse fifth toe muscle.[226]

The main advantage of *in vitro* preparations is the ability to study a single muscle group or isolated muscle fibers of interest without the confounding effects of adjacent or antagonist muscle groups. In addition, exact length changes and the velocity of length changes of the total muscle and individual sarcomeres can be accurately measured in real time. The release of muscle proteins and enzymes into the physiological bath can also be detected that may be indicative of injury or metabolic fatigue. Measures of performance, such as isometric twitch tension, maximal isometric force, work done during stretch or shortening actions, and power absorbed during stretch or produced during shortening can be quantified. Much information regarding muscle function and injury mechanics has been attained using *in vitro* models.

There are several shortcomings of *in vitro* preparations. Because the muscle has been excised from the animal and tissue viability is time-limited, *in vitro* preparations are only suited for single exposures. Thus, the effect of repetitive exposures on muscle response, adaptation, and injury cannot be studied. In addition, the effect of changes in muscle performance on biomechanics about the joint axis cannot be examined. Also, the effect of muscle synergists on performance about the joint axis cannot be studied. Because human muscle testing is performed about the joint axis of interest with functioning muscle synergists, generalizing from *in vitro* results to *in vivo* function can be difficult. Also, because normal neural and vascular supplies have been ablated, muscle performance, response to injurious perturbations, and changes in neural recruitment patterns can be affected. For example, an intact vascular supply is instrumental in replenishing depleted energy stores and removing toxins, and bringing in appropriate cellular infiltrates for muscle repair and remodeling. *In vitro* models by nature are intrinsically ischemic due to the absence of an intact vascular supply. An intact neural supply is beneficial for proper muscle activation and to facilitate the study of neural changes in response to injury and repair. Other models that maintain intact neural and vascular supplies and connections with supporting tissues can provide data that is more physiologically representative about intact muscle function.

15.3.2.2 *In Situ* Models

In situ models address the need for muscle injury models that have the advantages of the *in vitro* models but also have normal neural and vascular supplies intact, and the ability to test whole muscles or muscle groups. Typical *in situ* models involve surgical ligation of the distal tendon of the muscle or muscle group of interest leaving the neural and vascular supplies intact. The target muscle or muscle group is stimulated via the exposed nerve or by use of percutaneous electrodes. Measurement of muscle contractile forces and control of length changes is usually via attachment of the distal tendon that was surgically ligated to a computer-controlled servomotor and load measurement transducer. This model of

contraction-induced injury provides a more physiologically representative preparation than the ischemic, noninnervated *in vitro* model.

In situ models have been used to study injury mechanics in mice EDL muscles,[35,37–39,65,72,75,129,177,178,313] rat EDL,[176,276–278] rat adductor longus,[268,281] rabbit tibialis anterior (TA),[27,161,162,165,191,192,201,206] rabbit soleus muscle,[23] rabbit EDL,[219] and rabbit triceps surae.[264] The parameters used to produce injury in those studies ranged from single stretch models that consisted of stretch outside of the typical physiological range (usually greater than 130% of the resting or optimal muscle length, l_o) of the muscle[27,38,39,129,191,192,201,219] upto 1800 contractions within the normal physiological range (typically 70% l to 130% l_o).[161,165]

In situ models are well suited for acute muscle injury studies where changes in contractile forces subsequent to an injurious perturbation are of interest. Exact length changes and lengthening velocity of the muscle–tendon group can be controlled and monitored during testing. However, the invasive nature of this model precludes it from being applicable to repetitive injury models because the target muscle–tendon complex cannot be left exposed for more than a single session. Also, in most *in situ* studies, the forces or torques are not tested about the normal joint axis where effects of synergist muscles will have an effect on the resultant forces or torques about the joint axis. Also, the transmission of muscle–tendon forces through the joint axis to the target output limb via the mechanical advantage of that joint could not be studied. However, results from *in situ* studies have provided much information about the causal factors in acute muscle injury and the resultant physiological responses.

15.3.3.3 *In Vivo* Animal Models

It was apparent from *in vitro* and *in situ* findings that it would be beneficial to investigate muscle response and injury mechanics by testing about the normal joint axis of the target muscle and also be concerned with the invasiveness of the procedure. By using a noninvasive procedure, the confounding effects implicit in the required surgical procedure of *in situ* preparations are removed, and the temporal response after exposure can be examined. Also, a noninvasive preparation would be ideally suited for the study of muscle response from repeated exposures. *In vivo* models address these issues by facilitating testing about the normal joint axis in a noninvasive manner, with intact neural and vascular systems, and intact muscle–tendon systems.

Most *in vivo* models can be categorized as either volitional or nonvolitional models. Volitional models are those in which the movement tasks are performed voluntarily using different types of motivational tools. In contrast, nonvolitional models are those in which the animal typically is anesthesized and muscle contraction is initiated and controlled by an external electrical source.

15.3.3.3.1 Nonvolitional Models

In order to fulfill a need for more control and quantitation of *in vivo* muscle function, Wong and Booth in the late 1980s and early 1990s used electrical stimulation of the rat plantar flexors and a weighted pulley bar apparatus that would provide isotonic resistance to the plantar flexors.[302–304] This approach controlled the number of repetitions, the activation of the muscle group, the temporal arrangement of the repetitions, and loading of the plantar flexors about the joint axis. However, this model did not use a servomotor to control the range of motion or velocity and acceleration of the movement, and did not measure dynamic forces of the plantar flexors.

This approach was refined in the early 1990s to provide better control of the kinematics of the movement by use of an electrical servomotor. *In vivo* dynamometry incorporated electrical servomotors, load cells to measure forces, and potentiometers and tachometers to measure the kinematics of the movement to comprise a total testing system[63] (Figure 15.6). Dynamometry can be used to control and measure the biomechanical loading signature in real-time via control of muscle activation levels, range of motion of the muscle action, type of muscle action (isometric, shortening contractions, lengthening contractions, or stretch-shortening), velocity, acceleration, number of repetitions, duty cycle, and exposure duration.[63] The main difference between dynamometry and the Wong and Booth model is that the electrical

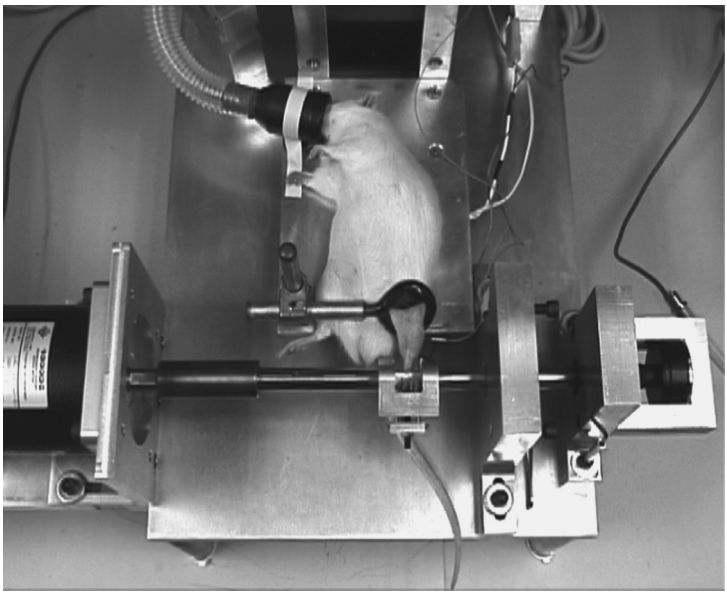

FIGURE 15.6 *In vivo* dynamometer with an anaesthetized rat on the heated $X-Y$ positioning table. The left foot is secured in the load cell fixture and the knee is secured in 90° flexion.

servomotor (in the dynamometer) controls the movement kinematics and measures the resultant muscle response during those movements. In the Wong and Booth model, the muscle forces produced the shortening-only movement about the joint axis where the kinematics was not controlled.

The first reported *in vivo* rodent dynamometer was developed by Ashton-Miller for the study of biomechanical behavior of the plantar and dorsi flexor muscles of the mouse hindlimb.[12] This approach also has been used to study rabbit dorsiflexors,[26,86,163,164,206] rat dorsiflexors,[62,97,116,152,154,156,157] mouse dorsiflexors,[132,168,283] and rat plantar flexors.[63,296–299] Typical exposures ranged from 20 repetitions of the rat plantar flexors[298] to 900 repetitions in the rabbit dorsiflexors.[86,163,164,206] Typical angular velocities were based on the animal being tested: 75 deg/sec for rabbit dorsiflexors,[163] upto 500 deg/sec for rat dorsiflexors,[97,152] and upto 2000 deg/sec for mouse dorsiflexors.[132,168,283] Ranges of angular velocities were selected based on the volitional capability of the muscle group and animal species in order to be physiologically representative.

The major benefit of nonvolitional *in vivo* models is the ability to study muscle function and injury mechanics about the joint axis of the target muscles. Thus, the normal muscle, tendon, and bone attachments are intact as well as the neural and vascular supplies. The synergistic function of muscle agonists and lateral transmission of adjacent muscle forces is also preserved.

One major limitation of nonvolitional models is the use of artificial electrical stimulation to invoke muscle contractions. Unlike voluntary contractile activity, which is submaximal and characterized by a selective recruitment of motor units, nonvolitional contractile activity is typically supramaximal because electrical stimulation involves the activation of all motor units of the target muscle. Thus, caution must be exercised when making inferences from comparisons between muscle responses from supramaximal electrical stimulation and voluntary submaximal contractions.

15.3.3.3.2 Volitional Models

Volitional *in vivo* models represent a more physiologically representative animal model for the study of muscle injury and adaptation. Volitional models differ from nonvolitional models in that normal muscle recruitment is employed via normal central nervous system control, and the pace of the activity is controlled by the animal, not the testing equipment or the investigator. One of the earliest reported

models of volitional lifting was developed by Gordon in 1967 using weights attached to the back of rats during vertical crawling and other exercises.[104–107] This type of work was furthered by Stone et al.[261,262] and Ho et al.[120] in rats. Gonyea et al. extended this model to cats, also using weight lifting exercise.[100–102] Weights also have been added to wing muscles of chickens, roosters, and other birds to produce an overload model designed to study the skeletal muscle response to persistent overload.[3,83,166,246]

Animal treadmill models were developed in the early 1980s as a way to invoke voluntary repetitive eccentric muscle actions capable of producing muscle injury.[10,234] It was found that downhill treadmill locomotion produced an eccentric bias on the soleus muscle in the plantar flexor group that resulted in distinguishable signs of injury.[10,66,202,234] Treadmill exposure has been used to study a wide variety of physiological variables and typical exposures range from 30 to 150 min per session for a single session in rats[10,66,153,202,277] upto five sessions per week for 10 weeks duration,[16,109] and upto 9 h exposure in a single session for mice[185] to study acute injury response and adaptation and reduction of injury susceptibility after repetitive exposure. The treadmill studies are similar to those conducted in humans although the animal exposures are typically longer in duration.

In volitional treadmill and resistance training models, the exposure biomechanic, such as muscle forces or torques, or the number of muscle contractions (repetitions) are not controlled or quantified during the activity. This lack of quantitation makes it difficult to relate physiological outcomes to specific parameters of performance and loading history, which can differ widely across individual animals.

Some researchers have employed operant conditioning procedures to produce the kinds of repetitive muscle loadings that are relevant for the study of exercise-induced physiological responses. In these approaches, voluntary responses were motivated by various consequences, such as food rewards,[102,141,308] intracranial stimulation,[94] or electric shock to the tail or feet.[71,84,120,266] The species, target muscle groups, and training protocols, however, differed widely among these models. For example, Gonyea et al.[98,101,102] trained cats to grasp and move a weighted bar with the forelimb repeatedly in 30-min sessions conducted 5 days per week for upto 87 weeks. Barbe et al.[19] trained rats upto 8 weeks to repeatedly reach their forelimbs into a small tube to retrieve food pellets. Yarasheski et al.[308] trained rats over an 8-week period to climb a wire-mesh ladder with weights secured to their tails, and Klitgaard[141] trained rats over a 36-week period to enter a vertical tube and use their hindlimbs to lift a weighted ring. In other approaches, rats wore weighted jackets and were trained in sessions conducted 8 to 16 weeks to rear up on their hindlimbs to avoid an electric shock[71,120,266] or to receive brain stimulation.[94]

Many of these approaches have been developed for the study of adaptive or regenerative processes. For example, there is considerable evidence that under some conditions, voluntary repetitive exertions performed over several weeks or months can lead to muscle hypertrophy as evidenced by increases in either myofiber number or size.[71,101,266,301] Under other conditions, however, similar patterns of repetitive exertions have resulted in degenerative morphology[98] and inflammatory responses.[19] Unfortunately, many of these approaches lacked the necessary control and quantitation of the biomechanics of the movements to allow for thorough assessments of external and internal loadings that would be necessary to characterize dose–response relationships. In addition, few studies have specifically examined the effects of external loads on both internal loads and tolerances (as measured by biomechanical performance and physiological force tremor) and physiological or biochemical processes. Thus, little is known about the relation between specific parameters or changes in performance or physiological force tremor and physiological or biochemical processes.

15.4 Injury Mechanisms

15.4.1 Acute Muscle Injury

Skeletal muscle is a unique tissue in the body since it has both passive and active properties that are exhibited during muscle contraction. Because skeletal muscle can generate force during contraction

for movement of the limbs and external work, as well as the absorption of work, it can produce loads on other tissues such as tendons, joints, and nerves. There have been extensive studies to date on acute contraction-induced muscle injury using both animal and human models. Studies of soft-tissue injury resulting from acute strain overload have been conducted using animal and human models. Indeed, a number of studies on contraction-induced injury have been conducted in rodents and rabbits. These studies have used *in vivo, in situ*, and *in vitro* preparations to investigate muscle injury.

15.4.2 Eccentric Muscle Actions in Acute Myofiber Injury

Eccentric contractions are known to cause a greater amount of damage in muscles. This suggests that high load tensions in fibers may be more important than physiologic considerations in the etiology of the injury process.[7,253,254] High mechanical forces produced during eccentric muscle actions have been causal in the underlying etiology of muscle strain injuries.[10,284,285] This was thought to be due to high fiber stresses in the contractile apparatus due to high forces transmitted axially to the contractile proteins. High mechanical forces produced during muscular contractions, particularly in eccentric exercise, where forces are distributed over relatively small cross-sectional areas of muscles, cause disruption of proteins in skeletal muscle fibers and connective tissues.[6,11] Eccentric contractions have been shown to result in ultrastructural damage immediately after exposure,[87] and 1–3 days after exposure.[116,178] The extent of histological damage is difficult to quantify by light or electron microscopy immediately after injurious exposure because only single sarcomeres or small groups of scattered sarcomeres are affected.[39] Damage is accompanied by a loss of contractile force with visible interfiber damage.[11] The reduction in contractile force is temporary (lasting days) and is accompanied by muscle soreness.[8] The isometric force deficit, a functional measure defined as the difference in isometric force before and after an eccentric contraction protocol, has been shown to be the best indicator of the magnitude of contraction-induced injury.[75,190,289]

Most work in eccentric contraction-induced injury has been done on small mammals *in situ*. To determine which parameters were responsible for muscle injury, some of the early studies focused on comparing muscles that were passively stretched through a range of motion to muscles that were actively stretched through the same range of motion. Muscles that were exposed to 30 min of passive stretches (TA and EDL) showed no loss in force; however, force loss was evident in the group that underwent 30 min of lengthening contractions of the same muscle group.[73,74] The role of passive stiffness was investigated in rat TA *in situ*, and stiffness increased after eccentric and isometric protocols; however, there was no correlation between injury and increased passive stiffness.[155] McCully and Faulkner[177] used mouse EDL muscles and found that the force decrement due to eccentric contractions did not recover and thus was due to mechanical insult to the tissue. Those results were corroborated by Lieber and Fridén[161] using New Zealand white rabbit TA muscle *in situ* and by Van Der Meulen,[276,277] using rat TA muscle *in situ*. Initial muscle length in conjunction with work input was also determined to be a factor in eccentric contraction-induced injury based on *in situ* single stretches to 170% L_o ("L_o" denotes the length of a particular muscle where it generates the highest force).[129] In contrast, Warren et al.,[284] observed no difference in force deficit due to initial length although L_o was less than 100%. Thus, results from *in situ* studies indicated that eccentric contraction-induced injury results from high mechanical forces in conjunction with the muscle being at long fiber lengths on the descending limb of the length–tension relationship.

The results of studies conducted using *in vivo* models of humans and animals have been consistent with *in vitro* and *in situ* results. Armstrong[10] and Ogilvie[202] used rodent treadmill testing to investigate eccentric contraction-induced injury. The purpose of these studies was to investigate the relationship between eccentrically biased treadmill exercise and skeletal muscle injury. The response of the concentrically contracting TA was compared to the eccentrically contracting soleus muscle. The control group (no exercise) and the concentrically contracting TA muscle showed no signs of injury but the eccentrically contracting soleus muscle did result in fiber injury. Newham[186] used human subjects and stimulated elbow flexors superimposed over maximum voluntary eccentric contractions to assure that muscle

contractions were maximal. The results of their study showed that the loss of force was due to changes in contractile elements, not the level of muscle activation. This was the first study to suggest that the force decrement resulting from eccentric muscle actions is not the result of less muscle activation, but instead may have a mechanical etiology. Thus, the force deficit seen after eccentric contraction-induced injury in both humans and animals is due to damage of the contractile proteins and supporting structures, not central nervous system activation level.

Once eccentric contractions were identified as causing muscle injury, it was important to investigate how the injurious response could be modified by mechanical exposure factors (e.g., force, strain, strain rate, number of repetitions, and velocity). The primary factors that have been studied have some generalizability to occupational physical exposures.

