CHANGES IN SKELETAL MUSCLE ULTRASTRUCTURE AND STRENGTH PERFORMANCE FOLLOWING ACUTE RESISTANCE EXERCISE

By

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To my parents, who will be proud, and my dog Schooner, who keeps the world in perspective for me.

ABSTRACT

The purpose of this study was to examine changes in muscle ultrastructure and strength performance following a single bout of elbow flexor resistance exercise. Eight untrained males performed 8 sets of 8 repetitions at 80% concentric 1 RM. One arm performed only the concentric (CON) phase of the movement while the other performed only the eccentric (ECC) phase. Maximum isometric (MVC), low (LV) and high velocity (HV) concentric peak torque, and evoked contractile property measurements of the elbow flexors were made before and after the bout, and at 24, 48, 72 and 96 h. Needle biopsies were obtained from the biceps brachii prior to the exercise, immediately post-exercise from each arm (POST-CON, POST-ECC), and 48 h post-exercise from each arm (48H-CON, 48H-ECC). Electron microscopy was used to quantify the extent of fiber disruption in each sample. The severity of disruption was classified as focal (FOC), moderate (MOD), or extreme (EXT). All strength measurements decreased ($P \leq 0.05$) below pre-exercise values immediately post-ex in both arms, but dramatic differences were observed between arms during the subsequent recovery period. MVC, LV, HV and peak twitch torque (PTT) recovered to pre-ex values by 24 h in the CON arm. In the ECC arm, HV did not recover for at least 72 h, and MVC, LV and PTT remained depressed at 96 h. ANOVA revealed a greater ($P \leq 0.05$) number fibers were disrupted in

the POST-CON, POST-ECC, 48H-CON and 48H-ECC samples compared to BASE. Significantly more fibers appeared disrupted in the POST-ECC (82%) and 48H-ECC (80%) samples compared to the POST-CON (33%) and 48H-CON (37%) samples, respectively. In addition, the POST-ECC (41%) and 48H-ECC (50%) samples contained a greater number of fibers with EXT disruption compared to the POST-CON (13%) and 48H-CON (17%) samples. Decreases in MVC at 48 h correlated ($P \leq 0.05$) with the extent of EXT disruption in the 48H-CON and 48H-ECC samples. These data indicate that both the CON and ECC phase of weightlifting produce myofibrillar disruption, with the greatest disruption occurring during the ECC phase.

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PREFACE

The following thesis is presented in two chapters. Chapter I is a literature review related to the mechanisms of exercise-induced muscle fiber injury, repair, and adaptation in humans. Chapter II embodies the thesis research and is presented in a manuscript style suitable for publication.

ACKNOWLEDGEMENTS

This thesis should really be viewed as a group effort, since there are a number of individuals who contributed both directly and indirectly to the final product. In keeping with the spirit of the game I love, the following analogy can be viewed as a sincere "thank-you" to the people who assisted in the completion of this project.

First and foremost, I must acknowledge team manager J. Duncan MacDougall (my advisor and mentor), for giving me a spot on his roster. He patiently showed me the fundamentals of the game, and instilled confidence that one day I'll be in the Big Leagues if I just work hard enough. Base coaches Joe Blimkie, Neil McCartney, Digby Sale, and Bill Stauber (my examining committee) each helped with certain aspects of my game, and assisted in my transition from Class AA to AAA ball. Scouts Digby Elliott, Tim Lee, Jim Lyons, and Brian Maraj (the motor learning crew) helped set fielding positions by providing up-to-the-minute statistical reports. I'm particularly indebted to this group for always having the coffee pot on when I arrived at the ballpark, and never declining an invitation for a malted beverage after a game!

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TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

CHAPTER I

EXERCISE-INDUCED MUSCLE FIBER INJURY

1.1 INTRODUCTION

Strenuous, unaccustomed exercise results in damage to skeletal muscle (Armstrong et al., 1991; Ebbeling and Clarkson, 1989). Evidence of tissue injury includes morphological disruption of fibers (Fridén et al., 1981, 1983b; Newham et al., 1983a) the appearance of muscle enzymes in the blood (Clarkson et al., 1986; Schwane et al., 1983), changes in force production (Davies and White, 1981; Newham et al., 1983b), and delayed-onset muscle soreness (Asmussen, 1956; Schwane et al., 1983). The damage is not permanent, although it may take up to 10-14 days before the tissues are repaired (Jones et al., 1986; Manfredi et al., 1991; O'Reilly et al., 1987). This process may be a normal precursor to muscle adaptation (Armstrong, 1984, 1990), since the muscles appear more resilient to future damage following regeneration (Byrnes et al., 1985, Clarkson et al., 1988). Despite its prevalence, the causative factors and sequence of events in exercise-induced muscle damage are not well understood.

Although many types of exercise have been shown to produce tissue damage (Ebbeling and Clarkson, 1989), activities which contain large negative work components (i.e.,

eccentric muscle action) have consistently been purported as the most damaging (Fridén and Lieber, 1992). An eccentric muscle action (EMA) occurs when the muscle actively lengthens while producing force (Knuttgen and Komi, 1992). This review will focus on the energetics of EMA, and will summarize the unique physiological properties which have been implicated in exercise-induced muscle fiber injury. It will outline the proposed mechanisms for damage and the potential sites of insult, and will present a hypothetical model of the damage and repair process. Finally, the review will describe the changes in force production following eccentrically-biased exercise, and examine muscle fiber adaptation following EMA.

[An aside on terminology: the term "eccentric exercise" is liberally used by many investigators to describe a variety of exercise models which contain large negative work components (e.g., downhill running, eccentric cycle ergometry). The term "eccentrically-biased exercise" is probably more appropriate, since human motion seldom involves pure forms of isolated concentric, eccentric or isometric actions (Knuttgen and Komi, 1992). In addition, most of the exercise protocols utilized in these investigations are dynamic in nature. Very few studies have examined resistive-type models of exercise, which contain clearly defined concentric and eccentric phases. Whenever possible, investigations of this type will be highlighted.]

1.2 THE PHYSIOLOGY OF ECCENTRIC MUSCLE ACTION

Active research into the physiology of eccentric muscle actions in human subjects began in the early 1950's. Based on the pioneering work of several laboratories, Asmussen (1956) established that eccentric muscle action induced substantially greater muscle soreness than either concentric or isometric action. He attributed the pronounced soreness to "strain or damage to part of the muscle." Several decades of study have confirmed that the physiological properties of eccentric muscle action are inherently unique, and intimately related to the phenomenon of exercise-induced muscle fiber injury.

1.2.1 Energetics

Abbott et al. (1952) and Asmussen (1953), demonstrated that the physiological cost of negative work (eccentric muscle action) was substantially less than that of positive work, when performing cycling tasks at similar workloads. Further, both investigators showed that the relative difference in oxygen uptake between concentric and eccentric cycle ergometry increased with exercise intensity. One explanation for the discrepancy in metabolic cost was that fewer muscle fibers were required to maintain the same force output during eccentric exercise (Abbott et al., 1952). This assertion was supported by studies of isolated muscle preparations, where the energy requirement of a fiber (measured from its rates of

ATP and phosphocreatine breakdown, and metabolic heat production) was much lower when the muscle was stretched compared to when it shortened (Wilkie, 1968).

The integrated electromyographic (IEMG) activity of muscles performing concentric and eccentric actions, at the same force production, is also quite different (Bigland-Ritchie and Woods, 1976; Komi and Buskirk, 1972). [IEMG activity reflects the number of action potentials in a muscle, which indicates the recruitment of the corresponding muscle fibers and their firing frequency (Basmajian, 1978)]. Bigland-Ritchie and Woods (1976) examined the IEMG activity of m. vastus lateralis, and oxygen uptake, during positive and negative work on a motorized cycle ergometer. They demonstrated that, under comparable work loads, there was less IEMG activity and a smaller oxygen uptake during eccentric exercise. Further, they showed that the reduction in oxygen consumption was substantially greater than the decrease in IEMG, indicating that active muscle fibers require less oxygen when they are being stretched than when they shorten. Newham et al. (1983b), using a stepping task in which one leg acted concentrically while the contralateral leg acted eccentrically, confirmed that fewer motor units are recruited to produce a similar amount of negative work.

1.2.2 Relation to Muscle Injury

Two aspects of reduced energy cost can therefore be distinguished when muscles perform eccentric muscle actions: (a) decreased energy utilization of active fibers, and (b) reduced motor unit recruitment and/or activation (Stauber, 1989). The functional significance of these properties is that, under comparable workloads, eccentric muscle actions produce greater tension per cross-sectional area of active fiber than concentric actions. Although many types of unaccustomed exercise can produce muscle damage (Clarkson et al., 1986), eccentric exercise has consistently been established as the most damaging in both humans (Fridén et al., 1983b; Newham et al., 1983a) and animals (Armstrong et al., 1983b; Ogilvie et al., 1988). The remainder of this review will deal specifically with muscle fiber injury in humans, although investigations using animal models will also be discussed briefly.

1.3 INITIAL EVENT IN MUSCLE FIBER INJURY

The unique physiological properties of eccentric muscle action have been implicated as causative factors for muscle fiber injury. (For review, see Armstrong, 1990; Armstrong et al., 1991; Ebbeling and Clarkson, 1989; Evans and Cannon, 1991). Although a number of hypotheses have been presented, the specific initiating event underlying exercise-induced

muscle damage remains unknown. The potential mechanisms can essentially be categorized as either metabolic or mechanical in nature. The metabolic events proposed include high temperature, insufficient mitochondrial respiration, oxygen free radical production, and lowered pH level (Armstrong, 1990). The mechanical hypothesis is based on the common finding that eccentric muscle actions, while metabolically less demanding, result in greater myofiber injury compared to concentric or isometric actions.

1.3.1 Metabolic Hypotheses

A) High Temperature

Eccentric muscle actions may generate higher local temperatures than concentric actions at the same workload (Nadel et al., 1972). Elevated temperatures in the muscles could result in damage to the structural proteins of the fibers, particularly the contractile apparatus (Armstrong, 1984). There is evidence from myopathic conditions, such as malignant hyperthermia, that high muscle temperatures can result in severe degeneration of muscle fibers (Ebbeling and Clarkson, 1989). However, Armstrong et al. (1991) warn that the comparison of different muscle actions at equivalent absolute metabolic rates (as many studies have done) may not be valid, noting that the Fenn effect actually predicts a lower rate of heat production during eccentric muscle actions. They suggest that if a higher muscle temperature occurs during

eccentric exercise, it is not due to a higher rate of heat production within the muscle, but rather the result of a lower rate of heat removal.

B) Insufficient Mitochondrial Respiration

During high intensity exercise, the rate of ATP hydrolysis may exceed ATP synthesis, due to insufficient mitochondrial respiration. Attenuated ATP levels could compromise calcium (Ca^{2+}) removal from the cytoplasm, allowing an elevation in cytoplasmic Ca^{2+} levels (Armstrong, 1991). Studies with chemically skinned fibers have demonstrated that incubation of the muscle in Ca^{2+} concentrations of 0.5 to 8 $\mu mol \cdot L^{-1}$ (within the physiological range of contracting muscles) stimulates destruction of myofibrils and hypercontraction of sarcomeres (Duncan, 1987). Nonetheless, most researchers dispute this hypothesis, citing evidence that eccentric muscle actions, while less metabolically demanding, result in greater fiber damage. Armstrong et al. (1991) however, caution that focal lesions within the fiber may have a metabolic origin (i.e., localized areas of ATP depletion), and these areas would not be detectable from whole muscle assays. In this regard, Lieber and Fridén (1988) have suggested that eccentric exercise-induced fiber injury in rabbit tibialis anterior muscle may be metabolic in nature.