15.4.3 Factors Affecting Acute Muscle Injury

15.4.3.1 The Effect of Muscle Force on Myofiber Injury

The *in situ* results of eccentric contraction-induced injury have been supported in general by *in vitro* animal models. Warren[285] used isolated rat soleus muscles to investigate mechanical factors associated with the initiation of eccentric contraction-induced muscle injury. The results indicated that a reduction in contractile force was most related to high forces during lengthening. This finding was also supported by other work.[110,178,253] In Warren's model, the primary criterion used to quantify injury was a reduction in twitch tension (P_t), which has previously been shown in muscles injured by eccentric contractions. The eccentric contraction group was compared against muscles performing isometrically using the same stimulation protocol. This study clearly demonstrated that eccentric contraction-induced muscle injury has a mechanical etiology. The predominant factor was mechanical force during lengthening with failure occurring above 113% P_o ("P_o" denotes maximum isometric force of the target muscle). In a follow-up study by Warren,[284] the focus was to investigate whether injury is the result of high tensile force after one contraction or the result of multiple contractions. The protocol consisted of 0.25 L_o excursions with a velocity of 1.5 l_o/sec at a force of 180% P_o. Muscles performing more than eight eccentric contractions resulted in injury since marked force decrements were observed after the eighth contraction. This suggests that it requires more than one repetition to result in myofiber injury within the physiological range.

15.4.3.2 The Effect of Muscle Length Changes on Myofiber Injury

To determine if muscle strain was important in susceptibility to injury, Zerba and Faulkner[314] studied mouse EDL muscles *in situ*. Isometric force was checked immediately post-test and 3 days after the protocol. Only muscles stretched to 75% L_f (length of the muscle fiber) at L_f/sec produced injury 3 days later. Muscles stretched at lower velocities and fiber strain did not exhibit any signs of injury. They hypothesized that strain and strain rate were synergistic in the etiology of muscle injury. Lieber and Fridén[162,165] also investigated whether muscle damage was a function of muscle force or muscle strain. Rabbit TA muscle was used *in situ* via securing the distal tendon to a servomotor. The TA muscle was selected due to a 30° pennation angle and negligible angular rotation during stretch. Final results indicated that muscle strain (change in length of the muscle) produced the most profound changes in muscle performance (via a force decrement) and that muscle strain was more responsible than force during lengthening in producing contraction-induced injury. Warren et al.[284] found no observed difference in force deficit when stretches were initiated from either 85% L or 90% L_o. In contrast, Hunter and Faulkner,[129] MacPherson et al.,[172] and Brooks and Faulkner[39] found that 30% strain was necessary to produce a force deficit after a single stretch and larger force deficits resulted when stretches were initiated from a longer initial length, or terminated at a longer final length. This finding was also supported in multiple repetition models.[110,305] Thus, it appears from both single- and multiple repetition models that muscle length during stretch has an impact on the resultant force deficit and myofiber injury. However, the effect of muscle length repetitive exposures of eccentric muscle actions has not been investigated to date.

15.4.3.3 The Effect of Repetitions on Muscle Injury

There is clear evidence that the number of eccentric or SSC repetitions has an effect on the amount of resultant muscle injury and force deficit.[97,116] Models that have induced single stretches in muscle within the physiological range have not resulted in muscle damage or a pronounced force deficit.[39,129] In other studies, it required more than one stretch within the physiological range to produce muscle injury.[97,110,284,297,298] Repeated stretches that varied from 225 to 900 at a final length of 110% L_o have resulted in myofiber damage and a resultant force deficit.[35,163,177,178,313] Thus, the amount of loading does have a graded effect on both changes in muscle performance and the extent of myofiber injury.[97,116] However, the effect of repetitions on repetitive exposures of either eccentric muscle actions or SSC has not been studied to date.

15.4.3.4 The Effect of Other Mechanical Factors on Muscle Injury

Dynamic muscle forces and length changes that are measured during eccentric muscle actions can be dissected into components of the dynamic signature. Components such as peak force,[110,178] average force,[39] work during the stretch,[129] and fiber length[110,129] have been found to affect the magnitude of contraction-induced injury. The force deficit resultant from an injurious exposure has been predicted by: (i) work done during the stretch when initiated from optimal length,[39,172] (ii) initial length and work during the stretch when not initiated at optimal length,[129] or by (iii) peak force and initial length.[110] Within a given level of force output, eccentric muscle actions performed at longer ranges of motion or fiber length have resulted in larger isometric force deficits in both humans[188] and animals[110,129,305] than stretches performed at a shorter range of motion. However, in these studies, the change in work (calculated by integration of the force–muscle displacement curve) during repeated stretches was not reported.[39,110,305] Work during the eccentric phase or stretch (negative work) has been shown to be well correlated with the isometric force deficit after a single eccentric contraction.[129,172] However, the length perturbation in these studies was beyond the normal physiological range of the target muscle. However, muscles stretched within the normal physiological range have required more than one repetition to produce injury.[284,297,298] Studies of repetitive eccentric muscle actions in the physiological range may have more external validity than single stretch models that have been studied outside of the normal physiological range.

15.4.3.5 The Effect of Exposure Duration and Lengthening Velocity

To determine if exposure duration and lengthening velocity affected muscle injury, McCully and Faulkner[178] used EDL muscles of mice. Although measured for 15 min exercise duration, there was no change in P_o due to muscle fatigue after the initial 5 min. A velocity of 1 L_f/sec produced a deficit in P_o after 3 days while the 0.2 L_f/sec and 0.5 L_f/sec lengthening velocities did not. A drop in P_o was mostly associated with stretch velocity. It was theorized that loss of peak force after fatigue prevents further muscle damage. Muscle injury increased with eccentric exercise duration for upto 5 min (no further force decrements were observed with subsequent eccentric muscle actions), higher velocities shortened the duration time for injury, and muscle force was a critical component in producing injury. Warren et al.'s[284] *in vitro* results in rats also indicated that higher lengthening velocities produced larger force decrements. The velocity component in muscle injury was investigated by Scifres and Martin[235] using a Kin-Com dynamometer to test human subjects for eccentric leg extension performance. One leg was tested at 30°/sec while the other leg was tested at 120°/sec. Delayed onset muscle soreness (DOMS) was more pronounced at the higher velocity, which indicated that higher velocity may produce additional muscle fiber injury. To further examine the effect of velocity on contraction-induced injury, Lynch and Faulkner[169] used single permeabilized fibers from mice EDL muscles. The severity of contraction-induced injury was not affected by the velocity of stretch. Controversy still exists as to the role of lengthening velocity in acute injury to skeletal muscle fibers. Furthermore, the role of lengthening velocity in chronic contraction-induced injury has not been investigated thus far. The effect of exposure duration (length of exposure) on repetitive injury has not been investigated to date, however it may be an important factor in repetitive injury causation.

15.4.3.6 The Effect of Loading History on Injury Susceptibility

The understanding of the injury pathophysiology is important in preventing contraction-induced injury. The predisposing factors, which mitigate injury are also important. Anecdotal observations of physical activities that result in a high incidence of injury has led to the hypothesis that if physical activities are repeated with adequate recovery time, muscles will eventually become "trained" and no injury will occur. Conversely, disuse could increase contraction-induced injury susceptibility[286] (Cutlip et al., unpublished observations). Human muscle performance studies assessed the relationship between muscle strength, soreness, and the release of intracellular proteins into the serum. Komi and Buskirk[150] used a training protocol of isometric, shortening, and eccentric muscle actions of the elbow flexors for 4 days/week for 7 weeks. Muscle soreness peaked after the first week and then disappeared. Maximum force during shortening and eccentric muscle actions increased throughout the 7-week test period. Newham et al.[186] also studied the effect of eccentric muscle actions of the elbow flexors performed on three occasions separated by 2 weeks. Muscle soreness was reduced after the second and third sessions, but was still present. Plasma levels of creatine kinase (a muscle-associated enzyme) were also greatly elevated after the first session, but were not elevated after the second and third sessions. Recovery of the maximum force was more rapid and complete after each subsequent session. A similar study indicated that a pre-training session of 24 eccentric contractions of the elbow flexors reduced muscle soreness and force deficit due to a 70 eccentric contraction protocol administered 2 weeks later.[60] Serum creatine kinase that showed an increase after the first session, diminished after the later session, which indicates a temporal relationship between creatine kinase levels and muscle soreness.

Results of studies employing small rodents (mice and rats) have been consistent with human studies. In one study, 198 eccentric contractions of the dorsi flexors were administered to anesthetized rodents *in vivo* once every 7 days for 6 weeks using a dynamometer.[76] The initial exposure to the protocol produced a 60% force deficit, which returned to 80% of the pretest value at 7 days. By the sixth week, no decline in force was observed. Muscles demonstrated an adaptive response, with increased whole muscle mass. Results from human and animal studies indicate that muscles can be trained to perform maximal eccentric muscle actions without injury; however, training must be continuous. The result of these studies raises an important issue in the study of muscle injury. Instituting training sessions containing eccentric contractions produces short-term force deficits and resultant myofiber disruption and inflammation, which later ceases and the muscle returns to normal function. The transient effects (temporary soreness and inflammation) are reduced with subsequent sessions. The preceding results pose an important issue about how the desirable effects of training can be distinguished from pathological changes that occur due to repetitive motion? Also, what characteristics of the dynamic inputs (force, strain, strain rate, number of repetitions) produce adaptive versus pathological responses in a chronic model?

15.4.3.7 The Effect of Age

Age can be an important factor in injury susceptibility and recovery. Zerba et al.[313] tested the hypothesis that muscles of old mice were more susceptible to injury than muscles of young and adult mice by attaching the distal tendon of the EDL muscle to a servomotor. The left leg was exposed to stretch while the right leg served as control. Results indicated a 27% deficit in P_o, and that old mice (43% deficit) were more susceptible to injury than young and adult mice. *In vitro* results of mice EDL muscles conducted by Faulkner et al.[72] indicated that old mice are more susceptible to injury and intrinsic differences in single permeabilized fibers account for the difference. Subsequent *in situ* and *in vitro* work by Brooks and Faulkner[37] and Faulkner et al.[72] supports earlier results that initial eccentric contraction-induced injury increases in old age. Single stretches of whole muscle and single permeabilized fibers (which rules out effects of excitation–contraction coupling, or membrane and extracellular effects) represented an effective method of focusing on factors that contribute to contraction-induced injury. Because their *in vitro* findings supported their *in situ* work, it suggested that failure has a mechanical etiology (due to work input) and increased injury susceptibility with age may be due to

decreased protein synthesis, and thus an increased population of weak sarcomeres. In contrast, Willems and Stauber did not find a difference in isometric force deficit after 30 eccentric stretches in old (39%) and young rats (35%).[297] While muscle weights were similar in the young (4 months) and old (24 months) groups, peak force was lower in the old group during the eccentric stretches. Thus, injury studies using rodents should be cognizant of age as a possible contributing or confounding factor in the response to contraction-induced injury. The effect of age on the adaptive or pathological response to a chronic administration of high force eccentric muscle actions has not been studied thus far. The age of the animals selected for study is an important factor in acute injury and may affect the response to repetitive exposure.

15.4.3.8 Recovery Kinetics

Recovery kinetics also is important in the evolution of the injury process. The primary determinant of the time course required for recovery is the magnitude of the secondary injury that results after the initial mechanically induced injury. The secondary injury cascade includes an inflammatory response, free radical damage, phagocyte infiltration, and eventual phagocytosis of the cytoplasm in areas of damaged fibers.[169] Depending on the magnitude of the initial contraction-induced injury, full recovery of normal structure and function requires from 7 to 30 days.[35,75,126,127,177,178,276] Faulkner et al.[75] investigated the recovery kinetics (sampled from 1 h post-test to 30 days post-test) after an acute exposure to eccentric contractions while leaving the distal tendon intact. The extent of injury to the EDL and TA muscles of mice was determined from 1 h to 30 days after passive shortening and lengthening, and eccentric and concentric contractions. No injury was produced in the control or passively lengthened and shortened groups. The active eccentric protocol produced a 50% decrease in P_o, which did not recover by 3 days; however, total recovery was evident at 30 days thus indicating that recovery from eccentric contractions is a prolonged process.[75,178] These findings were also supported in both rats[157,276] and rabbits[191] *in situ*, and mice,[130,132,168,288] *rabbits*,[86,163] rats (Cutlip, unpublished observations), and humans[58] *in vivo*. The results of these studies clearly showed that the recovery process is not due to metabolic fatigue, but fiber injury that requires days or weeks to recover functional performance and repair fiber lesions.

15.4.3.9 The Effect of Sex on Injury Susceptibility

There have been reports of sex differences in both human and animal models of exercise-induced muscle damage.[57,58] Creatine kinase activity in rats[4,5,18] and humans[70,247,263] after muscle injury have been reported with differing results. Differences in the inflammatory response after muscle injury have also been reported in both humans[263] and rats[252] with females exhibiting less cellular infiltrates into the damaged area of the target muscle. Komulainen et al. reported that there was less myofiber damage via changes in structural proteins in female than male rats after exposure to downhill treadmill exercise.[153] In contrast, Stupka et al. reported that muscle damage was similar in males and females after exposure to injurious eccentric exercise.[263] It has been shown that males have higher baseline plasma levels of muscle proteins, such as creatine kinase, myoglobin, and skeletal troponin-I, but when expressed with respect to baseline levels, the increase after injurious exercise was not different between men and women.[247] Sex-related hormones such as estradiol may be responsible for reducing the inflammatory response after injurious exposure. Work done in female rodents and male rodents supplemented with estradiol showed a reduced amount of leukocyte invasion, less focal inflammation, and resultant myofiber necrosis.[252,271] Indeed, estradiol may have a protective effect that reduces the amount of membrane damage resulting from eccentric damage.[270] Several studies have also reported no difference in force loss after injury between males and females in rats[298] and humans[34,220,233] or in the time required for recovery. However, females did have a greater loss of range of motion after injury than males.[220] The results from animal studies indicate that females showed a blunted inflammatory response as compared to their male counterparts, but in humans this does not seem to be the case. Results from human studies tend to show a delayed recovery in females, particularly with the use of oral contraceptives.[230]

15.4.3.10 SSC-Induced Injury

A viable method to study eccentric and concentric muscle performance simultaneously in the context of muscle injury is via SSCs. SSCs (reciprocal eccentric/shortening contractions) have been studied in the context of human locomotion and athletic performance[14] and have been shown to produce muscle injury due to the eccentric component of the cycle.[97,125] Natural muscle function involves SSCs and this model provides a sound physiological foundation in which to study muscle mechanics and injury.[149] Both the change in concentric and eccentric muscle function before and after an injury protocol (that includes eccentric contractions), and the change in concentric and eccentric muscle function during the injury protocol can be investigated using SSCs. Recently, the relationship between changes in negative and positive work and the isometric force deficit after an injurious exposure has been investigated and the study showed a positive correlation between the change in negative work and the isometric force deficit resultant from injury.[62] Also, changes in real-time muscle function during SSCs (Figure 15.7) are also positively correlated with isometric force deficit and the degree of myofiber injury after an injurious exposure.[97] Specifically, the decay in peak eccentric forces and the decay in force enhancement during each stretch during an injury protocol were positively correlated with isometric force deficit and the degree of fiber injury. This study also demonstrated that changes in real-time eccentric force production during SSCs are indicative of resultant performance decrement (Figure 15.8) and myofiber damage days later (Figure 15.9). Exposure to equivalent isometric contractions did not result in myofiber injury (Figure 15.9). Increasing the number of repetitions of SSCs results in an increase in the degree of myofiber injury (Figure 15.10).

15.4.3.11 Chronic Injury

It is well understood that acute exposure to unaccustomed eccentric muscle actions can result in injury, but an initial exposure can be beneficial to reduce injury resulting from a subsequent exposure.[58,60] This protective effect from an initial exposure has been shown to reduce the effects of a subsequent exposure 1 week upto 6 months from the initial exposure (with no intervening exercise).[59,179] Thus, there is a

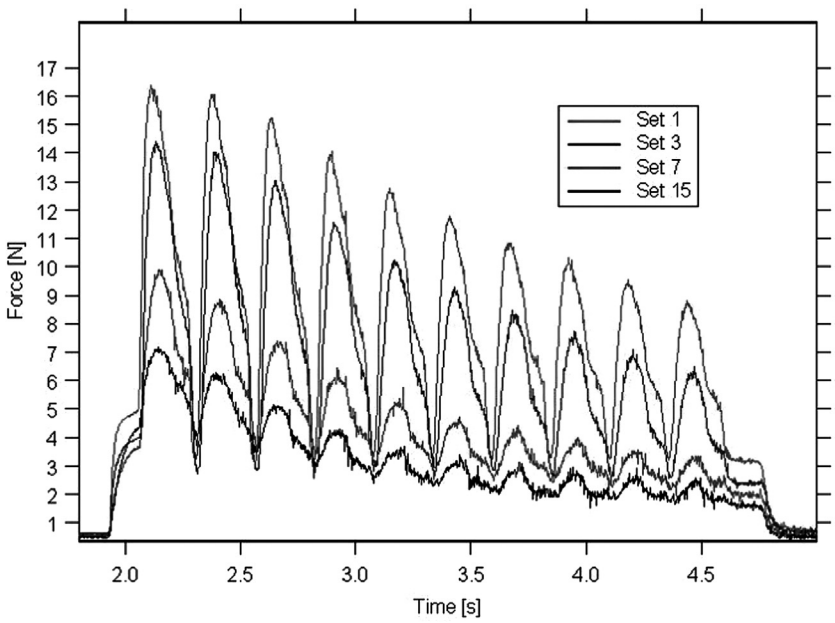

FIGURE 15.7 Fifteen sets of injurious SSCs performed on active dorsiflexor muscles at 1-min intervals. The curves are the force response (N) of the dorsi flexor muscles during sets 1, 3, 5, 7, and 15. The SSCs were conducted at a range of motion of 70–120–70° in a reciprocal fashion at 500 deg/sec.

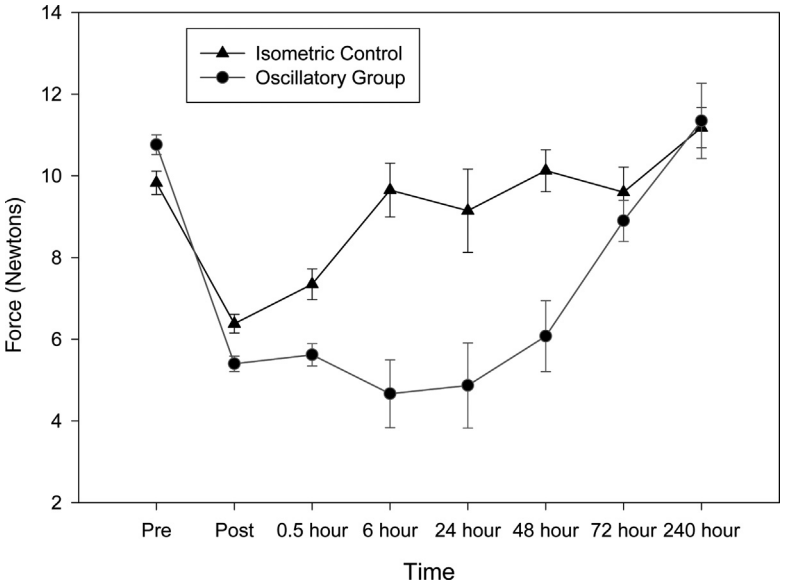

FIGURE 15.8 Time course of change in isometric force resultant from exposure to SSCs (oscillatory group) and isometric contractions (isometric control) over a 10-day period.

pronounced reduction in soreness, loss of strength, and release of enzymes, such as creatine kinase after the second exposure than the initial exposure.[17,58,60] Also, a second exposure of eccentric contractions does not appear to delay the recovery from the first exposure. Typically, recovery from eccentric contraction-induced injury takes at least 10 days for recovery of performance and remodeling of muscle fiber disruption.[96] If the second session is administered within 6 days after the first exposure, the recovery time was unaffected.[194] Thus, the normal recovery process is not affected by at least one intervening injurious exposure. Clearly there is an adaptation that takes place after one exposure to injurious eccentric contractions that ameliorates the injurious response to subsequent exposures. The mechanism by which this takes place is not clearly understood at this time. Exposure to a mild session of eccentric contractions still provides a protective effect to further sessions of more intense eccentric exposure.[40,60]

Some authors postulate that the adaptive mechanism could be at the level of the central nervous system where recruitment patterns could be adapting to recruit in a different fashion such that motor unit recruitment would be appropriately synchronized to reduce asymmetric stresses in the muscle fibers.[126,194] However, the repeated bout effect has also been found in electrically stimulated animal models, which would argue against the hypothesis that neural factors provide the adaptive effect.[179] Thus, the mechanism for adaptation must be in the muscle fibers themselves. There could be weak sarcomeres as a result of deconditioning that are more susceptible to stresses generated during eccentric contractions. The notion that stress-susceptible fibers exist was first postulated by Armstrong et al.[8] If fragile fibers were compromised after an initial session of eccentric muscle actions, the target muscle should lose muscle volume as a result of the loss of those fragile fibers. This hypothesis was supported by findings in human elbow flexors where there was a loss of muscle volume (approximately 10%) 14 days after an injurious exposure to eccentric muscle actions. In contrast, after a second session performed 8 weeks later, there was no loss of muscle volume. The authors concluded that the fragile fibers were lost after the first session and are repaired over time. As repaired fibers replace the fragile fibers, the muscle becomes more resistant to eccentric contraction-induced damage.[85] However, if muscle is not used, fragile fibers may again develop that can be injured at some juncture. There is also evidence that one session of injurious eccentric contractions can ameliorate the resultant damage from the

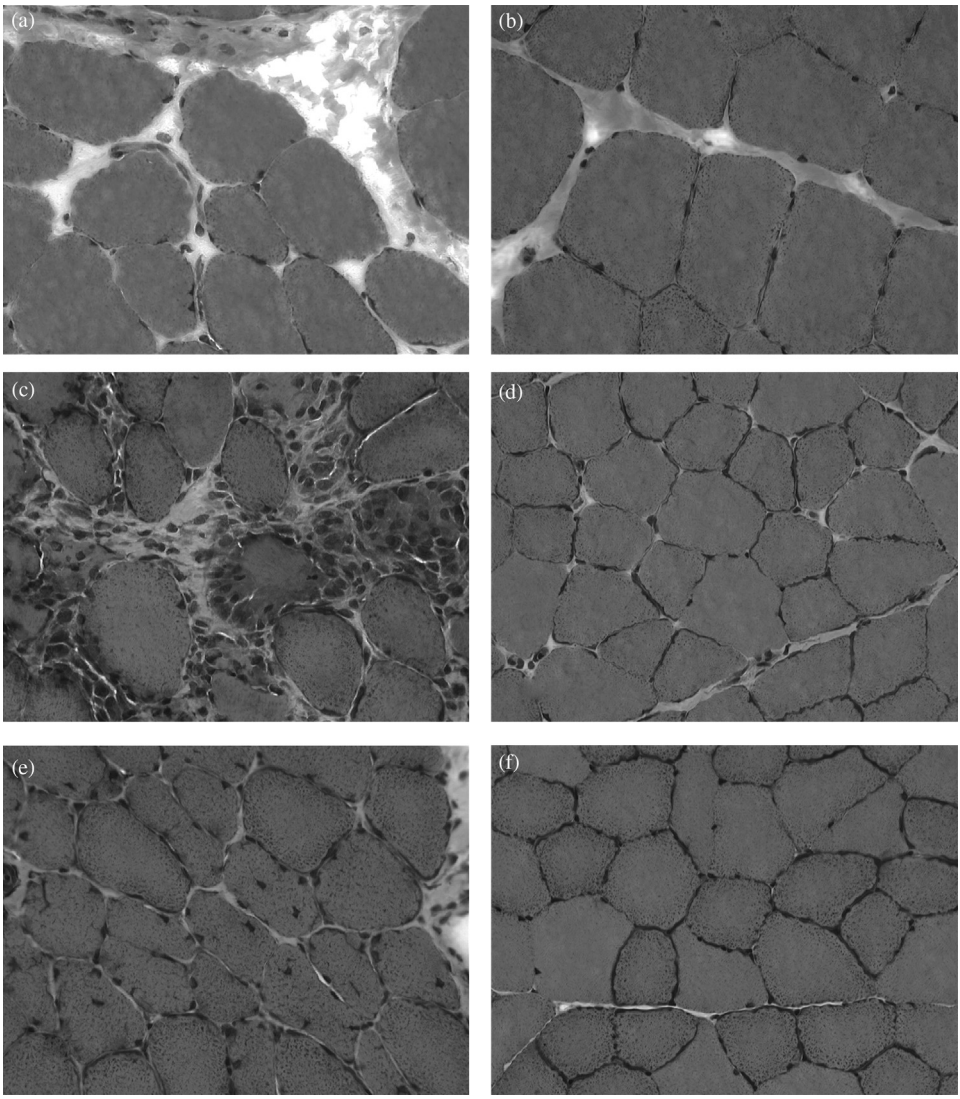

FIGURE 15.9 Cross-sections of TA muscle (H&E stain, 40× objective). After exposure to SSCs: (a) 6 h, (c) 72 h, and (e) 240 h. After isometric contractions: (b), 6 h, (d) 72 h, and (f) 240 h.

second session, even if the sessions are spaced 6 months apart.[198] This indicates that in normal subjects, the protective effect can last for some months, but the protective effect predictably dissipates with time.

Another explanation for this protective effect is the notion that muscles can add sarcomeres after an initial injurious exposure as a means to reduce the number that are at their extreme length, thus reducing injury.[58] The addition of sarcomeres after an injurious session of eccentric-biased exercise has been shown in both human[292] and animal[170] studies. Apparently, concentric muscle training reduces the number of sarcomeres and does not provide the level of protection in repeated sessions that eccentric training does.[292]

Few studies have investigated muscle response to the chronic administration of eccentric, concentric, and isometric muscle actions. The limited number of studies which have been conducted did not control the dynamics of movement, quantify the forces during the movement, or quantify the changes in performance longitudinally throughout the protocol. Investigation of *in vivo* muscle functional,

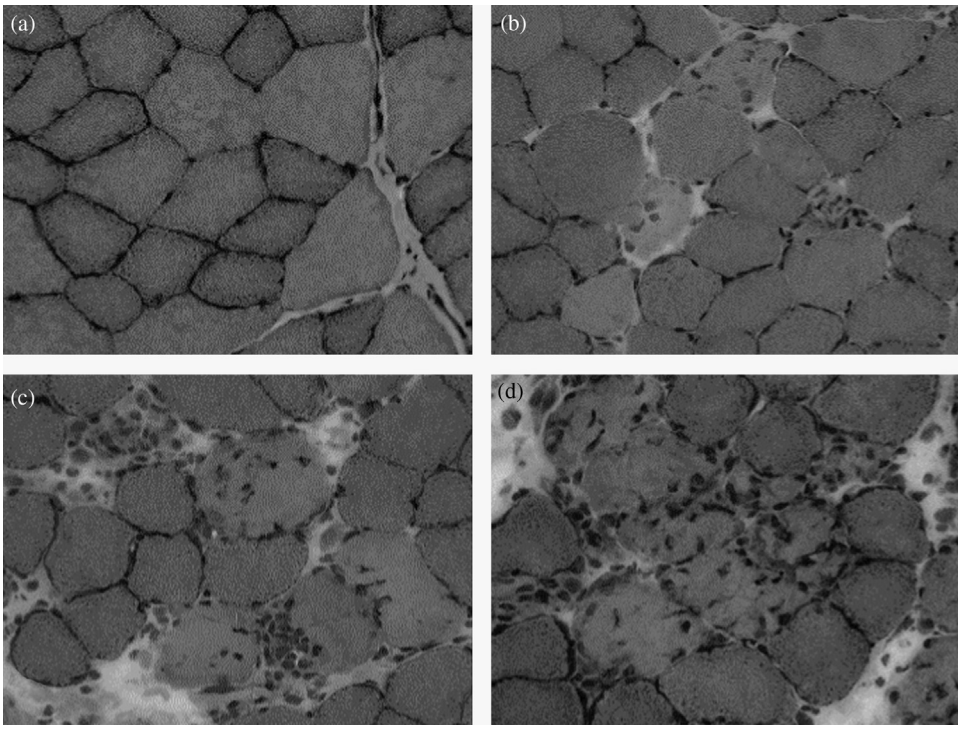

FIGURE 15.10 Histology of control muscle with no injury (a) and exposure to SSCs of 30 repetitions (b), 70 repetitions (c), and 150 repetitions (d).

histological, and biochemical responses to a chronic administration of muscle actions will further elucidate the factors, which predispose or mitigate contraction-induced injury. It will also further the understanding of the functional changes that result, characterize the inflammatory and repair kinetics longitudinally, and determine whether biochemical markers are representative of muscle performance or injury.

In summary, the study of different factors and their effect on muscle injury indicates that increasing force, task duration, number of repetitions, and range of motion exacerbate muscle injury. Older animals also have an increased susceptibility to injury. Training, particularly if involves eccentric muscle actions, can provide a protective effect from contraction-induced muscle injury. These findings should be of use to those attempting to ameliorate occupational muscle injury incidence.

15.5 Cellular Biology of Muscle Damage

Our understanding of the cellular and molecular mechanisms involved in mediating muscle damage and repair after strain, or low force, repetitive motion inducedinjuries is rapidly increasing.[20,58] Understanding how muscle tissue responds to these various stressors, and the time course of those responses, is necessary for defining approaches that can be taken to reduce the chances of obtaining an injury, or approaches to enhance the rate of recovery once an injury occurs. The cellular changes that occur as a result of acute muscle strain and repetitive motion are different. Strain injuries are associated with structural damage to myofibers, blood vessels and nerves, and cause edema and inflammation.[90,255] Repetitive motion damage can be associated with muscle ischemia, pain, and mitochondrial dysfunction.[137,243] Details regarding the cellular responses to repetitive use and strain injuries are discussed here.

15.5.1 Strain Injuries and Skeletal Muscle

Strain injuries are the result of three basic processes: (1) Initially, excessive forces coupled with lengthening result in structural damage to muscle cells (myofibers), including tearing of the cell membrane (i.e., the sarcolemma). (2) Structural damage to the sarcolemma results in an increase in intracellular Ca^{2+} levels, modifications of myofiber proteins and lipids, and the activation of intracellular pathways that regulate the injured muscle's response to damage. (3) Pro- and anti-inflammatory factors (i.e., cytokines and chemokines) are released by local tissues and infiltrating immune cells. Cytokines and chemokines cause inflammation and stimulate cellular pathways mediating muscle regeneration and repair. These three processes are involved in mediating the extent of physical damage, functional changes of the muscle, pain, and repair.

15.5.2 Physical Damage to Tissue

Strain injuries are caused by exposure of muscle and other soft tissues, including vasculature, nerves, and tendons to excessive strain or lengthening.[86,89,165] Studies performed in both humans,[88,90,91,187,190] and animals[10,161,257] have demonstrated that exposure to lengthening contractions results in physical damage to muscle tissue that can include shearing of myofibers, the loss of intermediate filaments and mitochondria, damage to the extracellular membrane, and disruptions in Z-line alignment (i.e., Z-line streaming). Immunostaining for structural proteins that maintain the integrity of the myofiber, such as desmin, titin, and fibronectin, have demonstrated that there are disruptions of the exo- and endosarcomeric membranes,[86,163,164] and of the extracellular matrix[163,257] in strain-injured muscle tissue. In lengthening contraction-induced injuries, damage within the muscle is most often seen at the myotendinous junction and at specific sarcomeres.[95,114,192,201] In fact, it has been hypothesized that there is a population of sarcomeres that are weaker, and tear more easily under lengthening conditions.[86,183,265] The stretch-induced damage to the extracellular matrix, sarcomeres, and critical cell organelles is associated with reduced isometric muscle force,[62,96,286,289] but not with pain. It has been hypothesized that these initial structural changes initiate a chain of events that may maintain an injury-induced force deficit, result in inflammation and pain, and stimulate pathways important for regulating muscle repair and regeneration.

15.5.3 Intracellular Ca^{2+} and Muscle Damage

Studies examining the effects of lengthening contractions on myofibers have demonstrated that there is an influx of Ca^{2+} into muscle cells[131,287,290] and mitochondria[66,267] in muscle exposed to damaging contractions. Calcium influx into myofibers can be increased in two ways. First, extreme stretch and strain can open the stretch-sensitive Ca^{2+} channels in the cell membrane. However, treating animals with a calcium channel blocker (i.e., verapamil) prior to injury only partially reduces the calcium influx into the cell,[9] and therefore, Ca^{2+} must be entering via an additional pathway. Injury-induced tears and damage to the sarcolemma can also allow Ca^{2+} to leak into the cell from the extracellular fluid.[290] In *in vitro* studies, membrane damage, and enzyme efflux can be prevented in muscle exposed to contractions by removing Ca^{2+} from the extracellular buffer.[135] Treating animals with Ca^{2+} chelators (molecules that preferentially bind Ca^{2+} and prevent it from entering the cell) during and after treadmill running also reduces tissue damage and the influx of Ca^{2+} in myofibers (66). Thus, this early influx of Ca^{2+} appears to play a critical role in mediating contraction-induced injuries to muscle tissue.

Increases in free Ca^{2+} within myofibers can result in the degradation of myofiber proteins and lipids, and result in the degeneration of damaged myofibers.[42] As mentioned earlier, intracellular Ca^{2+} homeostasis within myofibers is maintained by the SR. As intracellular Ca^{2+} levels rise, the SR normally increases the rate of Ca^{2+} uptake to keep free intracellular concentrations fairly stable. However, it has been demonstrated that the SR is less capable of sequestering additional Ca^{2+} after muscle has been exposed to lengthening contractions.[42] This reduction in Ca^{2+} up- take by the SR is associated with a reduction in muscle force[282] and may contribute to the rise in free intracellular Ca^{2+} after injury-inducing contractions.[42] Thus, the damage-induced loss of Ca^{2+} homeostasis in injured myofibers

may mediate the acute reduction in isometric force, and it may initiate other cellular mechanisms that exacerbate myofiber damage.

Increases in intracellular Ca^{2+} can also alter myofiber structure and integrity by modifying proteins and lipids in damaged cells.[163,304] For example, calpain proteases, which are activated by increases in intracellular Ca^{2+}, are elevated in muscle after exercise.[21] Calpain can cleave signaling, cytoskeletal, and myofibrillar proteins.[228,229] This cleavage may act to target proteins for degradation by other proteases including ubiquitin.[228] *In vitro* studies have demonstrated that activated calpain degrades desmin and stimulates the release of α-actinin, thereby inducing Z-line streaming.[22] Calpain-mediated modifications of proteins can also stimulate the activity of a number of intracellular signaling proteins important for mediating cellular responses to damage. For example, the signaling molecule, protein kinase C (PKC), is activated by calpain cleavage.[228] This active form of PKC can affect myofiber function by modifying proteins that are already present in the cell or by acting upon transcriptional pathways to regulate gene expression.[232] Calpain may also act as a chemotactic signal to enhance neutrophil infiltration into the damaged tissue.[22]

A number of other intracellular signaling systems are also activated by increased intracellular Ca^{2+} levels in myofibers. For example, intracellular Ca^{2+} liberates phospholipase A2 (PLA2) from the extracellular membrane. PLA2 acts to increase arachidonic acid production and the synthesis of prostaglandins, which can stimulate inflammation and cause pain.[204,244,279,280]

Other signaling systems that may be indirectly activated in response to contraction-induced injury and increases in intracellular Ca^{2+} include the extracellular receptor kinase 1-2 (ERK1-2), p38 mitogen activating protein kinase,[51] and c-JUN NH_2-terminal kinase pathways.[47,117,200] These pathways are stimulated in response to growth factors, cellular stress, and injury. Activation of these pathways regulates transcriptional and translational activity in many cell types including muscles. Although the increase in intracellular Ca^{2+} and associated proteolysis and lipolysis have been traditionally thought to exacerbate muscle injury, Ca^{2+}-induced activation of cell signaling pathways and proteolysis might also activate cell systems necessary for initiating cellular repair and regeneration.