C) Oxygen Free Radical Production

The metabolic stress induced by high intensity exercise may enhance the rate of oxygen free radical generation (Jenkins, 1988). Increased production of these extremely reactive molecules could exceed the protective capabilities of the scavenger enzyme and antioxidant systems, resulting in free radical attack on skeletal muscle tissue. Lipid peroxidation, a consequence of free radical generation in which hydroxyl radicals combine with lipids in the cell membrane, is especially disruptive to cell integrity (Sjödin et al., 1990). Maughan et al. (1989) investigated lipid peroxidation in humans following a 45 min. bout of downhill running, and suggested a relationship between free radical generation and exercise-induced muscle fiber injury. However, the metabolic changes that accompany endurance exercise (e.g., body temperature, substrate availability, hormone status) may be responsible for the increase in free radical production observed (Ebbeling and Clarkson, 1989). An increase in lipid peroxidation following short term, intense eccentric exercise has not been documented.

D) Lowered pH Level

The lowered pH hypothesis is based on the fact that muscle fiber injury following endurance exercise resembles ischaemia-induced cell damage (Ebbelling and Clarkson, 1989). Indeed, the accumulation of lactic acid remains the most

popular explanation for delayed muscle soreness in the lay exercise community (Armstrong, 1984). Experimental evidence, however, does not support the hypothesis that muscle damage following exercise is due to elevated [H^+]. Schwane et al. (1983) investigated the relation between blood lactate levels and delayed onset muscle soreness (DOMS) following level and downhill running in humans. They showed that downhill running (which primarily involves eccentric muscle actions) produced less lactate than level running (which consists of similar concentric and eccentric components), but resulted in significantly greater DOMS. Since eccentric exercise has been established as the most damaging type in humans (Newham et al., 1983a), it seems reasonable to assume that lowered muscle pH is not a primary agent in exercise-induced muscle damage. Animal investigations have confirmed that eccentric muscle actions produce substantially less lactate than concentric actions at the same power output (Armstrong et al., 1983a), yet result in greater fiber damage (Armstrong et al., 1983b).

1.3.2 Mechanical Hypothesis

The mechanical hypothesis is based on the common finding that eccentric muscle actions require less energy, yet cause greater fiber injury, than comparable concentric or isometric actions. This theory is consistent with the observation that high specific tensions (i.e., force per cross sectional area)

appear to be generated in active muscle fibers during eccentric muscle actions (Armstrong, 1990). During active muscle lengthening, the mechanical forces of stretch could potentially damage the sarcolemma, sarcoplasmic reticulum (SR), contractile apparatus, basal lamina, or surrounding connective tissue (Stauber, 1989).

Several variations of the mechanical hypothesis have been proposed. A number of investigators have suggested that the loss of Ca^{2+} homeostasis may play a primary role in muscle injury (Armstrong, 1990; Armstrong et al, 1991). It is known that sarcomere length varies within a fiber during eccentric muscle action (Colomo et al., 1988). Severe lengthening of some sarcomeres could disrupt normal Ca^{2+} cycling by damaging the sarcolemma or SR system (Armstrong et al., 1991). The inability of the cell to properly sequester Ca^{2+} could cause the fiber to enter a rigor-like state (Stauber, 1989). Tidball and Daniel (1986) hypothesized that, if only one fiber was involved in the rigor, "shear forces" could develop between adjacent myofibers, resulting in myofilament damage. Alternatively, increases in intracellular $[Ca^{2+}]$ may stimulate Ca^{2+}-sensitive enzymes (e.g., proteases, phospholipases) which are active in fiber degeneration (Byrd, 1992).

Armstrong et al. (1991) analyzed muscle fiber injury from a materials science standpoint, noting that skeletal muscle meets the criteria of a ductile material (i.e., a material that may elongate by 5% or more). Based on materials

fatigue theory, they hypothesized that the energy absorbed by a muscle being lengthened (i.e., performing negative work) must be dissipated either in the form of heat or plastic deformation. Deformation of a fiber, due to a single contraction or the cumulative effects of many contractions, could result in structural damage to the cell. While acknowledging the limitations inherent in this type of analysis, the authors cited experimental evidence to support such a theory (for review, see Armstrong et al., 1991).

1.4 STRUCTURAL EVIDENCE OF MUSCLE DAMAGE

Direct damage to the myofibrillar contractile apparatus has been observed in needle biopsy samples from human subjects following eccentric exercise (Fridén et al., 1981; Fridén et al, 1983b; Newham et al., 1983a). A variety of exercise models have been used to induce damage, including (i) continuous eccentric cycle ergometry (Fridén et al., 1983b; Fridén, 1984a; Manfredi et al., 1991; O'Reilly et al., 1987), (ii) bench stepping (Newham et al., 1983a), and (iii) repeated (anaerobic) bouts of downhill running (Fridén et al., 1981; Fridén et al., 1988). The most common structural component affected is the Z disc (Z band), which appears broadened, smeared, out of register, and sometimes totally disrupted (Fridén et al., 1981). Z band "streaming" is a non-specific indicator of muscle damage, and is often seen in patients with

neuromuscular disorders (Fridén, 1984b). However, it has also been observed in normal human muscle (Reske-Nielson and Harmsen, 1972). Meltzer et al. (1976) examined biopsies from the vastus lateralis or peroneus brevis of 34 healthy individuals and reported some degree of streaming in 2.6% and 6.7% of the fibers examined in the males and females, respectively. The exact cause of Z band streaming following eccentric muscle action is unknown, but may be related to disruption of the myofibrillar cytoskeleton (Fridén et al., 1884; Waterman-Storer, 1991).

1.4.1 Ultrastructure of the Z Disc and Cytoskeleton

The Z disc is a platelike protein structure to which the actin filaments anchor, and serves as an insertion site for various intermediate filaments (Sheterline, 1983). Under the electron microscope (which projects a two dimensional image), the Z disc corresponds to the dark narrow line which bisects the I-band between adjacent sarcomeres. At higher resolution, it appears as a jagged zig-zag, with thin filaments projecting from the "points" formed on either side (Pollack, 1992). In cross section, the Z disc appears as a square lattice, with actin filaments forming "dots" at the four corners of each square (Figure 1). The square lattice is distinctly different from the hexagonal lattice of thin filaments revealed in cross sections taken through the A-band. The built-in mismatch between the square and hexagonal lattice apparently provides

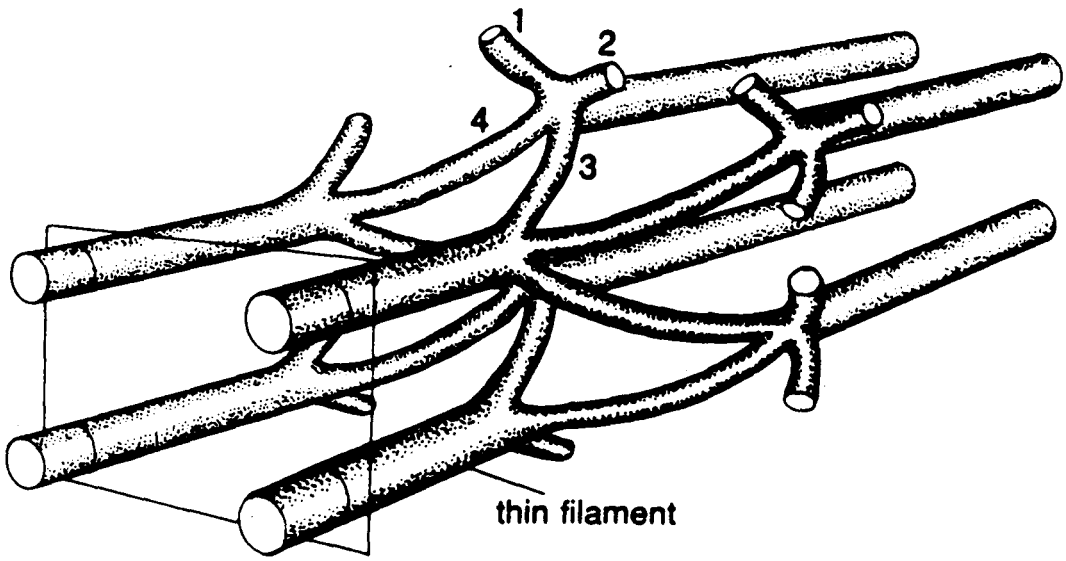


Figure 1. Schematic model of the Z disc (Pollack, 1990; after Knappeis and Carlsen, 1962). Intersecting plane illustrates the square lattice formed by 4 thin filaments.

a mechanism for increasing the myofibrillar content of the fibers during growth or as a result of exercise training (Goldspink, 1992). When force is developed, the displacement of thin filaments causes an oblique pull (mechanical stress) on the Z disc, causing it to rip and resulting in the formation of two daughter myofibrils (Goldspink, 1971).

The precise organization of the Z disc, and its chemical composition, are not known in detail. One of the proteins associated with it is α-actinin, an actin-binding protein which appears to hold the thin filaments in place and in register (Billeter and Hoppeler, 1992). In addition, there is a matrix of cytoskeletal proteins which interconnect to form a honeycomb-like sheet along the periphery of the Z disc (Waterman-Storer, 1991). The myofibrillar cytoskeleton (Figure 2) is a network of regulatory components (e.g., actin, microtubules, intermediate filaments) which provide the actual physical framework for contraction (Cooke, 1985). Although often referred to as a single entity, the cytoskeleton is actually made up of two distinct sets of filaments: the exosarcomeric cytoskeleton and endosarcomeric cytoskeleton (Wang, 1985). The exosarcomeric cytoskeleton, comprised mainly of the intermediate filament desmin, is located peripherally and links adjacent sarcomeres both transversely and longitudinally at the Z disc (Tokuyasu et al., 1983). Desmin therefore appears to maintain the lateral register of

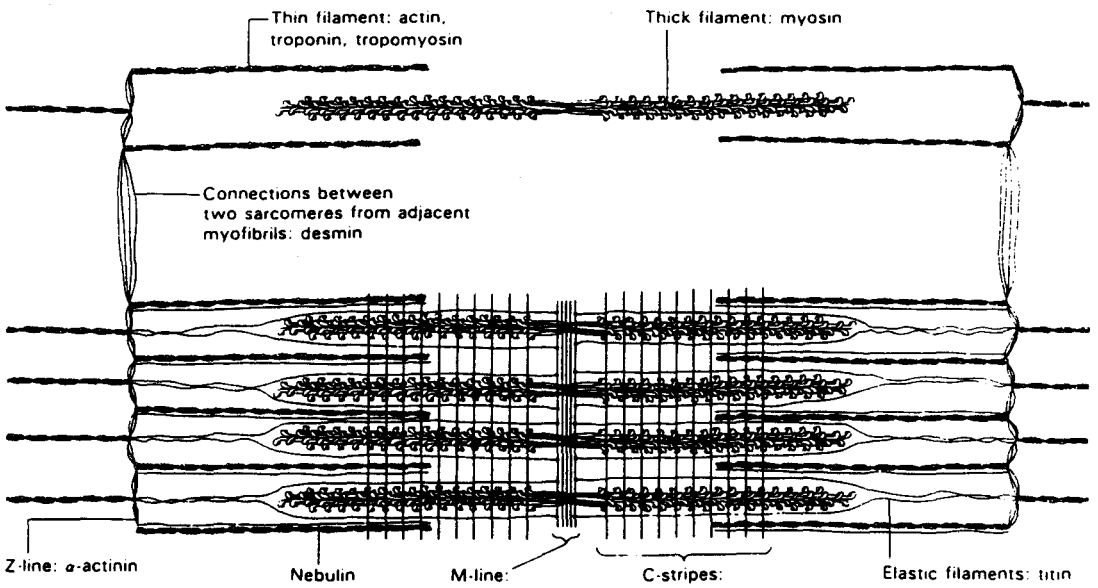


Figure 2. Schematic representation of the cytoskeletal proteins in the sarcomere (Billeter and Hoppeler, 1992).

the sarcomeres, and limits extreme length changes within individual sarcomeres (Waterman-Storer, 1991).