15.5.4 Muscle Inflammation

The physical disruptions of muscle fibers along with increases in intracellular Ca^{2+} are the initial effects of muscle injury. However, force deficits, muscle swelling, and soreness occur 1 to 7 days after the initial injury, and are associated with muscle inflammation.[10,89,97,163] Neutrophils, the first immune cells to enter damaged tissue, actually infiltrate muscle within 2 h of the initial injury.[79,269] We are only beginning to understand the complex roles that various immune cell types play in the damage and repair processes. However, it has been demonstrated that neutrophils phagocytize degenerating fibers and debris produced by injury. In addition, neutrophils can participate in the production and release of free radicals from damaged tissue, which can exacerbate the damage.[25] Neutrophils can also produce proteases and a number of cytokines, including tumor necrosis factor-α (TNF-α;[67]) and IL1-β.[272] These cytokines might increase muscle catabolism and degradation and act to attract monocytes to the site of the injury.[2,113,182]

Monocytes/macrophages, the other inflammatory cells commonly seen in injured muscle, can be found between 12 h and 14 days after the initial muscle injury.[224,251] In rats, macrophages expressing specific cell-surface molecules, including ED1 and ED2, have been identified in damaged muscle tissues.[156,157,251] ED1 expressing macrophages infiltrate damaged and necrotic tissues and remove debris. These macrophages also express pro-inflammatory cytokines including TNF-α.[64,310] Besides increasing muscle catabolism and promoting protease activity, TNF-α also activates the transcription factor, nuclear factor κB (NF-κB), to stimulate transcription of genes encoding for proteins that are part of the ubiquitin proteolytic pathway.[112,180] TNF-α may also stimulate the transcription of other pro-inflammatory cytokines including IL-1β and IL-6 and the chemokine monocyte chemoattractant protein (MCP-1[15,307]). The increased production of cytokines and chemokines by inflammatory cells

in damaged tissues enhance local pathways mediating tissue inflammation and may act to exacerbate damage during the first 5 days after muscle injury.

15.5.5 Muscle Regeneration and Repair

The infiltration of immune cells along with the release of pro-inflammatory cytokines appears to enhance muscle damage. During the acute phase of the injury, both pain and force deficits appear to be reduced by treating animals with nonsteroidal anti-inflammatory drugs (NSAIDs). For example, rabbits, which were exposed to a session of repeated eccentric contractions and treated with the NSAID flurbiprofen, showed improved functional recovery during the first week after injury as compared to controls.[181] However, 4 weeks after the injury, the NSAID-treated animals demonstrated reduced force generation. The authors hypothesized that treatment with NSAIDs may have interfered with or delayed the recovery process in these animals.[181] In humans with muscle damage caused by downhill walking, treatment with over the counter doses of acetaminophen or ibuprophen results in a decrease in pain, but these anti-inflammatory agents also decrease protein synthesis, which may be needed for muscle repair.[209,273,274] These findings suggest that inhibition of the inflammatory response interferes with normal recovery of muscle after a strain-induced injury.

Inflammatory cells, particularly macrophages, may stimulate myofiber regeneration through a number of different mechanisms. ED2 expressing macrophages, also referred to as resident macrophages, are thought to play a role in muscle repair and myofiber regeneration.[43,156,157,251] These macrophages may stimulate growth and repair by releasing a number of factors that could stimulate the division, migration, and differentiation of muscle precursor cells. These factors include, fibroblast growth factor-2 (FGF-2[77,236]), insulin-like growth factor-1 (IGF-1[1,119,216]) and hepatocyte growth factor (HGF[236,312]). In addition, ED2-expressing macrophages may also release the anti-inflammatory cytokines IL-6[44,93] and IL-15.[45,214,215] Thus, although inflammation causes pain and appears to exacerbate myofiber damage, the inflammatory process also appears to be necessary for complete repair of tissues and recovery of muscle function.[213]

15.5.6 Muscle Injuries Associated with Low-Force Repetitive Tasks

Low-force repetitive tasks involve movements that require little force generation by a muscle. Instead, the muscle action may need to be maintained over long periods of time, or repeated over and over again during a work cycle.[241,243] Injuries caused by these types of activities are not usually associated with inflammation or large areas of myofiber degeneration, but instead are characterized by muscle pain and/or rapid fatiguability of the muscle.[133,138,159,239,240] Low-force repetitive task injuries are prevalent in people working with computers. With the increase in computer use, both at home and in the workplace, understanding the mechanisms underlying this type of damage is crucial.

Injury due to overuse has been studied in people with trapezius myalgia. This disorder is often seen in workers whose job requires them to maintain stable upper body postures for extended periods of time, such as computer and clerical work.[133,138] Trapezius mylagia is associated with the appearance of ragged type I muscle fibers and with a decrease in muscle blood flow to the injured region. This ragged appearance of myofibers is an indicator of mitochondrial dysfunction in the cell.[137,158,159] The dysfunction and pain associated with trapezius myalgia and with other overuse injuries have been linked to changes in Ca^{2+} regulation in the damaged area, changes in the pH of the intra- and extracellular fluids, and changes in the local concentrations of specific ions involved in mediating muscle activity.[134,203,239] These biochemical alterations may have profound effects on myofiber metabolism and on the activity of sensory pathways carrying pain information.[243] Although recent studies have identified some of the factors that play a role in inducing injuries that are due to overuse, more research needs to be done to determine how these various physiological changes act together to result in pain and injury.

15.5.7 Long-Term Changes in Muscle in Response to Injuries

Most muscle injuries are acute, and recovery is complete within a couple weeks of the initial injury. However, long-term exposure to repetitive motion, load bearing or awkward postures in the workplace has been associated with an increased risk of developing more chronic musculoskeletal disorders.[24] These more chronic disorders include tendonitis, fibromyalgia, myositis, osteoarthritis, and synovitis.[19,41,111,211] Recent studies have focused on the biological mechanisms by which overuse, or acute strain injuries, may act to chronically affect muscle function and pain.

Understanding the biological underpinnings responsible for generating chronic disorders has been difficult because a number of factors, including age, general health, genetic predisposition, stress level, and length of exposure to a strenuous job, influence the development of these conditions.[24,111] Thus, what we know about the mechanisms responsible for generating chronic musculoskeletal disorders comes from studying people with chronic disorders (such as workers with trapezius myalgia), and by examining the effects of a few limited animal models of chronic damage or genetic models of chronic inflammation and muscular atrophy. Because the mechanisms involved in mediating trapezius mylagia were discussed previously, this discussion will focus on animal models used to study the etiology of chronic disorders.

Most animal models of chronic injury have used repeated injections of a toxin[52,225] or muscle overload of a single limb caused by paralyzing or binding the opposite limb, or by removing some muscle, and thereby forcing the remaining muscle to assume the additional load.[257] These models result in myofiber degeneration and the infiltration of macrophages.[255] In addition, there is an expansion of the extracellular matrix, along with an increase in collagen expression and fibrosis in muscle[92,256] and tendon[46] tissues, which leads to scarring. Muscle fibrosis results in decreased muscle strength and flexibility and is often associated with pain.[25,184,256,300] Thus, determining which mechanisms are responsible for increasing fibrosis, and interfering with these mechanisms may reduce the effects of chronic overload on muscle tissues.

Models of muscular dystrophy, such as dystrophin-deficient *mdx* mice, have also been used to try to determine how chronic damage may affect muscle tissue and function. *Mdx* mice, which are a model for Duchene's muscular dystrophy, are lacking the dystrophin protein. Dystrophin, along with dystroglycan, links the extracellular matrix to the cytoskeleton of the myofiber.[121,122] The absence of dystrophin compromises the strength of the sarcolemma, making it more susceptible to breakage.[217,218] Once the sarcolemma is damaged, Ca^{2+} can enter the cell.[99,123,124] This rise in intracellular Ca^{2+} activates the same proteases and lipases that are activated after muscle injury. Because there is persistent myofiber damage and necrosis occurring in *mdx* mice and people with Duchene's muscular dystrophy, there is also persistent inflammation.[108,249,250] In fact, it has been hypothesized that this inflammation exacerbates the muscular atrophy and loss of function. This is supported by the fact that treating Duchenne's patients with immunosuppressive drugs improves muscle strength.[139] Studies of *mdx* mice also demonstrate that these animals display a chronic inflammatory response, with the gene expression of many pro-inflammatory factors being chronically increased as compared to control animals.[212] The increased expression of these inflammatory factors is associated with fibrosis, muscle atrophy, and eventually necrosis.

The studies performed using these various animal models indicate that muscle overuse, or long-term inflammation, may result in chronic muscle damage. However, it is still unclear which occupational exposures, if any, result in long-term muscle inflammation. Future studies examining the cellular effects of repetitive loading on muscle tissue will help determine which mechanisms may underlie the development of chronic musculoskeletal disorders.

15.5.8 Biomarkers of Injury

One of the goals of occupational research is to find biomarkers that can be used to determine if a person is at risk for developing a muscle injury or for diagnostic purposes. The approaches that have been used

to assess contraction-induced damage have been reviewed previously.[289] In humans, these approaches include examining force production, histology, blood levels of muscle-associated proteins, and reports of pain.

Force production has commonly been used to assess muscle function because reductions in force are often correlated with injury. Changes in force are immediate, and maintained for a number of days following the exposure that caused the injury.[10,61,87,96,291] Although muscle damage is often associated with a force deficit, force measurements alone should not be used to diagnose injury because muscle fatigue can also result in reductions in force.[80–82]

Overuse disorders can also be associated with losses in force, however a better marker for these disorders is muscle fatigue.[30,133] Although injured muscle does demonstrate signs of fatigue,[80,299] fatigue is also seen prior to the generation of injury, and it is believed that fatigue and the cellular changes associated with this physiological state, may be in part responsible for generating overuse disorders.[243] Thus, depending on when measurements are taken, fatigue can be used as a biomarker for predicting the development of a disorder, or for diagnosing a disorder.

Increases in the circulating concentrations of certain muscle-associated proteins, including creatine kinase, lactate dehydrogenase, and myoglobin are often associated with strain-induced muscle injuries.[54,195,223,227,248] Increases in these circulating proteins do not correlate well with injury-induced force deficits during the first 24 h of injury,[56,60,186] and thus, the concentrations of these proteins do not serve as good markers for the early diagnosis of muscle injury. In addition, certain proteins, such as creatine kinase, fluctuate in response to muscle activity, and not solely in response to muscle injury.[55,187,289]

Histological examination of muscle biopsies can be used to diagnose muscle injury. As mentioned previously, infiltration of immune cells, disruptions of the sarcolemma, and necrosis can be seen in muscles with strain-induced injuries.[90,163,255] However, these changes are not usually apparent until 24–48 h after the injury,[90] and thus, they cannot be used to identify injuries early during the process. With overuse disorders, ragged red fibers are found by histological examination of biopsy tissue.[159] These ragged red fibers appear to be strongly immunopositive for cytochrome C oxidase (an enzyme involved in mitochondrial function), or immunonegative for this marker.[158] Thus, the presence of ragged red fibers, and their cytochrome C oxidase phenotype, can be used as a marker of muscle disorders.

Pain or muscle soreness is one of the most common symptoms used to determine if there is muscle damage. However, with strain-induced injuries, muscle soreness is not apparent until 24–48 h after the injury occurred,[55,90] and it does not correlate well with injury-induced force deficits.[50,69,136] Therefore, pain is not a good marker to use for early diagnoses of strain injuries. Pain is also common in overuse disorders and is associated with muscle fatigue,[159,240] the presence of ragged red fibers,[158,159] and reductions in blood flow.[158] Pain has also been correlated with biochemical changes associated with fatigue.[30,240,242] Because fatigue often precedes actual damage, pain and muscle fatigue may serve as biomarkers for predicting muscle damage.

Based on our current knowledge the diagnoses of muscle strains can most quickly be done by checking for force deficits. Reports of muscle soreness, or the presence of myofiber proteins in the blood can be used to confirm that the force deficit is due to an injury. Depending on the timing of events, reports of pain and measurements of muscle fatigue can also be used to indicate that injury may occur, or to diagnose an injury. Although most biomarkers are currently used for diagnosis, research focusing on pain, fatigue, and their association with biomarkers may provide a means for determining when an individual is at risk for developing a muscle strain or disorder.

15.6 Recommendations for Future Work

15.6.1 The Need for More Refined *In Vivo* Models

The most refined *in vivo* models to date have used dynamometry and electrical stimulation to study the target muscle of interest. However, these models have generally been used to study acute muscle injury.

In vivo dynamometry is well suited for the study of chronic muscle injury and adaptation and studies of this kind would be beneficial in elucidating the physiological response to repetitive biomechanical loading. Biomechanical loading parameters, such as number of repetitions per day, the velocity and acceleration of the movement, the range of motion of the movement, muscle force or torque during each movement, and the rest interval between sets, should be controlled during experiments. Controlling and quantifying the biomechanical loading profile is essential for rigorous study of muscle injury and adaptation for both acute and chronic exposure studies. Also, it is important to have the ability to vary the biomechanical inputs to study the effects of different inputs, and how the level of those inputs, such as more or less repetitions, higher or lower velocities or acceleration, and so on, affects the physiological response after a single exposure and/or multiple exposures.

In addition to controlling the biomechanical inputs, it is also important in volitional models to not only provide an apparatus to facilitate controlled movement, but to appropriately instrument the apparatus to monitor movement dynamics in real time. This will allow for quantitation of the biomechanical loading profile within each exposure session for the study of both acute and chronic muscle response. This approach is similar to *in vivo* dynamometry in that the biomechanical loading signature is recorded during each exposure, but different from *in vivo* dynamometry in that the movement profile is not controlled by an external source like a servomotor, but controlled by the animal.

In vivo models, whether volitional or nonvolitional, can provide a wealth of information about the effects of biomechanical loading inputs on acute and chronic physiological responses. Refinement of these models to control and monitor the biomechanical loading signature has been done in nonvolitional models and is currently being accomplished in volitional models.

15.6.2 The Need for Tissue Mechanobiology Studies

There is a clear need for tissue mechanobiology studies to determine the failure or injury mechanics of soft tissues and ultimately the repair kinetics after acute or chronic injury. From a muscle perspective, dose–response models would be important to develop in both an acute and chronic framework. In the acute framework, the effect of number of repetitions, rest between sets of exposures, range of motion, movement kinematics such as velocity, acceleration, and jerk, and force on the amount of soft-tissue injury should be fully investigated. The changes in soft tissue at the cellular level commensurate with injury should also be investigated. Defining damage at the tissue level is important to develop a consensus about what is soft-tissue injury. The time course of these changes after an acute exposure and the mechanisms of repair should also be fully characterized. Factors such as level of conditioning, age, and gender, and other relevant comorbid factors on injury susceptibility and rate of repair are also important considerations.

In the chronic framework, the threshold for injury under sustained or repeated loading should be determined. This should be based on the foundation developed by acute injury studies regarding the effect of different biomechanical parameters and any intrinsic interplay. Additional parameters such as exposure duration (number of days, weeks, or months) and the duty cycle (rest period between exposures) should also be considered. Using this framework, the time frame for injury to develop, the sustainment of injury, and any repair that can take place during repeated exposures should be studied. Functional measurements as well as noninvasive biological measurements should be made to monitor the status of the animal. Patterns of rest and reuse after injury are also important to consider.

15.6.3 Summary

There has been much work in the area of exercise-induced muscle injury over the past 30 years. The relation between factors such as force, range of motion, number of repetitions, exposure duration, velocity, age, gender, and training on muscle injury have been studied and have a clear occupational relevance. Also, the cellular mechanisms responsible for the injury and repair processes have been well studied to date. The relation between muscle function, myofiber damage, pain, and molecular indicators

of injury have also been studied. Clearly, more work needs to be done in this area, particularly regarding the physiological response to long-term repetitive loading. The findings to date indicate that eccentric muscle actions result in muscle damage and recovery from this injury can require upto 1 month. Increasing the biomechanical exposure such as force, number of repetitions, and range of motion can exacerbate the magnitude of injury response. Increased age can also increase injury susceptibility. The encouraging news is that training can reduce the injurious response and adaptation can take place, particularly if the appropriate rest intervals are included. There are biomarkers currently being studied that may indicate the evidence of myofiber injury that have the appropriate level of sensitivity and specificity needed for occupational monitoring. The area of soft-tissue pathomechanics can provide a wealth of information that will be of value to ergonomists and occupational health professionals in the quest to reduce the incidence of occupational musculoskeletal disorders.