The endosarcomeric cytoskeleton consists of the filamentous proteins titin and nebulin, which "coexist" with actin and myosin within the sarcomere (Wang, 1985). Titin, the largest protein so far isolated, stretches from M-line to Z band and is believed to form an elastic filament (Fürst et al., 1988; Maruyama, 1985). Investigations using fluorescently labelled monoclonal antibodies raised to purified titin suggest that titin found in the I-band is elastic, while titin in the A-band is bound to the outside of the thick filaments (Horowits and Podolsky, 1987; Whiting et al., 1989). These filaments apparently provide a source of passive tension, and keep the thick filaments centered between the two Z discs during the generation of active force (Funatsu et al., 1990; Horowits et al., 1986). Nebulin appears to be rigidly bound to the Z disc and runs parallel to actin (Waterman-Storer, 1991). Although less well understood than titin, nebulin is believed to control the number of actin monomers joined to each other in a thin filament (Billeter and Hoppeler, 1992). Nebulin may also play a role in thin filament packing geometry, or help actin interdigitate with myosin when the sarcomere is stretched beyond overlap (Mayamura et al., 1989; Wang and Wright, 1988).

1.4.2 Myofibrillar Disruption Following Eccentrically-Biased Exercise

Following 30 min of cycling on an ergometer modified for eccentric work, Fridén et al. (1983b) reported "disturbances ... found to originate from the myofibrillar Z-bands" in 32% of muscle fibers examined 1 h after the bout, and 52% of fibers 3 d later. Using a point counting method, the authors determined that the relative fiber area occupied by these disturbances was 1.6% and 2.4%, respectively. The damage was random across the fiber, with some regions appearing normal and other areas showing extreme sarcomeric disruption.

Newham et al. (1983a) examined the extent of ultrastructural damage following a 20 min stepping task, in which the quadriceps of one leg acted concentrically, while the contralateral leg acted eccentrically. Needle biopsy samples were taken from the quadriceps of each leg before the exercise, immediately after, and 24-48 h later. The authors quantified the damage as (i) "focal" (a lesion which affected 1-2 adjacent sarcomeres or 1-2 adjacent myofibrils), (ii) "extensive" (a lesion which affected more than two adjacent sarcomeres and more than two adjacent myofibrils, or a fiber which contained more than ten focal areas), or (iii) "very extensive" (a fiber which contained more than one extensive area). No morphological abnormalities were observed in any of the samples taken before the exercise, or from the legs which performed positive work. Samples from the eccentrically-

exercised legs revealed disturbances in 40% of the fibers immediately after the exercise (16% focal, 16% extensive, 8% very extensive). In the samples taken an average of 30 h post-exercise, 57% of the fibers showed evidence of disruption (6% focal, 23% extensive, 28% very extensive). In addition to Z band streaming, the authors also reported the complete loss of Z bands in some sarcomeres, as well as displaced organelles and widened A bands.

The phenomenon of Z disc streaming (i.e., Z discs which appear out of register) has been attributed to disruption of the exosarcomeric cytoskeleton, and in particular the intermediate filament desmin (Fridén et al., 1984; Waterman-Storer, 1991). Fridén et al. (1984), using immunofluorescence localization of desmin in eccentrically exercised human muscle, observed longitudinal extensions of fluorescent material between successive Z discs in biopsies 3 d post-exercise. The authors attributed the change in staining pattern to (i) cytoskeletal damage induced directly by the high tension or (ii) sarcomerogenesis, a secondary response to the initial injury. Waterman-Storer (1991) proposed that both explanations may be viable: following initial disruption of the intermediate filament cytoskeletal "Z bridges", various cellular processes eventually prepare the damaged area for the insertion of new Z discs. The role of the endosarcomeric proteins (titin and nebulin) in exercise-induced muscle damage is completely unknown. Several authors have hypothesized that

disruption of titin in particular could result in the displacement of myosin filaments following eccentric exercise (Fridén and Lieber, 1992; Waterman-Storer, 1991). Further investigation of the myofibrillar cytoskeleton is clearly warranted to define the precise role it plays in the sequence of exercise-induced muscle fiber injury.

1.4.3 Fiber Type Involvement

It appears that Type II fibers are most vulnerable to injury during eccentrically-biased exercise. Fridén (1983b) reported Type II fibers were predominantly affected following a bout of eccentric cycle ergometry (by a ratio of approximately 3:1). In a subsequent study, Fridén et al. (1988) conducted a thorough ultrastructural fiber typing of damaged fibers (using a classification scheme based on the number and width of M-bridges and Z band widths). They reported that, 2 h following repetitive bouts of downhill running, 36% of the fibers examined showed evidence of damage. Of these injured fibers, 80% were classified as Type IIb. Interestingly, PAS-stained sections of the same biopsies generally indicated glycogen depletion in Type I and IIb, but not IIa fibers. O'Reilly et al. (1987) also reported substantial Type II fiber involvement with eccentric cycle ergometry (in contrast to similar concentric exercise in which Type I fibers are almost exclusively recruited). These reports suggest that eccentrically-biased exercise may utilize

a greater percentage of Type II fibers than the corresponding concentric exercise of similar workload. Since Type II fibers show the narrowest Z-bands (Eisenberg, 1983), it has been proposed that these fibers (especially Type IIb) may be more vulnerable to repetitive, high tension stress (Fridén et al., 1988; Fridén and Lieber, 1992).

An alternative explanation for the greater damage observed in Type II fibers is that these fibers may generate higher specific tensions (i.e., force per cross sectional area). Assuming Type II fibers correspond to fast motor units (Burke, 1981), evidence is available to support such a hypothesis. Bodine et al. (1987) isolated single motor units of the cat tibialis anterior muscle and classified each unit as fast, fatiguable (FF), fast, fatigue-intermediate (FI), fast, fatigue-resistant (FR), or slow, fatigue-resistant (S). Specific tension was calculated by dividing the maximum tension of a unit by its total cross sectional area. They reported the overall mean specific tension of the fast motor units (FF, FI and FR combined) was significantly greater than that of the slow units. Several investigations using human subjects have reported no difference is specific tension between fiber types (Miller et al., 1993; Schantz et al, 1983), but the methods employed in these studies are much less precise than those used by Bodine et al. (1987).

1.4.4 Resistance Training Studies

To my knowledge, no investigation has been undertaken to specifically examine myofibrillar disruption following eccentric resistance exercise. Staron and colleagues have published several reports of muscle damage following traditional, 6-10 RM isotonic resistance training, which contains both concentric and eccentric components (Staron et al., 1989, 1991, 1992). In one study, 24 women completed a 20 wks heavy resistance training program for the lower extremities (Staron et al., 1989). Light microscopy examination of needle biopsy samples revealed evidence of myofiber degeneration (e.g., fiber necrosis, phagocytosis, monocyte infiltration) and regeneration (e.g., internal nuclei, small normal fibers) in 29% of the subjects.

Staron et al. (1992) examined the effects of repeated biopsy sampling on muscle morphology in strength-training and nontraining men and women. The purpose of the study was to compare muscle damage induced by the biopsy procedure alone with damage induced by the biopsy procedure combined with resistance training. One group of subjects performed an 8 wks progressive resistance training program, while a nontraining group served as controls. Biopsy samples were taken from both groups at the beginning and every two weeks during the study. The samples were extracted from m. vastus lateralis at the same location and depth each time. With the exception of one specimen, there was no evidence of ultrastructural damage in

any of the initial samples. Subsequent biopsy samples revealed evidence of fiber degeneration and regeneration in both groups. However, the training subjects had approximately four times as many damaged fibers as the nontraining group (9% vs 2%). In addition, only the training subjects showed evidence of internal disruption within the fibers (e.g., Z band streaming, myofibrillar disruption). The authors concluded that muscle damage induced by the biopsy procedure was not completely repaired after two weeks. Further, they observed: "resistance training appears to cause additional damage primarily consisting of myofibrillar disorganization."

1.5 ADDITIONAL SITES OF MORPHOLOGICAL DAMAGE

1.5.1 Sarcolemma

The sarcolemma (plasma membrane) functions as a barrier to maintain the ionic and electrical gradients between the interior and exterior of the muscle cell. The appearance of muscle proteins (particularly creatine kinase, CK) in the blood following eccentric exercise has been attributed to sarcolemmal disruption (Armstrong, 1990). Clarkson and colleagues have conducted a number of studies which have shown large increases in serum CK following "high-force eccentric exercise" of the elbow flexors (Clarkson and Tremblay, 1988; Clarkson et al., 1986; Ebbeling and Clarkson, 1990). The model they use to induce damage is a series of maximal

eccentric actions performed on a pulley apparatus, in which the investigator pulls down on a lever while the subject resists (for review, see Clarkson et al., 1992). The exercise regimen consists of two sets of 35 maximal eccentric actions (one every 15 s), with 5 min recovery between sets. A characteristic finding of these studies is the extremely delayed time course of CK release: it does not show a precipitous increase until 48 h, and usually reaches peak values approximately 4 d post-exercise (Clarkson et al., 1992). Following various forms of prolonged exercise (e.g., marathon running, endurance triathlons), CK activity typically peaks within 24-48 hours post-exercise (Noakes, 1987).

It is plausible that the delayed CK response following intense eccentric exercise may be due to physical disruption of the membrane. Several investigators have hypothesized that active lengthening of the fiber could physically disrupt the sarcolemma, allowing calcium (Ca^{2+}) to enter the cell across its electrochemical gradient (Armstrong, 1984; Newham et al., 1983a; Stauber, 1989). Abnormally high levels of intracellular Ca^{2+} can rapidly lead to cell dysfunction by stimulating a number of Ca^{2+}-activated proteases and phospholipases (Byrd, 1992). These compounds, active in muscle autolysis, may be responsible for the delayed time course for CK release by further degrading the cell membrane following initial injury (Ebbeling and Clarkson, 1989).