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References

1. Adams, G.R., Autocrine and/or paracrine insulin-like growth factor-I activity in skeletal muscle, *Clin. Orthop.*, (403 Suppl), S188–S196, 2002.
2. Alvarez, B., Quinn, L.S., Busquets, S., Lopez-Soriano, F.J., and Argiles, J.M., Direct effects of tumor necrosis factor alpha (TNF-alpha) on murine skeletal muscle cell lines. Bimodal effects on protein metabolism, *Eur. Cytokine Netw.*, 12(3), 399–410, 2001.
3. Alway, S.E., Attenuation of Ca(2+)-activated ATPase and shortening velocity in hypertrophied fast twitch skeletal muscle from aged Japanese quail, *Exp. Gerontol.*, 37(5), 665–678, 2002.
4. Amelink, G.J., Kamp, H.H., and Bar, P.R., Creatine kinase isoenzyme profiles after exercise in the rat: sex-linked differences in leakage of CK-MM, *Pflugers Arch.*, 412(4), 417–421, 1988.
5. Amelink, G.J., Koot, R.W., Erich, W.B., Van Gijn, J., and Bar, P.R., Sex-linked variation in creatine kinase release, and its dependence on oestradiol, can be demonstrated in an in-vitro rat skeletal muscle preparation, *Acta Physiol. Scand.*, 138(2), 115–124, 1990.
6. Armstrong, R.B., Mechanisms of exercise-induced delayed onset muscular soreness: a brief review, *Med. Sci. Sports Exerc.*, 16(6), 529–538, 1984.
7. Armstrong, R.B., Muscle damage and endurance events, *Sports Med.*, 3(5), 370–381, 1986.
8. Armstrong, R.B., Initial events in exercise-induced muscular injury, *Med. Sci. Sports Exerc.*, 22(4), 429–435, 1990.
9. Armstrong, R.B., Duan, C., Delp, M.D., Hayes, D.A., Glenn, G.M., and Allen, G.D., Elevations in rat soleus muscle [Ca2+] with passive stretch, *J. Appl. Physiol.*, 74(6), 2990–2997, 1993.
10. Armstrong, R.B., Ogilvie, R.W., and Schwane, J.A., Eccentric exercise-induced injury to rat skeletal muscle, *J. Appl. Physiol.*, 54(1), 80–93, 1983.
11. Armstrong, R.B., Warren, G.L., and Warren, J.A., Mechanisms of exercise-induced muscle fibre injury, *Sports Med.*, 12(3), 184–207, 1991.
12. Ashton-Miller, J.A., He, Y., Kadhiresan, V.A., McCubbrey, D.A., and Faulkner, J.A., An apparatus to measure in vivo biomechanical behavior of dorsi- and plantarflexors of mouse ankle, *J. Appl. Physiol.*, 72(3), 1205–1211, 1992.
13. Asmussen E. Positive and Negative Muscular Work. *ACTA Physiol. Scand.*, 28, 365–382, 1953.
14. Avela, J. and Komi, P.V., Reduced stretch reflex sensitivity and muscle stiffness after long-lasting stretch-shortening cycle exercise in humans, *Eur. J. Appl. Physiol. Occup. Physiol.*, 78(5), 403–410, 1998.

15. Baggiolini, M., Dewald, B., Moser B., Chemokines., In: Gallin, J.I., Snyderman, R., editors. *Inflammation: Basic Principles and Clinical Correlates*. 3rd ed., Lippincott Williams & Wilkins, Philadelphia, 1999, pp. 419–431.
16. Ballor, D.L., Tommerup, L.J., Smith, D.B., and Thomas, D.P., Body composition, muscle and fat pad changes following two levels of dietary restriction and/or exercise training in male rats, *Int. J. Obes.*, 14(8), 711–722, 1990.
17. Balnave, C.D. and Thompson, M.W., Effect of training on eccentric exercise-induced muscle damage, *J. Appl. Physiol.*, 75(4), 1545–1551, 1993.
18. Bar, P.R., Amelink, G.J., Oldenburg, B., and Blankenstein, M.A., Prevention of exercise-induced muscle membrane damage by oestradiol, *Life Sci.*, 42(26), 2677–2681, 1988.
19. Barbe, M.F., Barr, A.E., Gorzelany, I., Amin, M., Gaughan, J.P., and Safadi, F.F., Chronic repetitive reaching and grasping results in decreased motor performance and widespread tissue responses in a rat model of MSD, *J. of Orthopaed. Res.*, 21(1), 167–176, 2003.
20. Barr, A.E. and Barbe, M.F., Pathophysiological tissue changes associated with repetitive movement: a review of the evidence, *Phys. Ther.*, 82(2), 173–187, 2002.
21. Belcastro, A.N., Skeletal muscle calcium-activated neutral protease (calpain) with exercise, *J. Appl. Physiol.*, 74(3), 1381–1386, 1993.
22. Belcastro, A.N., Shewchuk, L.D., and Raj, D.A., Exercise-induced muscle injury: a calpain hypothesis, *Mol. Cell. Biochem.*, 179(1–2), 135–145, 1998.
23. Benz, R.J., Friden, J., and Lieber, R.L., Simultaneous stiffness and force measurements reveal subtle injury to rabbit soleus muscles, *Mol. Cell. Biochem.*, 179(1–2), 147–158, 1998.
24. Bernard, B.P., Putz-Anderson, V., Burt, S.E., Cole, L.L., Fairfield-Estill, C., Fine, L.J. et al., Musculoskeletal disorders and workplace factors. A critical review of epidemiologic evidence for work-related musculoskeletal disorders of the neck, upper extremity, and low back, Cincinnati: NIOSH; 1997. Report No.: DHHS (NIOSH) Pub no. 97–141.
25. Best, T.M. and Hunter, K.D., Muscle injury and repair, *Phys. Med. Rehabil. Clin. N. Am.*, 11(2), 251–266, 2000.
26. Best, T.M., McCabe, R.P., Corr, D., Vanderby, R., Jr., Evaluation of a new method to create a standardized muscle stretch injury, *Med. Sci. Sports Exerc.*, 30(2), 200–205, 1998.
27. Best, T.M., Shehadeh, S.E., Leverson, G., Michel, J.T., Corr, D.T., Aeschlimann, D., Analysis of changes in mRNA levels of myoblast- and fibroblast-derived gene products in healing skeletal muscle using quantitative reverse transcription-polymerase chain reaction, *J. Orthop. Res.*, 19(4), 565–572, 2001.
28. Bigland-Ritchie, B., Graichen, H., and Woods, J.J., A variable-speed motorized bicycle ergometer for positive and negative work exercise, *J. Appl. Physiol.*, 35(5), 739–740, 1973.
29. Bigland-Ritchie, B. and Woods, J., Proceedings: Oxygen consumption and integrated electrical activity of muscle during positive and negative work, *J. Physiol.*, 234(2), 39P–40P, 1973.
30. Blangsted, A.K., Sogaard, K., Christensen, H., and Sjogaard G., The effect of physical and psychosocial loads on the trapezius muscle activity during computer keying tasks and rest periods, *Eur. J. Appl. Physiol.*, 2003.
31. Bobbert, M.F., Huijing, P.A., and van Ingen Schenau, G.J., A model of the human triceps surae muscle-tendon complex applied to jumping, *J. Biomech.*, 19(11), 887–898, 1986.
32. Bonde-Petersen, F., Henriksson, J., and Knuttgen, H.G., Effect of training with eccentric muscle contractions on skeletal muscle metabolites, *Acta. Physiol. Scand.*, 88(4), 564–570, 1973.
33. Bonde-Petersen, F., Knuttgen, H.G., and Henriksson, J., Muscle metabolism during exercise with concentric and eccentric contractions, *J. Appl. Physiol.*, 33(6), 792–795, 1972.
34. Borsa, P.A. and Sauers, E.L., The importance of gender on myokinetic deficits before and after microinjury, *Med. Sci. Sports Exerc.*, 32(5), 891–896, 2000.
35. Brooks, S.V. and Faulkner, J.A., Contraction-induced injury: recovery of skeletal muscles in young and old mice, *Am. J. Physiol.*, 258(3 Pt 1), C436–C442, 1990.
36. Brooks, S.V. and Faulkner, J.A., Isometric, shortening, and lengthening contractions of muscle fiber segments from adult and old mice, *Am. J. Physiol.*, 267(2 Pt 1), C507–C513, 1994.

37. Brooks, S.V. and Faulkner, J.A., The magnitude of the initial injury induced by stretches of maximally activated muscle fibres of mice and rats increases in old age, *J. Physiol.*, 497(Pt 2), 573–580, 1996.
38. Brooks, S.V. and Faulkner, J.A., Severity of contraction-induced injury is affected by velocity only during stretches of large strain, *J. Appl. Physiol.*, 91(2), 661–666, 2001.
39. Brooks, S.V., Zerba, E., and Faulkner, J.A., Injury to muscle fibres after single stretches of passive and maximally stimulated muscles in mice, *J. Physiol.*, 488(Pt 2), 459–469, 1995.
40. Brown, S.J., Child, R.B., Day, S.H., and Donnelly, A.E., Exercise-induced skeletal muscle damage and adaptation following repeated bouts of eccentric muscle contractions, *J. Sports Sci.*, 15(2), 215–222, 1997.
41. Byl, N., Wilson, F., Merzenich, M., Melnick, M., Scott, P., Oakes, A. et al., Sensory dysfunction associated with repetitive strain injuries of tendinitis and focal hand dystonia: a comparative study, *J. Orthop. Sports Phys. Ther.*, 23(4), 234–244, 1996.
42. Byrd, S.K., Alterations in the sarcoplasmic reticulum: a possible link to exercise-induced muscle damage, *Med. Sci. Sports Exerc.*, 24(5), 531–536, 1992.
43. Cantini, M., Massimino, M.L., Bruson, A., Catani, C., Dalla Libera, L., and Carraro U., Macrophages regulate proliferation and differentiation of satellite cells, *Biochem. Biophys. Res. Commun.*, 202(3), 1688–1696, 1994.
44. Cantini, M., Massimino, M.L., Rapizzi, E., Rossini, K., Catani, C., Dalla Libera, L. et al., Human satellite cell proliferation in vitro is regulated by autocrine secretion of IL-6 stimulated by a soluble factor(s) released by activated monocytes, *Biochem. Biophys. Res. Commun.*, 216(1), 49–53, 1995.
45. Carbo, N., Lopez-Soriano, J., Costelli, P., Busquets, S., Alvarez, B., Baccino, F.M. et al., Interleukin-15 antagonizes muscle protein waste in tumour-bearing rats, *Br. J. Cancer*, 83(4), 526–531, 2000.
46. Carpenter, J.E., Flanagan, C.L., Thomopoulos, S., Yian, E.H., and Soslowsky, L.J., The effects of overuse combined with intrinsic or extrinsic alterations in an animal model of rotator cuff tendinosis, *Am. J. Sports Med.*, 26(6), 801–807, 1998.
47. Carrasco, M.A., Riveros, N., Rios, J., Muller, M., Torres, F., Pineda, J. et al., Depolarization-induced slow calcium transients activate early genes in skeletal muscle cells, *Am. J. Physiol. Cell Physiol.*, 284(6), C1438–C1447, 2003.
48. Cavanagh, P., On "muscle action" vs "muscle contraction", *J. Biomech.*, 21, 69, 1988.
49. Chen, M.J., Shih, C.L., and Wang, K., Nebulin as an actin zipper. A two-module nebulin fragment promotes actin nucleation and stabilizes actin filaments, *J. Biol. Chem.*, 268(27), 20327–20334, 1993.
50. Chen, T.C. and Hsieh, S.S., Effects of a 7-day eccentric training period on muscle damage and inflammation, *Med. Sci. Sports Exerc.*, 33(10), 1732–1738, 2001.
51. Childs, T.E., Spangenburg, E.E., Vyas, D.R., and Booth, F.W., Temporal alterations in protein signaling cascades during recovery from muscle atrophy, *Am. J. Physiol. Cell Physiol.*, 285(2), C391–C398, 2003.
52. Chou, S.M. and Mizuno, Y., Induction of spheroid cytoplasmic bodies in a rat muscle by local tetanus, *Muscle Nerve*, 9(5), 455–464, 1986.
53. Clarkson, P.M., Eccentric exercise and muscle damage. *International Journal of Sports Medicine*, 18(Suppl 4), S314–S317, 1997.
54. Clarkson, P.M., Apple, F.S., Byrnes, W.C., McCormick, K.M., and Triffletti, P., Creatine kinase isoforms following isometric exercise, *Muscle Nerve*, 10(1), 41–44, 1987.
55. Clarkson, P.M., Byrnes, W.C., McCormick, K.M., Turcotte, L.P., and White, J.S., Muscle soreness and serum creatine kinase activity following isometric, eccentric, and concentric exercise, *Int. J. Sports Med.*, 7(3), 152–155, 1986.
56. Clarkson, P.M. and Ebbeling, C., Investigation of serum creatine kinase variability after muscle-damaging exercise, *Clin. Sci. (Lond.)*, 75(3), 257–261, 1988.
57. Clarkson, P.M. and Hubal, M.J., Are women less susceptible to exercise-induced muscle damage?, *Curr. Opin. Clin. Nutr. Metab. Care*, 4(6), 527–531, 2001.

58. Clarkson, P.M. and Hubal, M.J., Exercise-induced muscle damage in humans. *Am. J. Phys. Med. Rehabil.*, 81(11 Suppl), S52–S69, 2002.
59. Clarkson, P.M., Nosaka, K., and Braun, B., Muscle function after exercise-induced muscle damage and rapid adaptation, *Med. Sci. Sports Exerc.*, 24(5), 512–520, 1992.
60. Clarkson, P.M. and Tremblay, I., Exercise-induced muscle damage, repair, and adaptation in humans, *J. Appl. Physiol.*, 65(1), 1–6, 1988.
61. Cutlip, R.G. The Dynamic Parameters that Affect Eccentric-Contraction Induced Injury in an In Vivo Rodent Model [Doctoral Dissertation]: West Virginia University; 1995.
62. Cutlip, R.G., Geronilla, K.B., Baker, B.A., Kashon, M.L., Miller, G.R., and Schopper, A.W., Impact of muscle length during stretch-shortening contractions on real-time and temporal muscle performance measures in rats in vivo, *J. Appl. Physiol.*, 96(2), 507–516, 2004.
63. Cutlip, R.G., Stauber, W.T., Willison, R.H., McIntosh, T.A., and Means, K.H., Dynamometer for rat plantar flexor muscles in vivo, *Med. Biol. Eng. Comput.*, 35(5), 540–543, 1997.
64. De Bleecker, J.L., Meire, V.I., Declercq, W., and Van Aken, E.H., Immunolocalization of tumor necrosis factor-alpha and its receptors in inflammatory myopathies, *Neuromuscul. Disord.*, 9(4), 239–246, 1999.
65. Devor, S.T. and Faulkner, J.A., Regeneration of new fibers in muscles of old rats reduces contraction-induced injury, *J. Appl. Physiol.*, 87(2), 750–756, 1999.
66. Duan, C., Delp, M.D., Hayes, D.A., Delp, P.D., and Armstrong, R.B., Rat skeletal muscle mitochondrial [Ca2+] and injury from downhill walking, *J. Appl. Physiol.*, 68(3), 1241–1251, 1990.
67. Dubravec, D.B., Spriggs, D.R., Mannick, J.A., and Rodrick, M.L., Circulating human peripheral blood granulocytes synthesize and secrete tumor necrosis factor alpha, *Proc. Natl. Acad. Sci. U.S.A.*, 87(17), 6758–6761, 1990.
68. Edman, K.A., The velocity of unloaded shortening and its relation to sarcomere length and isometric force in vertebrate muscle fibres, *J. Physiol.*, 291, 143–159, 1979.
69. Eston, R.G., Finney, S., Baker, S., and Baltzopoulos, V., Muscle tenderness and peak torque changes after downhill running following a prior bout of isokinetic eccentric exercise, *J. Sports Sci.*, 14(4), 291–299, 1996.
70. Eston, R.G., Lemmey, A.B., McHugh, P., Byrne, C., and Walsh, S.E., Effect of stride length on symptoms of exercise-induced muscle damage during a repeated bout of downhill running, *Scand. J. Med. Sci. Sports*, 10(4), 199–204, 2000.
71. Farrell, P.A., Fedele, M.J., Hernandez, J., Fluckey, J.D., Miller, J.L., 3rd, Lang, C.H. et al., Hypertrophy of skeletal muscle in diabetic rats in response to chronic resistance exercise, *J. Appl. Physiol.*, 87(3), 1075–1082, 1999.
72. Faulkner, J.A., Brooks, S.V., and Zerba, E., Muscle atrophy and weakness with aging: contraction-induced injury as an underlying mechanism, *J. Gerontol. A. Biol. Sci. Med. Sci.*, 50 Spec No:124–129, 1995.
73. Faulkner, J.A., Jones, D.A., and Round, J.M., An Apparatus for Unilateral Exercise of the Lower Limb Muscles of Small Mammals, *J. Physiol. Lond.*, 365, 11P, 1985.
74. Faulkner, J.A., Jones, D.A., and Round, J.M., Injury to Skeletal Muscle of Mice by Lengthening Contractions. *J. of Physiol., Lond.*, 365, 75P, 1985.
75. Faulkner, J.A., Jones, D.A., and Round, J.M., Injury to skeletal muscles of mice by forced lengthening during contractions, *Q. J. Exp. Physiol.*, 74(5), 661–670, 1989.
76. Faulkner, J.A., Opiteck, J.A., and Brooks, S.V., Injury to skeletal muscle during altitude training: induction and prevention, *Int. J. Sports Med.*, 13 Suppl 1, S160–S162, 1992.
77. Fibbi, G., D'Alessio, S., Pucci, M., Cerletti, M., and Del Rosso, M., Growth factor-dependent proliferation and invasion of muscle satellite cells require the cell-associated fibrinolytic system, *Biol. Chem.*, 383(1), 127–136, 2002.
78. Fielding, R.A., Evans, W.J., Hughes, V.A., Moldawer, L.L., and Bistrian, B.R., The effects of high intensity exercise on muscle and plasma levels of alpha-ketoisocaproic acid, *Eur. J. Appl. Physiol. Occup. Physiol.*, 55(5), 482–485, 1986.

79. Fielding, R.A., Manfredi, T.J., Ding, W., Fiatarone, M.A., Evans, W.J., and Cannon, J.G., Acute phase response in exercise. III. Neutrophil and IL-1 beta accumulation in skeletal muscle, *Am. J. Physiol.*, 265(1 Pt 2), R166–R172, 1993.
80. Fitts, R.H., Cellular mechanisms of muscle fatigue, *Physiol. Rev.*, 74(1), 49–94, 1994.
81. Fitts, R.H., Muscle fatigue: the cellular aspects, *Am. J. Sports Med.*, 24(6 Suppl), S9–S13, 1996.
82. Fitts, R.H. and Holloszy, J.O., Effects of fatigue and recovery on contractile properties of frog muscle, *J. Appl. Physiol.*, 45(6), 899–902, 1978.
83. Fluck, M., Carson, J.A., Schwartz, R.J., and Booth, F.W., SRF protein is upregulated during stretch-induced hypertrophy of rooster ALD muscle, *J. Appl. Physiol.*, 86(6), 1793–1799, 1999.
84. Fluckey, J.D., Kraemer, W.J., and Farrell, P.A., Pancreatic islet insulin secretion is increased after resistance exercise in rats, *J. Appl. Physiol.*, 79(4), 1100–1105, 1995.
85. Foley, J.M., Jayaraman, R.C., Prior, B.M., Pivarnik, J.M., and Meyer, R.A., MR measurements of muscle damage and adaptation after eccentric exercise, *J. of Appl. Physiol.*, 87(6), 2311–2318, 1999.
86. Friden, J. and Lieber, R.L., Segmental muscle fiber lesions after repetitive eccentric contractions, *Cell Tissue Res.*, 293(1), 165–171, 1998.
87. Friden, J., Lieber, R.L., and Thornell, L.E., Subtle indications of muscle damage following eccentric contractions, *Acta Physiol. Scand.*, 142(4), 523–524, 1991.
88. Friden, J., Seger, J., Sjostrom, M., and Ekblom B. Adaptive response in human skeletal muscle subjected to prolonged eccentric training, *Int. J. Sports Med.*, 4(3), 177–183, 1983.
89. Friden, J., Sfakianos, P.N., and Hargens, A.R., Muscle soreness and intramuscular fluid pressure: comparison between eccentric and concentric load, *J. Appl. Physiol.*, 61(6), 2175–2179, 1986.
90. Friden, J., Sjostrom, M., and Ekblom, B., A morphological study of delayed muscle soreness, *Experientia*, 37(5), 506–507, 1981.
91. Friden, J., Sjostrom, M., and Ekblom, B., Myofibrillar damage following intense eccentric exercise in man, *Int. J. Sports Med.*, 4(3), 170–176, 1983.
92. Fritz, V.K. and Stauber, W.T., Characterization of muscles injured by forced lengthening, *II. Proteoglycans. Med. Sci. Sports Exerc.*, 20(4), 354–361, 1988.
93. Gallucci, S., Provenzano, C., Mazzarelli, P., Scuderi, F., and Bartoccioni, E., Myoblasts produce IL-6 in response to inflammatory stimuli, *Int. Immunol.*, 10(3), 267–273, 1998.
94. Garner, R.P., Terracio, L., Borg, T.K., and Buggy, J., Intracranial self-stimulation motivates weight-lifting exercise in rats, *J. Appl. Physiol.*, 71(4), 1627–1631, 1991.
95. Garrett, W.E., Jr., Muscle strain injuries. *Am. J. Sports Med.*, 24(6 Suppl), S2–S8, 1996.
96. Geronilla, K.B., Miller, G.R., Mowrey, K., Kashon, M.L., and Cutlip, R.G., The Mechanical and Histological Response of Rat Skeletal Muscle to Oscillatory Contractions, In: American College of Sports Medicine; 2001, *Medicine and Science in Sports and Exercise, Baltimore*, MD 2001, p. 474.
97. Geronilla, K.B., Miller, G.R., Mowrey, K.F., Wu, J.Z., Kashon, M.L., Brumbaugh, K. et al., Dynamic force responses of skeletal muscle during stretch-shortening cycles, *Eur. J. Appl. Physiol.*, 90(1–2), 144–153, 2003.
98. Giddings, C.J., Neaves, W.B., and Gonyea, W.J., Muscle fiber necrosis and regeneration induced by prolonged weight-lifting exercise in the cat, *Anat. Rec.*, 211(2), 133–141, 1985.
99. Gillis, J.M., Membrane abnormalities and ca homeostasis in muscles of the mdx mouse, an animal model of the duchenne muscular dystrophy: a review, *Acta Physiol. Scand.*, 156(3), 397–406, 1996.
100. Gonyea, W. and Bonde-Petersen, F., Alterations in muscle contractile properties and fiber composition after weight-lifting exercise in cats, *Exp. Neurol.*, 59(1), 75–84, 1978.
101. Gonyea, W.J., Role of exercise in inducing increases in skeletal muscle fiber number, *J. Appl. Physiol.*, 48(3), 421–426, 1980.
102. Gonyea, W.J. and Ericson, G.C., An experimental model for the study of exercise-induced skeletal muscle hypertrophy, *J. Appl. Physiol.*, 40(4), 630–633, 1976.
103. Gordon, A.M., Huxley, A.F., and Julian, F.J., The variation in isometric tension with sarcomere length in vertebrate muscle fibres, *J. Physiol.*, 184(1), 170–192, 1966.

104. Gordon, E.E. Anatomical and biochemical adaptations of muscle to different exercises, *Jama.*, 201(10), 755–758, 1967.
105. Gordon, E.E., Kowalski, K., and Fritts M., Adaptations of muscle to various exercises, *Studies in rats. Jama.*, 199(2), 103–108, 1967.
106. Gordon, E.E., Kowalski, K., and Fritts, M., Changes in rat muscle fiber with forceful exercises, *Arch. Phys. Med. Rehabil.*, 48(11), 577–582, 1967.
107. Gordon, E.E., Kowalski, K., and Fritts, M. Protein changes in quadriceps muscle of rat with repetitive exercises, *Arch. Phys. Med. Rehabil.*, 48(6), 296–303, 1967.
108. Gorospe, J.R., Tharp, M.D., Hinckley, J., Kornegay, J.N., and Hoffman, E.P., A role for mast cells in the progression of Duchenne muscular dystrophy? Correlations in dystrophin-deficient humans, dogs, and mice, *J. Neurol. Sci.*, 122(1), 44–56, 1994.
109. Gosselin, L.E., Attenuation of force deficit after lengthening contractions in soleus muscle from trained rats, *J. Appl. Physiol.*, 88(4), 1254–1258, 2000.
110. Gosselin, L.E. and Burton H., Impact of initial muscle length on force deficit following lengthening contractions in mammalian skeletal muscle, *Muscle Nerve*, 25(6), 822–827, 2002.
111. Hales, T.R. and Bernard, B.P., Epidemiology of work-related musculoskeletal disorders, *Orthop. Clin. North Am.*, 27(4), 679–709, 1996.
112. Hasselgren, P.O., Role of the ubiquitin-proteasome pathway in sepsis-induced muscle catabolism, *Mol. Biol. Rep.*, 26(1–2), 71–76, 1999.
113. Hasselgren, P.O., Pedersen, P., Sax, H.C., Warner, B.W., and Fischer, J.E., Current concepts of protein turnover and amino acid transport in liver and skeletal muscle during sepsis, *Arch. Surg.*, 123(8), 992–999, 1988.
114. Hasselman, C.T., Best, T.M., Seaber, A.V., and Garrett, W.E., Jr. A threshold and continuum of injury during active stretch of rabbit skeletal muscle, *Am. J. Sports Med.*, 23(1), 65–73, 1995.
115. Henriksson, J., Knuttgen, H.G., and Bonde-Petersen, F., Perceived exertion during exercise with concentric and eccentric muscle contractions, *Ergonomics*, 15(5), 537–544, 1972.
116. Hesselink, M.K., Kuipers, H., Geurten, P., and Van Straaten, H., Structural muscle damage and muscle strength after incremental number of isometric and forced lengthening contractions, *J. Muscle Res. Cell. Motil.*, 17(3), 335–341, 1996.
117. Hilder, T.L., Tou, J.C., Grindeland, R.E., Wade, C.E., and Graves, L.M., Phosphorylation of insulin receptor substrate-1 serine 307 correlates with JNK activity in atrophic skeletal muscle, *FEBS Lett.*, 553(1–2), 63–67, 2003.
118. Hill, A.V., The heat of shortening and the dynamic constants of muscle, *Proc. R. Soc. London Ser. B*, 126, 1938.
119. Hill, M., Wernig, A., and Goldspink, G., Muscle satellite (stem) cell activation during local tissue injury and repair, *J. Anat.*, 203(1), 89–99, 2003.
120. Ho, K.W., Roy, R.R., Tweedle, C.D., Heusner, W.W., Van Huss, W.D., and Carrow, R.E., Skeletal muscle fiber splitting with weight-lifting exercise in rats. *Am. J. Anatomy*, 157, 433–440, 1980.
121. Hoffman, E.P., Brown, R.H., Jr., and Kunkel, L.M., Dystrophin: the protein product of the Duchenne muscular dystrophy locus, *Cell*, 51(6), 919–928, 1987.
122. Hoffman, E.P., Knudson, C.M., Campbell, K.P., and Kunkel, L.M., Subcellular fractionation of dystrophin to the triads of skeletal muscle, *Nature*, 330(6150), 754–758, 1987.
123. Hopf, F.W., Reddy, P., Hong, J., and Steinhardt, R.A., A capacitative calcium current in cultured skeletal muscle cells is mediated by the calcium-specific leak channel and inhibited by dihydropyridine compounds, *J. Biol. Chem.*, 271(37), 22358–22367, 1996.
124. Hopf, F.W., Turner, P.R., Denetclaw, W.F., Jr., Reddy, P., and Steinhardt, R.A., A critical evaluation of resting intracellular free calcium regulation in dystrophic mdx muscle. *Am. J. Physiol.*, 271(4 Pt 1), C1325–C1339, 1996.
125. Horita, T., Komi, P.V., Nicol, C., and Kyrolainen, H., Effect of exhausting stretch-shortening cycle exercise on the time course of mechanical behaviour in the drop jump: possible role of muscle damage, *Eur. J. Appl. Physiol. Occup. Physiol.*, 79(2), 160–167, 1999.

126. Hough, T., Ergographic Studies in Muscular Soreness. *Am. J. Physiol.*, 7, 76–92, 1902.
127. Howell, J.N., Chleboun, G., and Conatser R., Muscle stiffness, strength loss, swelling and soreness following exercise-induced injury in humans. *J. Physiol.*, 464, 183–196, 1993.
128. Huijing, P.A., Architecture of the human gastrocnemius muscle and some functional consequences, *Acta Anat. (Basel)*, 123(2), 101–107, 1985.
129. Hunter, K.D. and Faulkner, J.A., Pliometric contraction-induced injury of mouse skeletal muscle: effect of initial length, *J. Appl. Physiol.*, 82(1), 278–283, 1997.
130. Ingalls, C.P., Warren, G.L., and Armstrong, R.B., Dissociation of force production from MHC and actin contents in muscles injured by eccentric contractions, *J. Muscle Res. Cell Motil.*, 19(3), 215–224, 1998.
131. Ingalls, C.P., Warren, G.L., and Armstrong, R.B., Intracellular Ca2+ transients in mouse soleus muscle after hindlimb unloading and reloading, *J. Appl. Physiol.*, 87(1), 386–390, 1999.
132. Ingalls, C.P., Warren, G.L., Williams, J.H., Ward, C.W., and Armstrong, R.B., E-C coupling failure in mouse EDL muscle after in vivo eccentric contractions, *J. Appl. Physiol.*, 85(1), 58–67, 1998.
133. Jensen, B.R., Schibye, B., Sogaard, K., Simonsen, E.B., and Sjogaard, G., Shoulder muscle load and muscle fatigue among industrial sewing-machine operators, *Eur. J. Appl. Physiol. Occup. Physiol.*, 67(5), 467–475, 1993.
134. Jensen, B.R., Sjogaard, G., Bornmyr, S., Arborelius, M., and Jorgensen, K., Intramuscular laser-Doppler flowmetry in the supraspinatus muscle during isometric contractions, *Eur. J. Appl. Physiol. Occup. Physiol.*, 71(4), 373–378, 1995.
135. Jones, D.A., Jackson, M.J., McPhail, G., and Edwards, R.H., Experimental mouse muscle damage: the importance of external calcium, *Clin. Sci. (Lond)*, 66(3), 317–322, 1984.
136. Jones, D.A., Newham, D.J., Round, J.M., and Tolfree, S.E., Experimental human muscle damage: morphological changes in relation to other indices of damage, *J. Physiol.*, 375, 435–448, 1986.
137. Kadi, F., Hagg, G., Hakansson, R., Holmner, S., Butler-Browne, G.S., and Thornell, L.-E., Structural changes in male trapezius muscle with work-related myalgia, *Acta Neurophathol.*, 95, 352–360, 1998.
138. Keller, K., Corbett, J., and Nichols, D., Repetitive strain injury in computer keyboard users: Pathomechanics and treatment principles in individual and group intervention, *J. Hand Therapy*, 11, 9–26, 1998.
139. Kissel, J.T., Lynn, D.J., Rammohan, K.W., Klein, J.P., Griggs, R.C., and Moxley, R.T., 3rd, et al., Mononuclear cell analysis of muscle biopsies in prednisone- and azathioprine-treated Duchenne muscular dystrophy, *Neurology*, 43(3 Pt 1), 532–536, 1993.
140. Klausen, K. and Knuttgen, H.G., Effect of training on oxygen consumption in negative muscular work, *Acta. Physiol. Scand.*, 83(3), 319–323, 1971.
141. Klitgaard, H., A model for quantitative strength training of hindlimb muscles of the rat, *J. Appl. Physiol.*, 64(4), 1740–1745, 1988.
142. Knutsson, E., Analysis of spastic paresis. In: Proceedings of the Tenth International Congress of the World Confederation for Physical Therapy, 1987, Sydney; 1987. p. 629–633.
143. Knuttgen, H.G., Oxygen debt, lactate, pyruvate, and excess lactate after muscular work, *J. Appl. Physiol.*, 17, 639–644, 1962.
144. Knuttgen, H.G., Human performance in high-intensity exercise with concentric and eccentric muscle contractions. *Int. J. Sports Med.*, 7 Suppl 1, 6–9, 1986.
145. Knuttgen, H.G. and Klausen K., Oxygen debt in short-term exercise with concentric and eccentric muscle contractions, *J. Appl. Physiol.*, 30(5), 632–635, 1971.
146. Knuttgen, H.G., Nadel, E.R., Pandolf, K.B., and Patton, J.F., Effects of training with eccentric muscle contractions on exercise performance, energy expenditure, and body temperature. *Int. J. Sports Med.*, 3(1), 13–17, 1982.
147. Knuttgen, H.G., Petersen, F.B., and Klausen K., Oxygen uptake and heart rate responses to exercise performed with concentric and eccentric muscle contractions. *Med. Sci. Sports*, 3(1), 1–5, 1971.

148. Komi, P.V., Measurement of the force-velocity relationship in human muscle under concentric and eccentric contractions, *Medicine and Sport*, 8, 224–229, 1973.
149. Komi, P.V., Stretch-shortening cycle: a powerful model to study normal and fatigued muscle, *J. Biomech.*, 33(10), 1197–1206, 2000.
150. Komi, P.V. and Buskirk, E.R., Effect of eccentric and concentric muscle conditioning on tension and electrical activity of human muscle, *Ergonomics*, 15(4), 417–434, 1972.
151. Komi, P.V. and Rusko, H., Quantitative evaluation of mechanical and electrical changes during fatigue loading of eccentric and concentric work. *Scand. J. Rehabil. Med.*, Suppl 3, 121–126, 1974.
152. Komulainen, J., Kalliokoski, R., Koskinen, S.O., Drost, M.R., Kuipers, H., and Hesselink, M.K., Controlled lengthening or shortening contraction-induced damage is followed by fiber hypertrophy in rat skeletal muscle, *Int. J. Sports Med.*, 21(2), 107–112, 2000.
153. Komulainen, J., Koskinen, S.O., Kalliokoski, R., Takala, T.E., and Vihko, V., Gender differences in skeletal muscle fibre damage after eccentrically biased downhill running in rats, *Acta. Physiol. Scand.*, 165(1), 57–63, 1999.
154. Komulainen, J., Takala, T.E., Kuipers, H., and Hesselink, M.K., The disruption of myofibre structures in rat skeletal muscle after forced lengthening contractions, *Pflugers Arch.*, 436(5), 735–741, 1998.
155. Kuipers, H. and Van der Meulen, J.H., Increased Passive Stiffness of Rat Muscle after Eccentric and Isometric Work, *Medicine and Science in Sports and Exercise*, 24(5), S1007, 1992.
156. Lapointe, B.M., Fremont, P., and Cote, C.H., Adaptation to lengthening contractions is independent of voluntary muscle recruitment but relies on inflammation. *Am. J. Physiol. Regul. Integr. Comp. Physiol.*, 282(1), R323–R329, 2002.
157. Lapointe, B.M., Frenette, J., and Cote, C.H., Lengthening contraction-induced inflammation is linked to secondary damage but devoid of neutrophil invasion, *J. Appl. Physiol.*, 92(5), 1995–2004, 2002.
158. Larsson, B., Bjork, J., Henriksson, K.G., Gerdle, B. and Lindman, R., The prevalences of cytochrome c oxidase negative and superpositive fibres and ragged-red fibres in the trapezius muscle of female cleaners with and without myalgia and of female healthy controls, *Pain*, 84(2–3), 379–387, 2000.
159. Larsson, S.E., Bodegard, L., Henriksson, K.G., and Oberg, P.A., Chronic trapezius myalgia. Morphology and blood flow studied in 17 patients, *Acta Orthop. Scand.*, 61(5), 394–398, 1990.
160. Lazarides, E., Intermediate filaments as mechanical integrators of cellular space, *Nature*, 283(5744), 249–256, 1980.
161. Lieber, R.L. and Friden, J., Selective damage of fast glycolytic muscle fibres with eccentric contraction of the rabbit tibialis anterior, *Acta Physiol. Scand.*, 133(4), 587–588, 1988.
162. Lieber, R.L. and Friden, J., Muscle damage is not a function of muscle force but active muscle strain, *J. Appl. Physiol.*, 74(2), 520–526, 1993.
163. Lieber, R.L., Schmitz, M.C., Mishra, D.K., and Friden, J., Contractile and cellular remodeling in rabbit skeletal muscle after cyclic eccentric contractions, *J. Appl. Physiol.*, 77(4), 1926–1934, 1994.
164. Lieber, R.L., Thornell, L.E., and Friden, J., Muscle cytoskeletal disruption occurs within the first 15 min of cyclic eccentric contraction, *J. Appl. Physiol.*, 80(1), 278–284, 1996.
165. Lieber, R.L., Woodburn, T.M., and Friden, J., Muscle damage induced by eccentric contractions of 25% strain, *J. Appl. Physiol.*, 70(6), 2498–2507, 1991.
166. Lowe, D.A., and Alway, S.E., Stretch-induced myogenin, MyoD, and MRF4 expression and acute hypertrophy in quail slow-tonic muscle are not dependent upon satellite cell proliferation, *Cell Tissue Res.*, 296(3), 531–539, 1999.
167. Lowe, D.A., Warren, G.L., Hayes, D.A., Farmer, M.A., and Armstrong, R.B., Eccentric contraction-induced injury of mouse soleus muscle: effect of varying [Ca2+]o, *J. Appl. Physiol.*, 76(4), 1445–1453, 1994.
168. Lowe, D.A., Warren, G.L., Ingalls, C.P., Boorstein, D.B., and Armstrong, R.B., Muscle function and protein metabolism after initiation of eccentric contraction-induced injury, *Journal of Applied Physiology*, 79(4), 1260–1270, 1995.