Elevated CK levels have also been demonstrated following isotonic resistance exercise in humans (Hikida et al., 1991; Paul et al., 1989). Paul et al. (1989) compared the serum CK response of weight-trained and untrained subjects following a bout of 6 weight lifting exercises performed at 70-80% 1 RM. Both groups demonstrated significant elevations in CK at 12 and 24 h post-exercise, which the authors attributed to skeletal muscle damage. The magnitude of the response in the untrained group (a 6 fold increase over baseline levels) was much lower than that reported by Clarkson and colleagues following high-force eccentric exercise (a 20-25 fold increase over baseline values). The results of these studies suggest that traditional 6-10 RM resistance training programs cause substantially less muscle damage than the extreme protocols utilized in many experiments (which are not typically used by individuals in training). However, caution must be used when comparing investigations of this nature, since there is considerable variability between (and even within) subjects in the serum enzyme response to exercise (Noakes, 1987). Regardless, until direct evidence of "holes" in the plasma membrane is presented, any explanation of exercise-induced muscle damage based on sarcolemmal disruption will remain hypothetical.

1.5.2 Sarcoplasmic Reticulum

The sarcoplasmic reticulum (SR) helps maintain normal contractile function by regulating the concentration of intracellular free Ca^{2+}. Disruption of the SR could allow intracellular levels of Ca^{2+} to rise precipitously, triggering the cascade of Ca^{2+}-mediated events involved in cell damage mentioned above. Several investigators have hypothesized that the high specific tensions which occur during eccentric muscle actions may mechanically damage the SR (Armstrong, 1990; Ebbeling and Clarkson, 1989). While it is conceivable that variations in sarcomere length (which occur during eccentric actions) could result in physical tearing of the SR, this has not been demonstrated experimentally (Armstrong et al., 1991).

Altered SR function has been observed following exhaustive treadmill exercise in rats (Byrd et al., 1989a) and horses (Byrd et al., 1989b). The authors of these investigations have suggested that metabolic factors (e.g., increased muscle temperature, decreased pH, oxygen free radical production) are responsible for SR dysfunction (for review, see Byrd, 1992). While these mechanisms may affect SR integrity following endurance exercise, it is unlikely that metabolic factors induce structural changes in the SR during forceful eccentric muscle actions. To clarify the mechanisms responsible, it will be necessary to examine SR function following eccentric resistive-type exercise using the standard

techniques employed by Byrd and colleagues (e.g., isolated SR vesicles).

1.5.3 Extracellular Matrix

The extracellular matrix (ECM) of skeletal muscle is an intricate network of macromolecules which surrounds and interconnects individual myofibers. The matrix is comprised of various connective tissues and is anchored to the sarcolemma of each fiber by continuous thin mats of specialized ECM called basal laminae (Alberts et al., 1989). In human muscle, the ECM consists of type IV and V collagen, fibronectin, laminin, and various specific proteoglycans (Stauber, 1989). The collagens are fibrous proteins which strengthen the matrix, fibronectin and laminin are adhesive glycoproteins which help various cells attach to the ECM, and proteoglycans form a gel-like substance in which the other components are imbedded (Alberts et al., 1989).

Damage to the ECM following eccentric muscle action has been documented in both humans (Stauber et al., 1990) and animals (Fritz and Stauber, 1988). Stauber et al. (1990) examined ECM integrity in untrained subjects following 70 maximal (isokinetic) eccentric actions of the elbow flexors. Forty eight h post-exercise, needle biopsy samples revealed separations of the ECM from myofibers, mast cell degranulation and increased plasma constituents (e.g., albumin, fibrinogen) in the extracellular space. Biopsy samples taken from the

non-exercised control arm of each subject did not exhibit any signs of damage. The authors hypothesized that the mechanical strain of the exercise disrupted the ECM, which initiated the inflammatory response (evidenced by the release of mast cell granules) and caused swelling. Since mast cell degranulation is known to release histamine (a known algesic), they also suggested that delayed onset muscle soreness may be mediated by ECM disruption rather than direct myofiber damage.

1.6 SEQUENCE OF EVENTS IN EXERCISE-INDUCED MUSCLE FIBER INJURY

The sequence of events associated with exercise-induced muscle fiber injury is not well defined (Ebbeling and Clarkson, 1989). The general lack of understanding may be largely attributed to methodological differences between investigations, including (i) the type of exercise model used to induce damage, (ii) the duration and intensity of the activity, (iii) the eccentric component of the exercise, and (iv) the variable periods of assessment following the initial insult. Understandably, there are few definitive assertions which can be made regarding the specific time course of events. One of the most consistent findings among many studies, however, is that of delayed myofiber damage. This refers to the common observation that following initial injury, further degradation of the fiber appears to take place

during the ensuing hours (Fridén et al., 1983b; Fridén et al., 1984; Newham et al., 1983a).

Newham et al. (1983a) reported that a larger number of fibers were damaged, and to a greater extent, an average of 30 h post-exercise compared to immediately after the bout (eccentric stair stepping). Fridén et al. (1983b) also reported more extensive damage at 3 d compared to 1 h post-exercise (eccentric cycle ergometry), although the biopsy samples were not from the same subjects. Fridén et al. (1984) observed lipofuscin granules, indicative of lysosomal activity, in areas of myofibrillar disruption 3 d following a bout of eccentric cycle ergometry. Biopsies taken on day 6 after exercise showed fibers which appeared essentially normal. The most common interpretation of these findings is that further degradation of the fiber occurs in the hours after exercise (Fridén and Lieber, 1992). This process is likely initiated by proteolytic and lipolytic systems (intrinsic to the fiber) which degrade damaged tissue before the myofibrillar structure can be repaired (Armstrong, 1990). Additional evidence supporting delayed myofiber damage following eccentric exercise includes the release of muscle specific enzymes (e.g., creatine kinase, lactate dehydrogenase), which also show a characteristic delayed response (Stauber, 1989).

A number of researchers believe that the process of delayed myofibrillar damage is initiated by increased

intracellular calcium, resulting in the activation of numerous compounds which further degrade the fiber (Armstrong, 1984; Armstrong, 1990; Armstrong et al., 1991; Fridén and Lieber, 1992). Armstrong (1990) proposed a model for exercise-induced muscle fiber injury, highlighting the central role of Ca^{2+} in myofiber damage. He noted that all of the potential initiating events (both metabolic and mechanical) could result from, or directly lead to high levels of intracellular Ca^{2+}. Armstrong et al. (1991) slightly revised his previous model (Armstrong, 1990), and presented a five stage model for exercise-induced muscle fiber injury: (1) an *initial event* occurs in the muscle fiber that inaugurates the injury process; (2) the initiating event leads to a focal loss of Ca^{2+} homeostasis (Ca^{2+} *overload phase*), which activates several degradative pathways intrinsic to the fiber; (3) *autogenetic mechanisms* (e.g., Ca^{2+}-activated proteases, lysosomal proteases) are activated at the local site of injury prior to, and continue after, the invasion of phagocytic and inflammatory cells into the sites of injury; (4) the *phagocytic phase* is in evidence 2-6 h after the injury, and functions to remove dead tissue and debris for several days; (5) the *regenerative phase*, beginning 4-5 d after injury, restores the muscle to its normal condition. Very little is known about the mechanisms underlying the first three stages of the model; the phagocytic and regenerative processes appear

similar to those following other types of muscle injury (Armstrong et al., 1992; Carlson and Faulkner, 1983).

1.7 CHANGES IN FORCE PRODUCTION FOLLOWING ECCENTRICALLY-BIASED EXERCISE

1.7.1 Maximal Voluntary Force

Immediately following forceful eccentric exercise, maximal force production is reduced and recovers gradually, such that a strength deficit may still remain after a week or more (Ebbeling and Clarkson, 1989). Serial concentric or isometric actions also lead to an immediate decrement in force production, but maximal strength is typically restored within a few hours (Clarkson et al., 1992). Following 70 maximal "high force" eccentric actions of the elbow flexors (described in section 1.5.1), subjects typically experience an immediate reduction in maximal isometric strength of over 50% (Clarkson et al., 1992). Similar reductions in elbow flexor strength have been observed following 70 maximal eccentric actions performed on a modified arm curl machine (Clarkson and Tremblay, 1988; Ebbeling and Clarkson, 1990) and 70 coupled concentric/eccentric actions on an isokinetic dynamometer (Donnelly et al., 1992). These studies reported strength recoveries ranging from 45-75% (compared to initial values) 4 days post-exercise. It is important to note, however, that the exercise protocols used in these investigations are not

typically employed in resistance training programs. The changes in force production following multiple set, 6-10 RM regimens of concentric and eccentric resistance exercise have not been systematically investigated.

A number of explanations have been offered for the prolonged strength loss observed following eccentric exercise, including (i) the perception of pain (which would prevent the subject from voluntarily producing maximal force), (ii) "overstretched" sarcomeres, and (iii) ultrastructural damage to the contractile tissue or elements involved in excitation-contraction coupling. It is unlikely that the force decrement is due to pain, since the time course for muscle soreness (which typically peaks 48 h post-exercise) is very different from that of strength loss (which peaks immediately after exercise and is partially recovered by 48 h). In addition, electrically stimulated contractions (which "bypass" the central nervous system) are also reduced following eccentric exercise (see section 1.7.2).

The overstretched sarcomere theory proposes that lengthening actions pull some of the fiber's central sarcomeres apart, thereby reducing the number of available cross bridges (Clarkson et al., 1992). Presumably, the sarcomeres are most stretched immediately after the exercise (when strength loss is maximal), and then gradually return to normal resting length (paralleling the recovery of strength). Although there is no evidence to substantiate such a theory,

Newham and Jones (1988) have demonstrated that strength loss is greater when eccentric actions are performed at long rather than short muscle lengths.

Alternatively, the data by Newham and Jones (1988) may be interpreted to suggest that greater ultrastructural damage is caused by lengthening actions performed at long muscle lengths (i.e., the strength loss is due to damaged myofibrils). Indeed, severe myofibrillar disorganization has been observed in muscle biopsy samples immediately following eccentric exercise in humans (Fridén et al., 1983b; Newham et al., 1983a). It appears, however, that the ultrastructural damage becomes worse in the days following the exercise bout, during the time that strength is recovering. Both Fridén (1983b) and Newham (1983a) reported more extensive disruption 1-3 days following eccentric exercise compared to immediately after the bout. If ultrastructural damage is strictly responsible for the reduction in force production, it is not clear why these two parameters do not correlate better.

1.7.2 Electrically Evoked Contractions

The force:frequency relationship has also been examined to assess the inherent force generating capacity of eccentrically exercised muscles (Ebbeling and Clarkson, 1989). Davies and White (1981) reported that, following a 60 min chair stepping exercise, the maximal twitch and tetanic tensions of the triceps surae (at 10,20,50 and 100 Hz) were

substantially reduced in the eccentric leg, while the contralateral concentric leg remained unaffected. Newham et al. (1987) examined the force generated by 20 Hz stimulation expressed as a percent of that at 100 Hz (20/100%) following 60-80 maximal eccentric actions of the elbow flexors. They reported the 20/100% value decreased to approximately 40% of initial immediately post-exercise, and remained significantly depressed until the ninth day after the bout. The most common explanation offered for "low frequency fatigue" is mechanical disruption of the sarcoplasmic reticulum (SR), which would impair normal Ca^{2+} cycling following each action potential (Ebbeling and Clarkson, 1989). Although no direct link has been established, there is evidence to support the possible involvement of the SR in exercise-induced fiber injury (see section 1.5.2).

1.7.3 Rapid Adaptation

Clarkson and Tremblay (1988) have reported that the performance of a single bout of high-force eccentric exercise can exert a protective effect on the fiber, such that the loss of isometric strength following a successive bout of similar exercise is markedly attenuated. In addition to greater force production, subjects experience less muscle soreness and demonstrate reduced levels of serum CK following the second bout of exercise (Clarkson et al., 1992). Although when and how this adaptation occurs is unknown, it has been shown to

occur very rapidly: subjects who performed a second exercise bout 5 d after the first already showed an adaptive response (Ebbeling and Clarkson, 1990). To explain the adaptation, a number of investigators have proposed that the muscle repairs itself after the first bout of exercise, such that the membranes, extracellular matrix, cytoskeleton, and myofibrils are more resilient to future damage (Clarkson and Tremblay, 1988; Fridén et al., 1983a; Stauber, 1989).

An alternative explanation for the rapid adaptation observed is that "fragile" fibers undergo lethal injury during intense eccentric exercise (Armstrong et al., 1983b). A population of fibers may exist which are degenerating or have been weakened through disuse of a motor unit recruitment pattern (Ebbeling and Clarkson, 1989). These "stress-susceptible" fibers may be destroyed during an initial bout of novel eccentric exercise, while stronger, healthier fibers survive and become more resilient to future damage.

1.8 MUSCLE DAMAGE AND THE HYPERTROPHY PROCESS

1.8.1 Importance of Eccentric Muscle Actions in Resistance Exercise

The performance of eccentric muscle actions (EMA) in resistance training programs appears to be crucial for the optimal development of muscle strength (Colliander and Tesch, 1990; Dudley et al., 1991; Komi and Buskirk, 1972) and

hypertrophy (Evans and Cannon, 1991; Hather et al., 1991). Dudley et al. (1991) examined the importance of EMA in strength adaptations to heavy resistance exercise in middle aged men (33 ± 1 y). Three groups of subjects performed leg extension exercises at an intensity equivalent to 6-12 RM, twice a week for 19 weeks. One group (CON/ECC) performed 4-5 sets of concentric and eccentric muscle actions, while the other two groups performed either 4-5 sets (CON) or 8-10 sets (CON/CON) of concentric muscle actions only. Following training, the increase in leg press 3 RM was greater ($p \leq 0.05$) for group CON/ECC (26%) than group CON/CON (15%) or group CON (8%). In the leg extension 3 RM, the increase for group CON/ECC (29%) was greater ($p \leq 0.05$) than group CON (16%), but not group CON/CON (24%). The authors hypothesized that the enhanced strength development observed following combined concentric and eccentric resistance exercise may be due to increased central activation, synchronization of motor units, and/or decreased input from neural inhibitory reflexes. It should be noted, however, that the 3 RM test (which involves 1 concentric and 2 coupled eccentric-concentric actions) may have favoured the CON/ECC training group.

Hather et al. (1991) examined the influence of EMA on skeletal muscle adaptations in the subjects who participated in the study by Dudley et al. (1991). Needle biopsy samples revealed that Type I and II fiber area increased 14% and 32%, respectively, in group CON/ECC post-training. Group CON/CON

only showed an increase in Type II fiber area (27%), while group CON showed no significant increase in either fiber type. Following four weeks of detraining, only group CON/ECC demonstrated mean fiber hypertrophy. It therefore appears that EMA are critical for optimizing muscle fiber hypertrophy during resistance training. Hather et al. (1991) also reported a shift in fiber subtype from IIb to IIa after the training period in all three groups. Type II fiber subtype conversion of this nature have been reported previously (Colliander and Tesch, 1990; Staron et al., 1989). Since all of the groups demonstrated a similar response however, the authors attributed the transformation in Type II subtype to the performance of concentric, rather than eccentric, muscle actions.

1.8.2 Muscle Damage and Satellite Cell Activation

The suggestion that EMA are important for the muscle hypertrophy which occurs in response to resistance training programs (Hather et al, 1991) may appear controversial, since it is generally accepted that EMA also cause substantial muscle damage. It is well established however that mammalian skeletal muscle possesses the ability to regenerate following exercise-induced injury (for review, see Carlson and Faulkner, 1983; Chapman, 1985). Indeed, the damage induced by eccentric exercise may represent an important stimulus for muscle growth (Stauber, 1989). Evans and Cannon (1991) have suggested that

the localized and systemic changes which accompany the repair process may also be essential for muscle hypertrophy.

The activation of satellite cells, a population of unspecialized reserve cells located beneath the basal lamina, is a critical event in muscle fiber regeneration (Carlson and Faulkner, 1983). Following an injury, mononucleated satellite cells fuse to form a multinucleated myotube, which develops into a new muscle fiber to replace the damaged one (Schultz, 1989). The regenerative process appears to follow a common pathway, regardless of the initiating event (Carlson and Faulkner, 1983). Satellite cell proliferation has been observed in animals following a variety of exercise protocols, including downhill running in rats (Darr and Schultz, 1987), and concentric resistance training in cats (Giddings et al, 1985).

Recruitment of satellite cells following injury appears to be confined to the damaged fiber (Schultz et al, 1986), suggesting that the regeneration process is simply one of replacement, with no net increase in fiber number. The role that satellite cell proliferation might play in the muscle hypertrophy response however, is unclear (i.e., is the fiber simply repaired to its original state, or does regeneration include the synthesis of additional contractile material?). Further investigation of the mechanisms underlying muscle damage and the hypertrophy process is clearly warranted before a conclusive link can be established between the two.

1.9 SUMMARY

This review has focused on the potential mechanisms responsible for exercise-induced muscle fiber injury, repair and adaptation in humans. Although the specific initiating event remains unknown, emphasis has been placed on the hypothesis that high specific tensions generated during eccentric muscle action (EMA) physically disrupt the fiber. Metabolic explanations of fiber injury cannot be discounted, but appear confounded by the fact that EMA require less energy, yet cause greater damage, than comparable concentric or isometric actions. It is clear that the fiber is repaired and becomes more resistant to future damage, but the mechanisms by which this is accomplished remain poorly understood.

Most investigations of exercise-induced muscle fiber injury have utilized continuous models of dynamic exercise such as downhill running or eccentric cycle ergometry. Resistive-type exercise models have also been used, but the protocols are extreme and do not mimic the type of exercise typically performed during resistance training. The purpose of the present investigation was to examine the changes in muscle ultrastructure and force production which occur following a single bout of traditional concentric or eccentric resistance exercise. The experiment was designed to systematically examine (i) the extent of fiber disruption immediately after and 48 h following the bout, and (ii) the

changes in voluntary and evoked strength immediately, 24, 48, 72 and 96 h post-exercise. Since resistance training typically involves both a concentric (lifting the weight) and eccentric (lowering the weight) component, a secondary purpose was to determine which component of the exercise resulted in greater fiber damage. This was accomplished by having the subjects lift the weight with one arm and lower it with the other so that the same absolute work was performed by each arm.

CHAPTER II

CHANGES IN SKELETAL MUSCLE ULTRASTRUCTURE AND STRENGTH PERFORMANCE FOLLOWING ACUTE RESISTANCE EXERCISE

2.1 INTRODUCTION

Strenuous, unaccustomed exercise can disrupt the fine structure of skeletal muscle tissue (Fridén et al., 1981, 1983). Activities which contain large eccentric components appear to be especially disruptive (Newham et al., 1983a). Direct evidence of myofibrillar disruption has been observed in needle biopsy samples from human subjects following downhill sprinting (Fridén et al., 1981, 1988), bench stepping (Newham et al., 1983a), and eccentric cycle ergometry (Fridén, 1984; Fridén et al., 1983b; Manfredi et al., 1991; O'Reilly et al., 1987). The most common structural component affected is the Z disc (Fridén et al., 1981, 1983), although disturbances to the A-band (Newham et al., 1983a), mitochondrion (Fridén et al., 1983b, 1988; Manfredi et al., 1991), sarcoplasmic reticulum (Manfredi et al., 1991; O'Reilly et al., 1987), cytoskeleton (Fridén et al., 1984; Waterman-Storer, 1992) and extracellular matrix (Stauber et al., 1990) have also been reported. Indirect evidence of tissue disruption includes the appearance of muscle enzymes in the blood (Clarkson et al., 1986; Schwane et al., 1983), changes in force production

(Davies and White, 1981; Newham et al., 1983b), and delayed-onset muscle soreness (Asmussen, 1956; Schwane et al., 1983).

Eccentric muscle action (EMA) is characterized by active lengthening of the muscle while it produces force. Myofibrillar disruption following eccentrically-biased exercise has been attributed to physical tearing of sarcomeres (Fridén et al., 1981, 1983), since EMA is known to generate greater tension per active fiber than comparable concentric action (Abbott et al., 1952; Asmussen, 1953; Bigland-Ritchie et al., 1976). Metabolic explanations of tissue disruption appear confounded by the fact that EMA requires less energy, yet causes greater disruption, than comparable concentric action (Clarkson et al., 1986; Newham et al. 1983a).

Most of the information regarding exercise-induced muscle fiber injury is based on studies which have utilized continuous models of dynamic exercise. Data regarding myofibrillar disruption following resistance exercise (which contains clearly defined concentric and eccentric components) are scarce. Clarkson and colleagues have conducted a number of investigations examining "high-force eccentric exercise" of the elbow flexors (Clarkson and Tremblay, 1988; Clarkson et al., 1986; Ebbeling and Clarkson, 1990; Stauber et al., 1990), but the protocols are extreme and do not mimic the type of exercise typically performed during resistance training. In addition, most of these studies have used indirect markers (e.g., serum CK, changes in force production) to assess the

extent of fiber disruption. Stauber et al. (1990) observed direct evidence of tissue disruption following 70 maximal eccentric actions of the elbow flexors, but the focus of the investigation was the extracellular matrix (not myofibrillar fine structure).

Recent evidence suggests that traditional (6-10 RM) resistance exercise causes skeletal muscle disruption in humans (Paul et al., 1989; Staron et al., 1992). Staron et al. (1992) reported evidence of myofibrillar disruption (including Z band streaming) in needle biopsy samples following 8 wks of progressive resistance training. No study has been undertaken, however, to systematically investigate myofibrillar disruption following a single bout of traditional resistance exercise in humans. The purpose of the present investigation, therefore, was to examine the changes in muscle ultrastructure and force production which occur following a single bout of elbow flexor resistance exercise. A secondary purpose was to determine which component of the exercise resulted in greater fiber disruption. This was accomplished by having the subjects lift the weight (concentric action) with one arm, and lower it (eccentric action) with the other, so that the same absolute work was performed by each arm. Specifically, we examined (i) the extent of fiber disruption immediately after and 48 h following the bout, and (ii) the changes in voluntary and evoked strength immediately, 24, 48, 72 and 96 h post-exercise.