169. Lynch, G.S. and Faulkner, J.A., Contraction-induced injury to single muscle fibers: velocity of stretch does not influence the force deficit, *Am. J. Physiol.*, 275(6 Pt 1), C1548–C1554, 1998.
170. Lynn, R. and Morgan, D.L., Decline running produces more sarcomeres in rat vastus intermedius muscle fibers than does incline running, *J. Appl. Physiol.*, 77(3), 1439–1444, 1994.
171. Macpherson, P.C., Dennis, R.G., and Faulkner, J.A., Sarcomere dynamics and contraction-induced injury to maximally activated single muscle fibres from soleus muscles of rats, *J. Physiol.*, 500 (Pt 2), 523–533, 1997.
172. Macpherson, P.C., Schork, M.A., and Faulkner, J.A., Contraction-induced injury to single fiber segments from fast and slow muscles of rats by single stretches, *Am. J. Physiol.*, 271(5 Pt 1), C1438–C1446, 1996.
173. Mair, J., Koller, A., Artner-Dworzak, E., Haid, C., Wicke, K., Judmaier, W. et al., Effects of exercise on plasma myosin heavy chain fragments and MRI of skeletal muscle, *J. Appl. Physiol.*, 72(2), 656–663, 1992.
174. Mair, J., Mayr, M., Muller, E., Koller, A., Haid, C., Artner-Dworzak, E. et al., Rapid adaptation to eccentric exercise-induced muscle damage, *Int. J. Sports Med.*, 16(6), 352–356, 1995.
175. Malm, C., Nyberg, P., Engstrom, M., Sjodin, B., Lenkei, R., Ekblom, B. et al., Immunological changes in human skeletal muscle and blood after eccentric exercise and multiple biopsies, *J. Physiol.*, 529 Pt 1, 243–262, 2000.
176. McArdle, A., van der Meulen, J.H., Catapano, M., Symons, M.C., Faulkner, J.A., and Jackson, M.J., Free radical activity following contraction-induced injury to the extensor digitorum longus muscles of rats, *Free Radic. Biol. Med.*, 26(9-10), 1085–1091, 1999.
177. McCully, K.K. and Faulkner, J.A., Injury to skeletal muscle fibers of mice following lengthening contractions, *J. Appl. Physiol.*, 59(1), 119–126, 1985.
178. McCully, K.K. and Faulkner, J.A., Characteristics of lengthening contractions associated with injury to skeletal muscle fibers, *J. Appl. Physiol.*, 61(1), 293–299, 1986.
179. McHugh, M.P., Connolly, D.A., Eston, R.G., and Gleim, G.W., Exercise-induced muscle damage and potential mechanisms for the repeated bout effect, *Sports Medicine*, 27(3), 157–170, 1999.
180. Medina, R., Wing, S.S., and Goldberg, A.L., Increase in levels of polyubiquitin and proteasome mRNA in skeletal muscle during starvation and denervation atrophy, *Biochem. J.*, 307 (Pt 3), 631–637, 1995.
181. Mishra, D.K., Friden, J., Schmitz, M.C., and Lieber, R.L., Anti-inflammatory medication after muscle injury. A treatment resulting in short-term improvement but subsequent loss of muscle function, *J. Bone Joint Surg. Am.*, 77(10), 1510–1519, 1995.
182. Moldawer, L.L., Svaninger, G., Gelin, J., and Lundholm, K.G., Interleukin 1 and tumor necrosis factor do not regulate protein balance in skeletal muscle, *Am. J. Physiol.*, 253(6 Pt 1), C766–C773, 1987.
183. Morgan, D.L., New insights into the behavior of muscle during active lengthening, *Biophys. J.*, 57(2), 209–221, 1990.
184. Mutsaers, S.E., Bishop, J.E., McGrouther, G., and Laurent, G.J., Mechanisms of tissue repair: from wound healing to fibrosis, *International Journal of Biochemistry & Cell Biology*, 29(1), 5–17, 1997.
185. Myllyla, R., Salminen, A., Peltonen, L., Takala, T.E., and Vihko, V., Collagen metabolism of mouse skeletal muscle during the repair of exercise injuries, *Pflugers Arch.*, 407(1), 64–70, 1986.
186. Newham, D.J., Jones, D.A., and Clarkson, P.M., Repeated high-force eccentric exercise: effects on muscle pain and damage, *J. Appl. Physiol.*, 63(4), 1381–1386, 1987.
187. Newham, D.J., Jones, D.A., and Edwards, R.H., Large delayed plasma creatine kinase changes after stepping exercise, *Muscle Nerve*, 6(5), 380–385, 1983.
188. Newham, D.J., Jones, D.A., Ghosh, G., and Aurora, P., Muscle fatigue and pain after eccentric contractions at long and short length, *Clin. Sci. (Lond)*, 74(5), 553–557, 1988.
189. Newham, D.J., Jones, D.A., Tolfree, S.E., and Edwards, R.H., Skeletal muscle damage: a study of isotope uptake, enzyme efflux and pain after stepping, *Eur. J. Appl. Physiol. Occup. Physiol.*, 55(1), 106–112, 1986.

190. Newham, D.J., McPhail, G., Mills, K.R., and Edwards, R.H., Ultrastructural changes after concentric and eccentric contractions of human muscle, *J. Neurol. Sci.*, 61(1), 109–122, 1983.
191. Nikolaou, P.K., Macdonald, B.L., Glisson, R.R., Seaber, A.V., and Garrett, W.E., Jr., Biomechanical and histological evaluation of muscle after controlled strain injury, *Am. J. Sports Med.*, 15(1), 9–14, 1987.
192. Noonan, T.J., Best, T.M., Seaber, A.V., and Garrett, W.E. Jr., Identification of a threshold for skeletal muscle injury, *Am. J. Sports Med.*, 22(2), 257–261, 1994.
193. Nosaka, K. and Clarkson, P.M., Effect of eccentric exercise on plasma enzyme activities previously elevated by eccentric exercise, *Eur. J. Appl. Physiol. Occup. Physiol.*, 69(6), 492–497, 1994.
194. Nosaka, K. and Clarkson, P.M., Muscle damage following repeated bouts of high force eccentric exercise, *Med. Sci. Sports Exerc.*, 27(9), 1263–1269, 1995.
195. Nosaka, K. and Clarkson, P.M., Changes in indicators of inflammation after eccentric exercise of the elbow flexors, *Medicine & Science in Sports & Exercise*, 28(8), 953–961, 1996.
196. Nosaka, K. and Clarkson, P.M., Variability in serum creatine kinase response after eccentric exercise of the elbow flexors, *International Journal of Sports Medicine*, 17(2), 120–127, 1996.
197. Nosaka, K., Clarkson, P.M., and Apple, F.S., Time course of serum protein changes after strenuous exercise of the forearm flexors, *Journal of Laboratory & Clinical Medicine*, 119(2), 183–188, 1992.
198. Nosaka, K., Clarkson, P.M., McGuiggin, M.E., and Byrne, J.M., Time course of muscle adaptation after high force eccentric exercise, *Eur. J. Appl. Physiol. Occup. Physiol.*, 63(1), 70–76, 1991.
199. Nurenberg, P., Giddings, C.J., Stray-Gundersen, J., Fleckenstein, J.L., Gonyea, W.J., and Peshock, R.M., MR imaging-guided muscle biopsy for correlation of increased signal intensity with ultrastructural change and delayed-onset muscle soreness after exercise, *Radiology*, 184(3), 865–869, 1992.
200. Oak, S.A., Zhou, Y.W., and Jarrett, H.W., Skeletal muscle signaling pathway through the dystrophin glycoprotein complex and Rac1, *J. Biol. Chem.*, 278(41), 39287–39295, 2003.
201. Obremsky, W.T., Seaber, A.V., Ribbeck, B.M., and Garrett, W.E., Jr., Biomechanical and histologic assessment of a controlled muscle strain injury treated with piroxicam, *Am. J. Sports Med.*, 22(4), 558–561, 1994.
202. Ogilvie, R.W., Armstrong, R.B., Baird, K.E., and Bottoms, C.L., Lesions in the rat soleus muscle following eccentrically biased exercise, *Am. J. Anat.*, 182(4), 335–346, 1988.
203. Ortenblad, N., Sjogaard, G., and Madsen, K., Impaired sarcoplasmic reticulum Ca(2+) release rate after fatiguing stimulation in rat skeletal muscle, *J. Appl. Physiol.*, 89(1), 210–217, 2000.
204. Ortenblad, N., Young, J.F., Oksbjerg, N., Nielsen, J.H., and Lambert, I.H., Reactive oxygen species are important mediators of taurine release from skeletal muscle cells, *Am. J. Physiol. Cell Physiol.*, 284(6), C1362–C1373, 2003.
205. Pardo, J.V., Siliciano, J.D., and Craig, S.W., A vinculin-containing cortical lattice in skeletal muscle: transverse lattice elements ("costameres") mark sites of attachment between myofibrils and sarcolemma, *Proc. Natl. Acad. Sci. USA*, 80(4), 1008–1012, 1983.
206. Patel, T.J., Cuizon, D., Mathieu-Costello, O., Friden, J., and Lieber, R.L., Increased oxidative capacity does not protect skeletal muscle fibers from eccentric contraction-induced injury, *Am. J. Physiol.*, 274(5 Pt 2), R1300–R1308, 1998.
207. Patel, T.J. and Lieber, R.L., Force transmission in skeletal muscle: from actomyosin to external tendons, *Exerc. Sport Sci. Rev.*, 25, 321–363, 1997.
208. Petersen, F.B. and Knuttgen, H.G., Effect of training with eccentric muscle contractions on human skeletal muscle metabolites, *Acta. Physiol. Scand.*, 80(4), 16A–17A, 1970.
209. Peterson, J.M., Trappe, T.A., Mylona, E., White, F., Lambert, C.P., Evans, W.J. et al., Ibuprofen and acetaminophen: effect on muscle inflammation after eccentric exercise, *Med. Sci. Sports Exerc.*, 35(6), 892–896, 2003.
210. Petrof, B.J., Shrager, J.B., Stedman, H.H., Kelly, A.M., and Sweeney, H.L., Dystrophin protects the sarcolemma from stresses developed during muscle contraction, *Proc. Natl. Acad. Sci. USA*, 90(8), 3710–3714, 1993.

211. Piligian, G., Herbert, R., Hearns, M., Dropkin, J., Landsbergis, P., and Cherniack, M., Evaluation and management of chronic work-related musculoskeletal disorders of the distal upper extremity, *American Journal of Industrial Medicine*, 37(1), 75–93, 2000.
212. Porter, J.D., Khanna, S., Kaminski, H.J., Rao, J.S., Merriam, A.P., Richmonds, C.R. et al., A chronic inflammatory response dominates the skeletal muscle molecular signature in dystrophin-deficient mdx mice, *Hum. Mol. Genet.*, 11(3), 263–272, 2002.
213. Prisk, V. and Huard, J., Muscle injuries and repair: the role of prostaglandins and inflammation, *Histol. Histopathol.*, 18(4), 1243–1256, 2003.
214. Quinn, L.S., Anderson, B.G., Drivdahl, R.H., Alvarez, B., and Argiles, J.M., Overexpression of interleukin-15 induces skeletal muscle hypertrophy in vitro: implications for treatment of muscle wasting disorders, *Exp. Cell Res.*, 280(1), 55–63, 2002.
215. Quinn, L.S., Haugk, K.L., and Grabstein, K.H., Interleukin-15: a novel anabolic cytokine for skeletal muscle, *Endocrinology*, 136(8), 3669–3672, 1995.
216. Rabinovsky, E.D., Gelir, E., Gelir, S., Lui, H., Kattash, M., DeMayo, F.J. et al., Targeted expression of IGF-1 transgene to skeletal muscle accelerates muscle and motor neuron regeneration, *Faseb J.*, 17(1), 53–55, 2003.
217. Rafael, J.A. and Brown, S.C., Dystrophin and utrophin: genetic analyses of their role in skeletal muscle, *Microsc. Res. Tech.*, 48(3–4), 155–166, 2000.
218. Rafael, J.A., Townsend, E.R., Squire, S.E., Potter, A.C., Chamberlain, J.S., and Davies, K.E., Dystrophin and utrophin influence fiber type composition and post-synaptic membrane structure, *Hum. Mol. Genet.*, 9(9), 1357–1367, 2000.
219. Reddy, A.S., Reedy, M.K., Best, T.M., Seaber, A.V., Garrett, W.E., Jr. Restriction of the injury response following an acute muscle strain, *Med. Sci. Sports Exerc.*, 25(3), 321–327, 1993.
220. Rinard, J., Clarkson, P.M., Smith, L.L., and Grossman, M., Response of males and females to high-force eccentric exercise, *J. Sports Sci.*, 18(4), 229–236, 2000.
221. Rodenburg, J.B., Bar, P.R., and De Boer, R.W., Relations between muscle soreness and biochemical and functional outcomes of eccentric exercise, *J. Appl. Physiol.*, 74(6), 2976–2983, 1993.
222. Rodenburg, J.B., Steenbeek, D., Schiereck, P., and Bar, P.R., Warm-up, stretching and massage diminish harmful effects of eccentric exercise, *Int. J. Sports Med.*, 15(7), 414–419, 1994.
223. Rogers, M.A., Stull, G.A., and Apple, F.S., Creatine kinase isoenzyme activities in men and women following a marathon race, *Medicine & Science in Sports & Exercise*, 17(6), 679–682, 1985.
224. Round, J.M., Jones, D.A., and Cambridge, G., Cellular infiltrates in human skeletal muscle: exercise induced damage as a model for inflammatory muscle disease? *J. Neurol. Sci.*, 82(1–3), 1–11, 1987.
225. Sadeh, M., Czyewski, K., and Stern, L.Z., Chronic myopathy induced by repeated bupivacaine injections, *J. Neurol. Sci.*, 67(2), 229–238, 1985.
226. Sam, M., Shah, S., Friden, J., Milner, D.J., Capetanaki, Y., and Lieber, R.L., Desmin knockout muscles generate lower stress and are less vulnerable to injury compared with wild-type muscles, *Am. J. Physiol. Cell Physiol.*, 279(4), C1116–C1122, 2000.
227. Sargeant, A.J. and Dolan, P., Human muscle function following prolonged eccentric exercise, *Eur. J. Appl. Physiol. Occup. Physiol.*, 56(6), 704–711, 1987.
228. Sato, K. and Kawashima, S., Calpain function in the modulation of signal transduction molecules, *Biol. Chem.*, 382(5), 743–751, 2001.
229. Sato, S., Yanagisawa, K., and Miyatake, T., Conversion of myelin-associated glycoprotein (MAG) to a smaller derivative by calcium activated neutral protease (CANP)-like enzyme in myelin and inhibition by E-64 analogue, *Neurochem. Res.*, 9(5), 629–635, 1984.
230. Savage, K.J. and Clarkson, P.M., Oral contraceptive use and exercise-induced muscle damage and recovery, *Contraception*, 66(1), 67–71, 2002.
231. Saxton, J.M., Clarkson, P.M., James, R., Miles, M., Westerfer, M., Clark, S., et al. Neuromuscular dysfunction following eccentric exercise, *Med. Sci. Sports Exerc.*, 27(8), 1185–1193, 1995.
232. Sayeed, M.M., Alterations in calcium signaling and cellular responses in septic injury, *New Horiz.*, 4(1), 72–86, 1996.