2.2 METHODOLOGY

2.2.1 Subjects

Eight healthy males (age = 21.8 ± 0.9 yr, height = 181.0 ± 5.3 cm, mass = 79.8 ± 5.5 kg), with no previous resistance training history, were recruited for the investigation. All subjects were fully advised as to the purposes of the study and associated risks, and gave written informed consent. The project was approved by the Human Ethics Committee of McMaster University (Appendix II).

2.2.2 Experimental Protocol

Subjects performed an isolated bout of elbow flexor resistance exercises on an incline bench using a standard weight training dumbbell. Only concentric muscle actions were performed with one arm (CON arm), and only eccentric muscle actions with the contralateral arm (ECC arm). The designation of each arm as either "CON" or "ECC" was randomly assigned and counterbalanced, so that in 4 subjects the CON arm was their dominant arm, and in 4 subjects the CON arm was their non-dominant arm. Prior to the exercise bout, each subject underwent a progressive loading protocol to determine the maximum weight that could be lifted (from full extension) with the CON arm (i.e., one repetition maximum; 1 RM). Exercise intensity was established as 80% of CON arm 1 RM.

Subjects were instructed to perform 8 sets of 8 repetitions, with 3 minutes recovery between sets. Each "repetition" actually consisted of the following sequence: (i) the subject began with the CON arm fully extended and the ECC arm fully flexed; (ii) an attendant placed the dumbbell in the hand of the CON arm, and the subject performed a concentric muscle action by raising the weight to the fully flexed position; (iii) upon completion, the attendant removed the weight from the hand of the CON arm and placed it into the hand of the fully flexed ECC arm; (iv) the subject then performed an eccentric muscle action by lowering the weight to full extension; (v) upon completion, the attendant removed the weight from the hand of the ECC arm, and the subject resumed the initial starting position (CON arm fully extended, ECC arm fully flexed). The range of motion that each arm went through (CON arm: full extension to full flexion; ECC arm: full flexion to full extension) was approximately 150°. No assistance or "spotting" was given, but subjects were verbally encouraged to maintain effort until they reached volitional failure. In those instances when the subject failed to complete 8 concentric arm actions, the same number of eccentric arm actions were performed with the opposite arm. Each subject therefore performed the same absolute amount of work with each arm.

Subjects were instructed to perform the CON and ECC phases of the movement in 2.0 s each. To monitor this, an

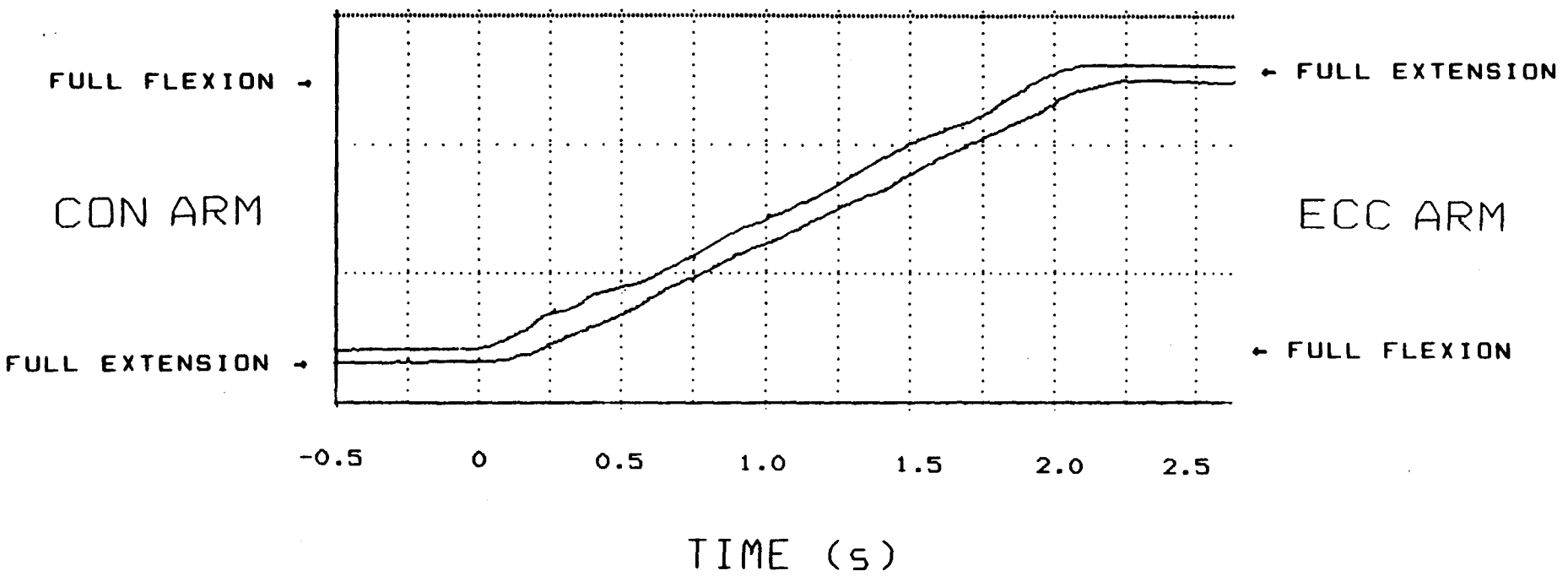
electro-goniometer (L-gon) was strapped to each arm during the exercise session. The device provided an analog voltage output signal at different elbow joint positions. A custom-made software package enabled the signals to be viewed over time, in the form of two velocity tracings on an oscilloscope. Both the subject and investigator were provided with constant feedback of the elbow position of each arm. The positioning of the CON arm L-gon was in opposition to the ECC arm L-gon, so that at identical arm velocities, their respective tracings "mapped" over one another (i.e., the velocity tracing formed by flexing the CON arm was identical to the trace formed by extending the ECC arm; see Figure 1). During the training session, the subjects were instructed to "match" the velocity tracing formed when the CON arm flexed with the tracing formed when the ECC arm extended.

2.2.3 Strength Measurements

A) Isometric strength and evoked contractile properties

Voluntary isometric peak torque (MVC) and evoked isometric twitch contractile properties of the elbow flexors were measured on a custom made dynamometer as described previously (McCartney et. al., 1988). Briefly, the upper arm rested on a horizontal plate and the supinated forearm was secured in the vertical plane with velcro straps to a second plate. The two plates were secured about a shaft, fixed to

Figure 1. Sample oscilloscope tracing of concentric (CON) and eccentric (ECC) arm action during the exercise session (axes have been modified slightly for clarity).


FULL FLEXION →
CON ARM
FULL EXTENSION →
← FULL EXTENSION
ECC ARM
← FULL FLEXION
-0.5
0
0.5
1.0
1.5
2.0
2.5
TIME (s)

produce an elbow joint angle of 1.83 rad (105°; 3.14 rad = 180° = full elbow extension). Previous research with this equipment has demonstrated that this joint angle lies along the flattened peak of the isometric strength curve for untrained athletes (Tsunoda et al., 1993) and near the maximum point of the peak twitch torque curve (O'Hagan et al., 1993). The shaft about which the plates were secured was equipped with a torque transducer consisting of strain gauges. The torque signal from the gauges was amplified, converted to a digital signal, and fed to a computer for on-line analysis.

Maximum, isometric twitch contractions of the elbow flexors were evoked by percutaneous stimulation. Large (3x4 cm) lead plate electrodes were placed over the belly of the biceps brachii and the forearm flexor compartment. These areas were marked with indelible ink to ensure that the electrodes were placed in the same position at each measurement session. Conductance cream was placed on the electrodes before they were secured to the arm with surgical tape. 50-100 μs square wave stimuli (Digitimer Stimulator model 3072, Great Neck, NY) were delivered with increasing voltage intensity until no further increase in twitch torque was observed on the storage oscilloscope. It was assumed that full activation of the muscle was achieved when twitch torque failed to increase with further increases in stimulus intensity. Voltage intensity at full activation ranged from 200-400 V. A custom-made software package enabled peak twitch

torque (N·m), time to peak torque (ms), half relaxation time (ms) and maximum rate of torque development ($N \cdot m \cdot s^{-1}$) to be measured. Three separate trials (for each arm) were administered and recorded, and the greatest twitch torque achieved used for statistical analysis. Twitch contractions were evoked prior to MVC measurements to avoid potentiation effects. The previously determined reproducibility (method error) of the evoked contractile and MVC measurements using this system ranged from 3-7%. During these and all subsequent strength measurement trials, the CON arm data was always collected prior to the ECC arm data.

MVC was considered to be the highest force production over a 5 s effort. This duration has been shown to be adequate for subjects to attain peak force (Sale, 1991). Three trials for each arm were given, interspersed with 60 s rest, and the highest value selected as the peak torque. The interpolated twitch technique, as described previously (Belanger and McComas, 1981), was used to assess the extent of motor unit activation during the maximal voluntary contractions. Briefly, if the application of a maximal indirect stimulus during a maximum voluntary contraction fails to evoke any additional torque, it is considered that all motor units are recruited and firing at optimal frequencies for tension development.

B) Concentric strength

Maximal low- and high-velocity concentric peak torque measurements of the elbow flexors were determined on an isokinetic dynamometer (Cybex II, Ronkonkoma, NY), and recorded on a two channel oscillograph recorder (Hewlett Packard 7402A). The terms "low" (LV) and "high" (HV) velocities corresponded to angular velocities of 0.52 $rad \cdot s^{-1}$ (30°/s) and 3.14 $rad \cdot s^{-1}$ (180°/s), respectively. The test of elbow flexor strength was conducted as described previously (Sale et al., 1987). The subject sat with the upper arm resting on a padded table and the elbow joint axis was aligned with the axis of the dynamometer lever arm. He grasped the handle of the lever arm with the forearm supinated, and began the contractions with the elbow fully extended. Two to four submaximal warm-up trials with each arm were allowed, followed by 1-2 min rest. Two maximal concentric actions at each velocity were then performed, interspersed with 1 min rest, for each arm. For the test trials, subjects were instructed to continue exerting maximal effort throughout the full range of movement. The velocity tests were administered in order from slow to fast, as suggested by Sale (1991) when conducting repeated tests on athletes.

2.2.4 Quantification of Fiber Disruption

A) Muscle Biopsies

Five percutaneous needle biopsy samples (Bergström, 1962) were obtained under local anaesthesia from each subject. A 50 mL syringe was used to provide "suction" in order to facilitate adequate sample sizes (Evans et al., 1982). One sample was taken from the distal lateral portion of the biceps brachii of the non-dominant arm 4-8 weeks prior to the training session, in order to establish "baseline" indices of myofibrillar configuration. Needle biopsy samples were similarly obtained from the biceps of both the CON and ECC arm immediately following the training session (within 20 min), and again 48 h following the training session.

Each sample was dissected of fat and connective tissue and appropriately divided into two sections. One section was immediately subdivided into several smaller pieces and placed in a fixative (2% gluteraldehyde) for electron microscopy preparation. The remainder of the sample was mounted in an embedding medium (OCT), and frozen by submersion in isopentane cooled with liquid nitrogen for subsequent histochemical and immunohistochemical analysis.

B) Electron Microscopy

Following initial fixation, the tissue samples were post-fixed in osmium tetroxide, dehydrated in graded baths of ethyl alcohol, and embedded in an epoxy resin (Spurr's). Four

to eight tissue blocks per specimen were obtained in this manner. Each block was semi-thin sectioned (0.5 μm) and stained with toluidine blue for light microscopy. An area containing at least 10 longitudinally oriented muscle fibers was identified, and ultra-thin sections (60-70 nm) were cut for electron microscopy. The sections were stained with uranyl acetate and lead citrate, mounted on 200 square copper grids, and viewed through a Philips 301 electron microscope (Philips Industries, Eindhoven, The Netherlands).

Individual fibers from each biopsy were studied and classified according to the extent of myofibrillar disruption (Table 1). To be included in the analysis, a fiber had to occupy at least 3 continuous square copper grids (in longitudinal section), or an area of 0.0217 mm^2. A maximum of 4 continuous grids (0.0289 mm^2) were examined per fiber. Based on this criterion, 40.3 ± 6.6 fibers (mean ± SD; range: 30-54) from each sample were examined in a blind fashion (the investigator knew which subject the sample was taken from, but was not aware of the sampling condition).

Specifically, a fiber was considered disrupted if the myofibrillar architecture of any sarcomere appeared "smeared." An area of disruption occupying 1-2 adjacent myofibrils and/or 1-2 continuous sarcomeres was designated "focal" (Newham et al., 1983a). A "moderate" area of disruption was defined as an area encompassing 3-10 continuous sarcomeres and/or 3-10 adjacent myofibrils. A fiber which contained more than 10

Table 1. Myofibrillar disruption classification scheme

Focal	Area encompassing 1-2 continuous sarcomeres and/or 1-2 adjacent myofibrils
Moderate	Area encompassing 3-10 continuous sarcomeres and/or 3-10 adjacent myofibrils, or more than 10 "focal" areas
Extreme	Area encompassing >10 continuous sarcomeres and >10 adjacent myofibrils, or more than 10 "moderate" areas

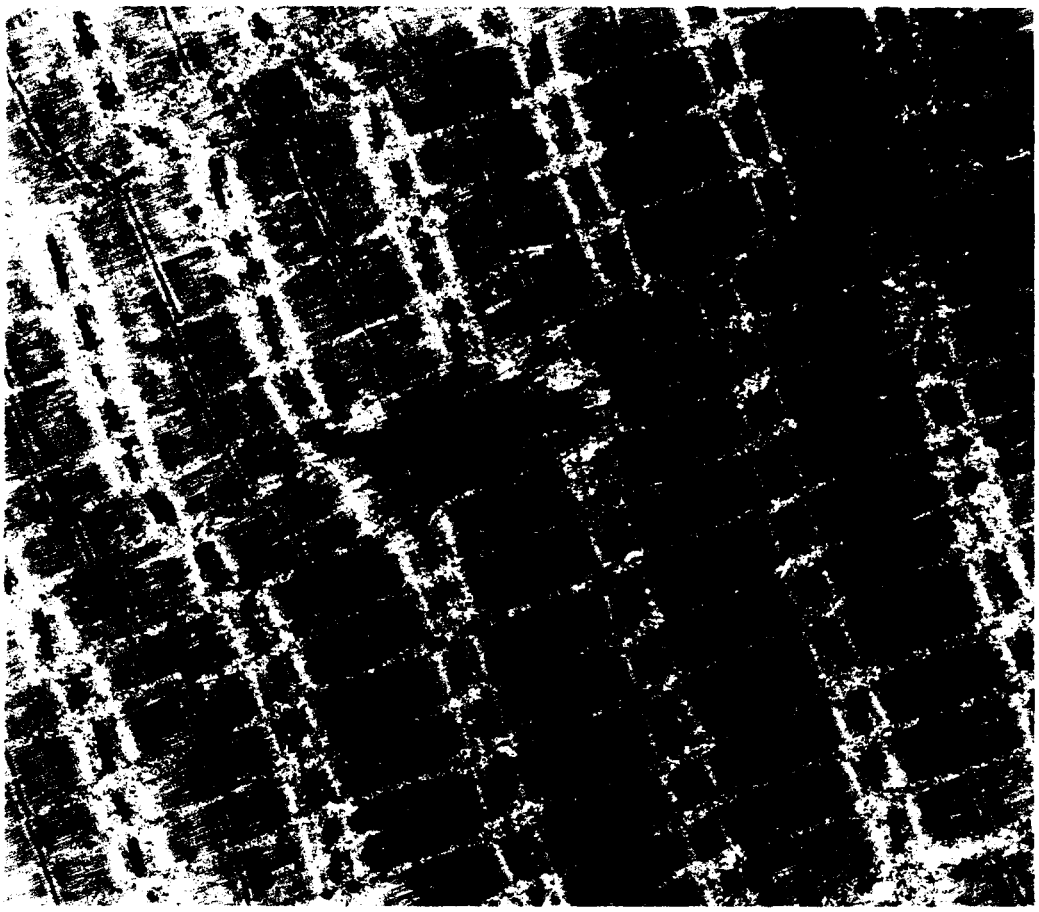


Figure 2. Micrograph illustrating focal area of disruption (x2600).

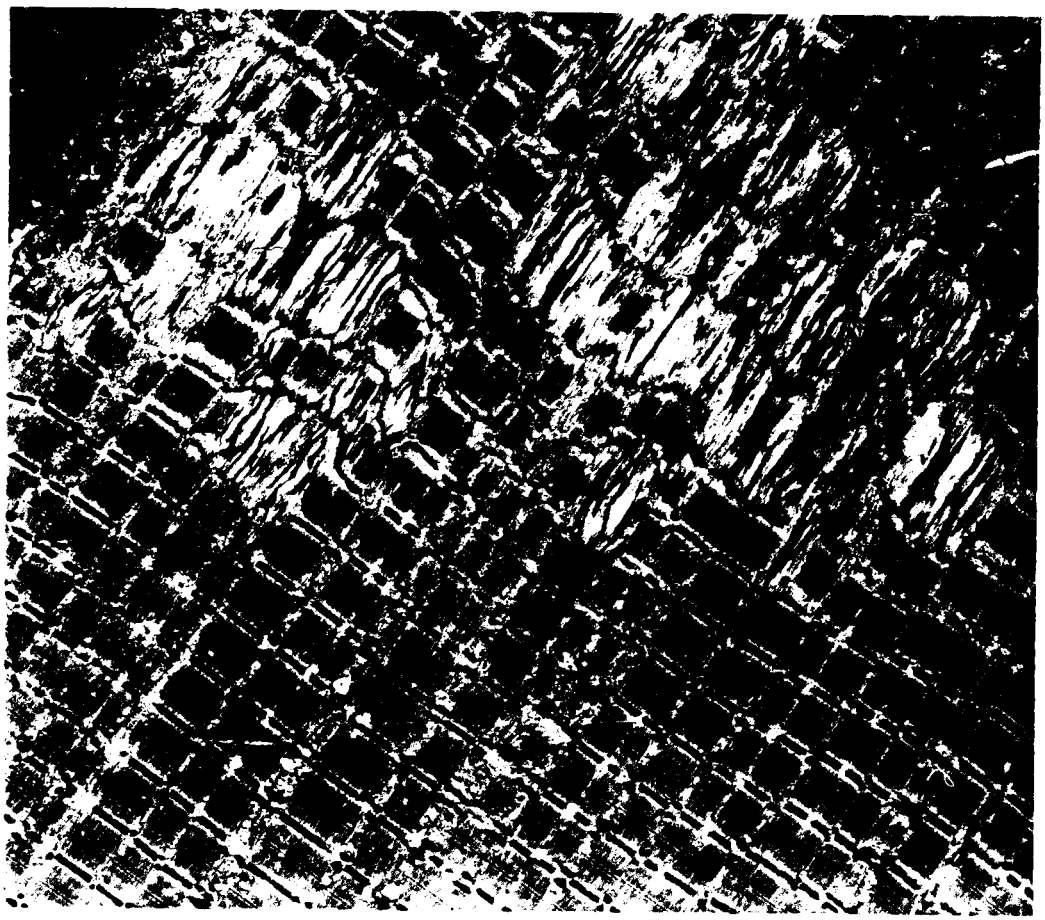


Figure 3. Micrograph illustrating moderate area of disruption (x720).

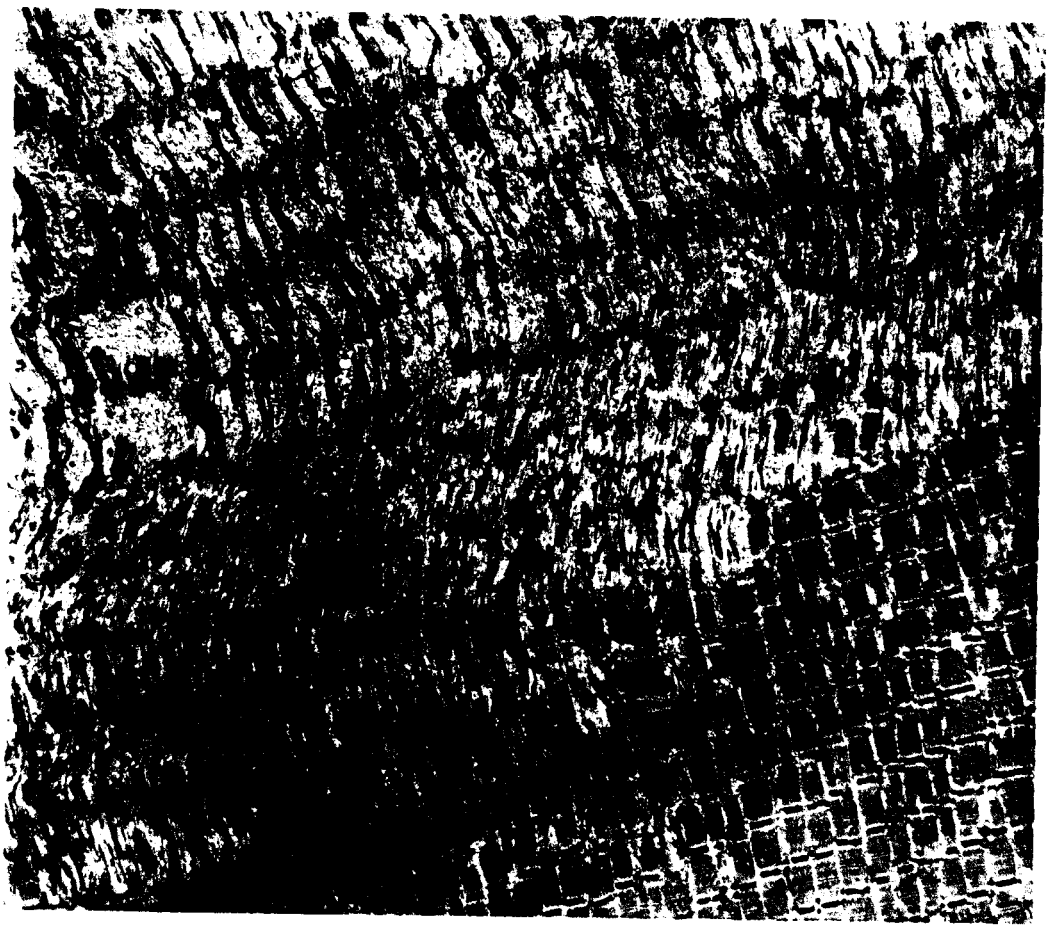


Figure 4. Micrograph illustrating extreme area of disruption (x550).

focal areas of disruption was also given a "moderate" rating. Finally, an area of disruption which spanned more than 10 continuous sarcomeres and 10 adjacent myofibrils was defined as "extreme". A fiber which contained more than 10 moderate areas of disruption was also given an "extreme" rating. (Refer to figures 2, 3 and 4 for sample photographs of each disruption category). For statistical analysis, the extent of fiber disruption was quantified in terms of (i) the number of disrupted fibers (percent of total), and (ii) the percentage of fibers in each disruption category (i.e., focal, moderate, extreme).