233. Sayers, S.P. and Clarkson, P.M., Force recovery after eccentric exercise in males and females. *Eur. J. Appl. Physiol.*, 84(1–2), 122–126, 2001.
234. Schwane, J.A. and Armstrong, R.B., Effect of training on skeletal muscle injury from downhill running in rats, *J. Appl. Physiol.*, 55(3), 969–975, 1983.
235. Scifres, J.C. and Martin, D.T., Velocity of Eccentric Contraction affects the Magnitude of Delayed Onset Muscle Soreness. *Medicine & Science in Sports & Exercise*, 24(5), S851, 1992.
236. Sheehan, S.M. and Allen, R.E., Skeletal muscle satellite cell proliferation in response to members of the fibroblast growth factor family and hepatocyte growth factor, *J. Cell Physiol.*, 181(3), 499–506, 1999.
237. Sherwood, L., Human Physiology: From cells to systems. 4th ed., Brooks–Cole, Pacific Grove, CA, 2001.
238. Singh, M. and Karpovich, P.V., Isotonic and isometric forces of forearm flexors and extensors, *J. Appl. Physiol.*, 21(4), 1435–1437, 1966.
239. Sjogaard, G., Muscle energy metabolism and electrolyte shifts during low-level prolonged static contraction in man, *Acta Physiol. Scand.*, 134(2), 181–187, 1988.
240. Sjogaard, G. and Jensen, B.R., Muscle Pathology with Overuse. In: Ranney, D., editor. Chronic Musculoskeletal Injuries in the Workplace. Philadelphia, PA: W.B. Saunders Company; 1997. p. 17–40.
241. Sjogaard, G., Lundberg, U., and Kadefors, R., The role of muscle activity and mental load in the development of pain and degenerative processes at the muscle cell level during computer work, *Eur. J. Appl. Physiol.*, 83(2–3), 99–105, 2000.
242. Sjogaard, G. and McComas, A.J., Role of interstitial potassium, *Adv. Exp. Med. Biol.*, 384, 69–80, 1995.
243. Sjogaard, G. and Sogaard, K., Muscle injury in repetitive motion disorders, *Clin. Orthop.*, (351), 21–31, 1998.
244. Smith, H.J. and Tisdale, M.J., Signal transduction pathways involved in proteolysis-inducing factor induced proteasome expression in murine myotubes, *Br. J. Cancer.*, 89(9), 1783–1788, 2003.
245. Smith, L.L., Fulmer, M.G., Holbert, D., McCammon, M.R., Houmard, J.A., Frazer, D.D. et al., The impact of a repeated bout of eccentric exercise on muscular strength, muscle soreness and creatine kinase, *Br. J. Sports Med.*, 28(4), 267–271, 1994.
246. Sola, O.M., Christensen, D.L., and Martin, A.W., Hypertrophy and hyperplasia of adult chicken anterior latissimus dorsi muscles following stretch with and without denervation, *Exp. Neurol.*, 41(1), 76–100, 1973.
247. Sorichter, S., Mair, J., Koller, A., Calzolari, C., Huonker, M., Pau, B. et al., Release of muscle proteins after downhill running in male and female subjects, *Scand. J. Med. Sci. Sports*, 11(1), 28–32, 2001.
248. Sorichter, S., Puschendorf, B., and Mair, J., Skeletal muscle injury induced by eccentric muscle action: muscle proteins as markers of muscle fiber injury, *Exercise Immunology Review*, 5, 5–21, 1999.
249. Spencer, M.J., Montecino-Rodriguez, E., Dorshkind, K., and Tidball, J.G., Helper (CD4(+)) and cytotoxic (CD8(+)) T cells promote the pathology of dystrophin-deficient muscle, *Clin. Immunol.*, 98(2), 235–243, 2001.
250. Spencer, M.J. and Tidball, J.G., Do immune cells promote the pathology of dystrophin-deficient myopathies? *Neuromuscul. Disord.*, 11(6–7), 556–564, 2001.
251. St. Pierre, B.A. and Tidball, J.G., Macrophage activation and muscle remodeling at myotendinous junctions after modifications in muscle loading, *Am. J. Pathol.*, 145(6), 1463–1471, 1994.
252. St. Pierre Schneider, B., Correia, L.A., and Cannon, J.G., Sex differences in leukocyte invasion in injured murine skeletal muscle, *Res. Nurs. Health*, 22(3), 243–250, 1999.
253. Stauber, W.T., Eccentric action of muscles: physiology, injury, and adaptation, *Exerc. Sport Sci. Rev.*, 17, 157–185, 1989.
254. Stauber, W.T., Measurement of Muscle Function in Man. In: Sports Injuries, International Perspectives in Physical Therapy: Churchhill-Livingstone; 1989.

255. Stauber, W.T., Fritz, V.K., Vogelbach, D.W., and Dahlmann, B., Characterization of muscles injured by forced lengthening I. Cellular infiltrates, *Med. Sci. Sports Exerc.*, 20(4), 345–353, 1988.
256. Stauber, W.T., Knack, K.K., Miller, G.R., and Grimmett, J.G., Fibrosis and intercellular collagen connections from four weeks of muscle strains, *Muscle Nerve*, 19(4), 423–430, 1996.
257. Stauber, W.T., and Smith, C.A., Cellular responses in exertion-induced skeletal muscle injury, *Mol. Cell Biochem.*, 179(1–2), 189–196, 1998.
258. Stevens, E.D., Relation between work and power calculated from force-velocity curves to that done during oscillatory work, *J. Muscle. Res. Cell. Motil.*, 14(5), 518–526, 1993.
259. Stevens, E.D., Effect of phase of stimulation on acute damage caused by eccentric contractions in mouse soleus muscle, *J. Appl. Physiol.*, 80(6), 1958–1962, 1996.
260. Stevens, E.D., and Syme, D.A., Effect of stimulus duty cycle and cycle frequency on power output during fatigue in rat diaphragm muscle doing oscillatory work, *Can. J. Physiol. Pharmacol.*, 71(12), 910–916, 1993.
261. Stone, M.H. and Lipner, H., Responses to intensive training and methandrostenelone administration. I. Contractile and performance variables. *Pflugers Arch.*, 375(2), 141–146, 1978.
262. Stone, M.H., Rush, M.E., and Lipner, H., Responses to intensive training and methandrostenelone administration: II. Hormonal, organ weights, muscle weights and body composition, *Pflugers Arch.*, 375(2), 147–151, 1978.
263. Stupka, N., Lowther, S., Chorneyko, K., Bourgeois, J.M., Hogben, C., and Tarnopolsky, M.A., Gender differences in muscle inflammation after eccentric exercise, *J. Appl. Physiol.*, 89(6), 2325–2332, 2000.
264. Sun, J.S., Tsuang, Y.H., Hang, Y.S., Liu, T.K., Lee, W.W., and Cheng, C.K., The deleterious effect of tetanic contraction on rabbit's triceps surae muscle during cyclic loading, *Clin. Biomech. (Bristol, Avon)*, 11(1), 46–50, 1996.
265. Talbot, J.A. and Morgan, D.L., Quantitative analysis of sarcomere non-uniformities in active muscle following a stretch, *J. Muscle Res. Cell Motil.*, 17(2), 261–268, 1996.
266. Tamaki, T., Uchiyama, S., and Nakano, S., A weight-lifting exercise model for inducing hypertrophy in the hindlimb muscles of rats, *Med. Sci. Sports Exerc.*, 24(8), 881–886, 1992.
267. Tate, C.A., Bonner, H.W., and Leslie, S.W., Calcium uptake in skeletal muscle mitochondria. I. The effects of chelating agents on the mitochondria from fatigued rats, *Eur. J. Appl. Physiol. Occup. Physiol.*, 39(2), 111–116, 1978.
268. Thompson, C.B., Choi, C., Youn, J.H., and McDonough, A.A., Temporal responses of oxidative vs. glycolytic skeletal muscles to K+ deprivation: Na+ pumps and cell cations, *Am. J. Physiol.*, 276(6 Pt 1), C1411–C1419, 1999.
269. Tidball, J.G., Inflammatory cell response to acute muscle injury, *Med. Sci. Sports Exerc.*, 27(7), 1022–1032, 1995.
270. Tiidus, P.M., Can estrogens diminish exercise induced muscle damage? *Can. J. Appl. Physiol.*, 20(1), 26–38, 1995.
271. Tiidus, P.M. and Bombardier, E., Oestrogen attenuates post-exercise myeloperoxidase activity in skeletal muscle of male rats, *Acta. Physiol. Scand.*, 166(2), 85–90, 1999.
272. Tiku, K., Tiku, M.L., Liu, S., and Skosey, J.L., Normal human neutrophils are a source of a specific interleukin 1 inhibitor, *J. Immunol.*, 136(10), 3686–3692, 1986.
273. Trappe, T.A., Fluckey, J.D., White, F., Lambert, C.P., and Evans, W.J., Skeletal muscle PGF(2)(alpha) and PGE(2) in response to eccentric resistance exercise: influence of ibuprofen acetaminophen, *J. Clin. Endocrinol. Metab.*, 86(10), 5067–5070, 2001.
274. Trappe, T.A., White, F., Lambert, C.P., Cesar, D., Hellerstein, M., and Evans, W.J., Effect of ibuprofen and acetaminophen on postexercise muscle protein synthesis, *Am. J. Physiol. Endocrinol. Metab.*, 282(3), E551–E556, 2002.
275. Triolo, R., Robinson, D., Gardner, E., and Betz, R., The eccentric strength of electrically stimulated paralyzed muscle, *IEEE Transactions on Biomedical Engineering*, 651–652, 1987.

276. van der Meulen, J.H., Exercise Induced Muscle Damage: Morphological, Biochemical, and Functional Aspects [Doctoral Dissertation]. Amsterdam: Universiteit van Amsterdam; 1991.
277. van der Meulen, J.H., Kuipers, H., and Drukker, J., Relationship between exercise-induced muscle damage and enzyme release in rats, *J. Appl. Physiol.*, 71(3), 999–1004, 1991.
278. van der Meulen, J.H., McArdle, A., Jackson, M.J., and Faulkner, J.A., Contraction-induced injury to the extensor digitorum longus muscles of rats: the role of vitamin E, *J. Appl. Physiol.*, 83(3), 817–823, 1997.
279. van der Vusse, G.J., de Groot, M.J., Willemsen, P.H., van Bilsen, M., Schrijvers, A.H., and Reneman, R.S., Degradation of phospholipids and triacylglycerol, and accumulation of fatty acids in anoxic myocardial tissue, disrupted by freeze-thawing, *Mol. Cell Biochem.*, 88(1–2), 83–90, 1989.
280. van der Vusse, G.J., van Bilsen, M., and Reneman, R.S., Is Phospholipid Degradation a Critical Event in Ischemia and Reperfusion-Induced Damage? *News in Physiological Sciences*, 49–53, 1989.
281. Vijayan, K., Thompson, J.L., Norenberg, K.M., Fitts, R.H., and Riley, D.A., Fiber-type susceptibility to eccentric contraction-induced damage of hindlimb-unloaded rat AL muscles, *J. Appl. Physiol.*, 90(3), 770–776, 2001.
282. Vollestad, N.K. and Sejersted, O.M., Biochemical correlates of fatigue. A brief review, *Eur. J. Appl. Physiol. Occup. Physiol.*, 57(3), 336–347, 1988.
283. Warren, G.L., 3rd, Williams, J.H., Ward, C.W., Matoba, H., Ingalls, C.P., Hermann, K.M., et al. Decreased contraction economy in mouse EDL muscle injured by eccentric contractions, *J. Appl. Physiol.*, 81(6), 2555–2564, 1996.
284. Warren, G.L., Hayes, D.A., Lowe, D.A., and Armstrong, R.B., Mechanical factors in the initiation of eccentric contraction-induced injury in rat soleus muscle, *J. Physiol.*, 464, 457–475, 1993.
285. Warren, G.L., Hayes, D.A., Lowe, D.A., Prior, B.M., and Armstrong, R.B., Materials fatigue initiates eccentric contraction-induced injury in rat soleus muscle, *J. Physiol.*, 464, 477–489, 1993.
286. Warren, G.L., Hayes, D.A., Lowe, D.A., Williams, J.H., and Armstrong, R.B., Eccentric contraction-induced injury in normal and hindlimb-suspended mouse soleus and EDL muscles, *J. Appl. Physiol.*, 77(3), 1421–1430, 1994.
287. Warren, G.L., Ingalls, C.P., and Armstrong, R.B., Temperature dependency of force loss and Ca(2+) homeostasis in mouse EDL muscle after eccentric contractions, *Am. J. Physiol. Regul. Integr. Comp. Physiol.*, 282(4), R1122–R1132, 2002.
288. Warren, G.L., Ingalls, C.P., Shah, S.J., and Armstrong, R.B., Uncoupling of in vivo torque production from EMG in mouse muscles injured by eccentric contractions, *J. Physiol.*, 515 (Pt 2), 609–619, 1999.
289. Warren, G.L., Lowe, D.A., and Armstrong, R.B., Measurement tools used in the study of eccentric contraction-induced injury, *Sports Medicine*, 27(1), 43–59, 1999.
290. Warren, G.L., Lowe, D.A., Hayes, D.A., Farmer, M.A., and Armstrong, R.B., Redistribution of cell membrane probes following contraction-induced injury of mouse soleus muscle, *Cell & Tissue Research*, 282(2), 311–320, 1995.
291. Warren, G.L., Lowe, D.A., Hayes, D.A., Karwoski, C.J., Prior, B.M., and Armstrong, R.B., Excitation failure in eccentric contraction-induced injury of mouse soleus muscle, *J. Physiol.*, 468, 487–499, 1993.
292. Whitehead, N.P., Allen, T.J., Morgan, D.L., and Proske, U., Damage to human muscle from eccentric exercise after training with concentric exercise, *J. Physiol.*, 512 (Pt 2), 615–620, 1998.
293. Wickiewicz, T.L., Roy, R.R., Powell, P.L., and Edgerton, V.R., Muscle architecture of the human lower limb, *Clin. Orthop.*, (179), 275–583, 1983.
294. Widrick, J.J., Costill, D.L., McConell, G.K., Anderson, D.E., Pearson, D.R., and Zachwieja, J.J., Time course of glycogen accumulation after eccentric exercise, *J. Appl. Physiol.*, 72(5), 1999–2004, 1992.
295. Wilkie, D.R., The relation between force and velocity in human muscle, *J. Physiol.*, 110, 249–280, 1950.
296. Willems, M.E. and Stauber, W.T., Isometric and concentric performance of electrically stimulated ankle plantar flexor muscles in intact rat, *Exp. Physiol.*, 84(2), 379–389, 1999.

297. Willems, M.E. and Stauber, W.T., Changes in force by repeated stretches of skeletal muscle in young and old female Sprague Dawley rats, *Aging (Milano)*, 12(6), 478–481, 2000.
298. Willems, M.E. and Stauber, W.T., Force deficits after repeated stretches of activated skeletal muscles in female and male rats, *Acta Physiol. Scand.*, 172(1), 63–67, 2001.
299. Willems, M.E. and Stauber, W.T., Fatigue and recovery at long and short muscle lengths after eccentric training, *Med. Sci. Sports Exerc.*, 34(11), 1738–43, 2002.
300. Williams, P., Kyberd, P., Simpson, H., Kenwright, J., and Goldspink, G., The morphological basis of increased stiffness of rabbit tibialis anterior muscles during surgical limb-lengthening, *J. Anat.*, 193 (Pt 1), 131–138, 1998.
301. Wirth, O., Gregory, E.W., Cutlip, R.G., and Miller, G.R., Control and quantitation of voluntary weight-lifting performance of rats, *J. Appl. Physiol.*, 95(1), 402–412, 2003.
302. Wong, T.S. and Booth, F.W., Skeletal muscle enlargement with weight-lifting exercise by rats, *J. Appl. Physiol.*, 65(2), 950–954, 1988.
303. Wong, T.S. and Booth, F.W., Protein metabolism in rat gastrocnemius muscle after stimulated chronic concentric exercise, *J. Appl. Physiol.*, 69(5), 1709–1717, 1990.
304. Wong, T.S. and Booth, F.W., Protein metabolism in rat tibialis anterior muscle after stimulated chronic eccentric exercise, *J. Appl. Physiol.*, 69(5), 1718–1724, 1990.
305. Wood, S.A., Morgan, D.L., and Proske, U., Effects of repeated eccentric contractions on structure and mechanical properties of toad sartorius muscle, *Am. J. Physiol.*, 265(3 Pt 1), C792–C800, 1993.
306. Wright, J., Huang, Q.Q., and Wang, K., Nebulin is a full-length template of actin filaments in the skeletal muscle sarcomere: an immunoelectron microscopic study of its orientation and span with site-specific monoclonal antibodies, *J. Muscle. Res. Cell Motil.*, 14(5), 476–483, 1993.
307. Wung, B.S., Cheng, J.J., Hsieh, H.J., Shyy, Y.J., and Wang, D.L., Cyclic strain-induced monocyte chemotactic protein-1 gene expression in endothelial cells involves reactive oxygen species activation of activator protein 1, *Circ. Res.*, 81(1), 1–7, 1997.
308. Yarasheski, K.E., Lemon, P.W., and Gilloteaux, J., Effect of heavy-resistance exercise training on muscle fiber composition in young rats, *J. Appl. Physiol.*, 69(2), 434–437, 1990.
309. Yeung, E.W., Bourreau, J.P., Allen, D.G., and Ballard, H.J., Effect of eccentric contraction-induced injury on force and intracellular pH in rat skeletal muscles, *J. Appl. Physiol.*, 92(1), 93–99, 2002.
310. Zador, E., Mendler, L., Takacs, V., de Bleecker, J., and Wuytack, F., Regenerating soleus and extensor digitorum longus muscles of the rat show elevated levels of TNF-alpha and its receptors, TNFR-60 and TNFR-80, *Muscle & Nerve*, 24(8), 1058–1067, 2001.
311. Zajac, F.E., Muscle and tendon: properties, models, scaling, and application to biomechanics and motor control, *Crit. Rev. Biomed. Eng.*, 17(4), 359–411, 1989.
312. Zeng, C., Pesall, J.E., Gilkerson, K.K., and McFarland, D.C., The effect of hepatocyte growth factor on turkey satellite cell proliferation and differentiation, *Poult. Sci.*, 81(8), 1191–1198, 2002.
313. Zerba, E. and Faulkner, J.A., A Single Lengthening Contraction can Induce Injury to Skeletal Muscle Fibers, *Physiologist*, 33, A122, 1990.
314. Zerba, E., Komorowski, T.E., and Faulkner, J.A., Free radical injury to skeletal muscles of young, adult, and old mice, *Am. J. Physiol.*, 258(3 Pt 1), C429–C435, 1990.