C) Histochemistry and Immunohistochemistry

The tissue was sectioned at -20°C in a cryostat at a thickness of 8 μm and collected on precleaned glass slides. Serial sections were collected by placing 2 sections per slide on 6 consecutively numbered slides. The first 3 slides from each sample were used to determine muscle fiber composition: following preincubation at pH 4.3, 4.6 or 10.0, the fibers were classified as either type I, IIa or IIb after staining for myofibrillar ATPase activity at pH 9.4. The remaining 3 slides were stored in airtight containers at -20°C until further immunohistochemical analysis. [As of this date, the immunohistochemical analysis (under the direction of Dr. William Stauber at West Virginia University) had not been

completed. The results of this analysis will be included in the final manuscript to be submitted for publication.].

2.2.5 Data Analysis

Strength and contractile property data were analyzed using a two factor (2x6; arm x measurement time) repeated measures ANOVA. Electron microscopy data were analyzed using: (i) a one factor (1x5; sampling time) repeated measures ANOVA to examine the total number of fibers disrupted at each biopsy sampling time, and (ii) a two factor (2x2; arm x sampling time) repeated measures ANOVA to examine the severity of fiber disruption in the post-exercise samples. Statistical significance was accepted as $P \leq 0.05$. Significant main effects and interactions were further analyzed using a Tukey HSD post hoc test.

2.3 RESULTS

2.3.1 Strength Measurements

A) Isometric strength

Changes in isometric peak torque (MVC) and motor unit activation (MUA) for each arm are presented in Table 2. In the CON arm, MVC decreased ($P \leq 0.05$) below pre-exercise (PRE) values immediately post-exercise (POST), but did not differ from PRE at any other measurement time (24, 48, 72 or 96 h following the exercise bout). In the ECC arm, MVC was lower

($P \leq 0.05$) POST, 24, 48, 72 and 96 h compared to PRE values. The changes in MVC are illustrated in Figure 5. MUA demonstrated a main effect for measurement time: when the data were collapsed across arms, MUA was significantly ($P \leq 0.05$) lower POST compared to PRE, but did not differ from PRE at any other measurement time.

B) Concentric strength

Changes in low (LV) and high velocity (HV) concentric peak torque for each arm are presented in Table 2. In the CON arm, LV and HV showed similar changes: both decreased ($P \leq 0.05$) below PRE values POST, but did not differ from PRE at 24, 48, 72 or 96 h. In the ECC arm, LV decreased ($P \leq 0.05$) below PRE values POST, 24, 48, 72 and 96 h; HV decreased below PRE values POST, 24, 48 and 72 h, but was not significantly different at 96 h. The changes in LV and HV are illustrated in Figures 6 and 7, respectively.

2.3.2 Evoked Contractile Properties

Changes in peak twitch torque (PTT), time to peak torque (TPT), maximum rate of torque development (MRD), and half relaxation time (HRT) are summarized in Table 3. PTT and MRD showed similar changes: in the CON arm, both decreased ($P \leq 0.05$) below PRE values POST, but did not differ from PRE at 24, 48, 72 or 96 h. In the ECC arm, PTT and MRD decreased ($P \leq 0.05$) below PRE values POST, 24, 48, 72 and 96 h. HRT

decreased ($P \leq 0.05$) POST, 24 and 48 h in the ECC arm only, and TPT remained unchanged in either arm during the entire measurement period. The changes in PTT are illustrated in Figure 8.

2.3.2 Quantification of Fiber Disruption

The 5 biopsy sampling times are abbreviated as follows:

1. Baseline (pre-exercise): BASE
2. Immediately post-exercise (CON arm): POST-CON
3. Immediately post-exercise (ECC arm): POST-ECC
4. 48 h post-exercise (CON arm): 48H-CON
5. 48 h post-exercise (ECC arm): 48H-ECC

Examination of the BASE samples revealed that 96.9% of the fibers appeared normal (i.e., no evidence of fiber disruption) before the exercise session. Although half of the BASE samples (4 of 8) contained some evidence of fiber disruption, the overall number of affected fibers was quite small: 1.1% "focal" and 2.0% "moderate". Immediately following the exercise bout, a significantly greater number of fibers appeared disrupted in the POST-CON and POST-ECC samples compared to BASE. However, significantly more fibers were disrupted in the POST-ECC (81.7%) compared to the POST-CON (33.2%) samples.

The extent of fiber disruption observed in the 48H-CON and 48H-ECC samples was similar to those samples obtained

immediately post-exercise. Both samples contained a greater number of disrupted fibers compared to BASE, although significantly more fibers appeared disrupted in the 48H-ECC (79.9%) compared to 48H-CON (37.4%) samples. The total number of disrupted fibers, and number of disrupted fibers within each category, are illustrated in Figures 9 and 10, respectively. These data are also summarized in Table 4.

In addition to greater absolute disruption (i.e., number of fibers affected), the severity of disruption was also greater in the samples from the ECC arm. The number of fibers with "extreme" disruption showed a main effect for arm when the post-exercise data was collapsed across biopsy time: a greater ($P \leq 0.05$) number of fibers were disrupted in the samples taken POST-ECC (40.6%) and 48H-ECC (49.6%) compared to POST-CON (12.8%) and 48H-CON (17.1%).

The percent decrease in MVC at 48 h in the ECC arm significantly correlated ($P \leq 0.05$) with the percentage of fibers showing "extreme" disruption in the samples taken 48 h post-exercise from the ECC arm ($y = -0.98x + 118.52$, $r = -0.745$, $r2 = 0.556$). There was also a significant correlation ($P \leq 0.05$) between the percent decrease in MVC at 48 h in the CON arm and the percentage of fibers showing "extreme" disruption in the samples taken 48 h post-exercise from the CON arm ($y = -1.86x + 189.20$, $r = -0.864$, $r^2 = 0.746$). These correlations are illustrated in Figures 11 and 12, respectively.

Table 2. Voluntary strength measures and motor unit activation

	MVC (N·m)	LV (N·m)	HV (N·m)	MUA (%)
CON arm:				
PRE	74.3± 3.6	48.6± 1.6	34.1± 2.2	95.4± 2.0
POST	58.8± 2.8*	43.3± 2.3*	29.6± 1.9*	79.9± 4.7
24 HR	67.6± 4.5	46.3± 3.1	33.3± 2.9	90.9± 3.1
48 HR	68.5± 3.9	49.9± 2.6	33.8± 2.1	89.6± 2.6
72 HR	70.5± 4.2	49.3± 2.1	35.5± 2.2	97.3± 1.0
96 HR	73.0± 4.0	50.3± 2.0	25.8± 2.1	93.1± 2.3
ECC arm:				
PRE	72.0± 4.5	48.6± 2.6	35.6± 2.4	93.1± 1.8
POST	48.8± 7.0*+	31.6± 4.0*+	24.1± 3.1*+	87.2± 5.2
24 HR	46.4± 7.6*+	28.3± 4.2*+	24.1± 2.9*+	84.3± 9.2
48 HR	43.8± 9.2*+	31.9± 5.4*+	25.8± 4.5*+	90.5± 3.8
72 HR	55.4± 7.6*+	36.5± 4.6*+	32.1± 3.8*+	89.9± 3.5
96 HR	61.9± 7.6*+	39.9± 5.0*+	33.0± 4.0	95.1± 2.1

Voluntary strength measures and motor unit activation of the elbow flexors of the concentric (CON) and eccentric (ECC) arm at each measurement time (mean ± SE). MVC = isometric peak torque, LV = low-velocity (0.52 $rad \cdot s^{-1}$) concentric peak torque, HV = high-velocity (3.14 $rad \cdot s^{-1}$) concentric peak torque, MUA = motor unit activation.

* $P \leq 0.05$ from PRE (pre-exercise) value.

+ $P \leq 0.05$ between CON and ECC arms.

Table 3. Evoked contractile properties

	PTT (N·m)	TPT (ms)	MRD ($N \cdot m \cdot s^{-1}$)	HRT (ms)
CON arm:				
PRE	8.6± 0.8	56.2± 2.2	316.6± 31.3	67.1± 4.0
POST	5.1± 0.4*	53.6± 3.4	199.3± 22.2*	62.3± 4.3
24 HR	8.7± 0.9	53.7± 3.2	317.7± 38.1	64.6± 2.1
48 HR	8.4± 0.5	54.1± 2.6	282.6± 20.6	68.1± 2.5
72 HR	8.5± 0.8	60.3± 4.4	281.5± 28.6	66.8± 2.9
96 HR	8.1± 0.7	62.1± 2.6	278.1± 22.4	62.0± 2.3
ECC arm:				
PRE	8.6± 0.9	52.4± 3.1	317.7± 35.6	70.0± 2.2
POST	2.7± 0.5*+	42.0± 4.6	122.0± 26.2*+	28.6± 5.1*+
24 HR	6.0± 1.4*+	51.3± 4.9	223.2± 49.3*+	47.8± 7.6*+
48 HR	6.6± 1.4*+	53.7± 3.1	235.8± 44.7*+	54.8± 7.9*+
72 HR	6.9± 1.2*+	54.8± 3.4	248.6± 45.3*+	67.4± 5.3
96 HR	6.8± 1.0*+	57.7± 2.5	236.6± 32.4*+	65.3± 6.4

Evoked contractile properties of the elbow flexors of the concentric (CON) and eccentric (ECC) arms at each measurement time (mean ± SE). PTT = peak twitch torque, TPT = time to peak torque, MRD = maximum rate of torque development, HRT = half relaxation time

* $P \leq 0.05$ from PRE (pre-exercise) value.

+ $P \leq 0.05$ between CON and ECC arms.

Table 4. Quantification of Disrupted Fibers

	BASELINE	POST-CON	POST-ECC	48H-CON	48H-ECC
FOC	1.1± 0.7	11.4± 2.2	15.6± 5.3	7.0± 2.5	10.2± 4.1
MOD	2.1± 1.3	9.1± 3.2	25.6± 5.2	13.2± 4.4	20.0± 5.7
EXT	0.0± 0.0	12.8± 6.2	40.6±11.7$^{\#}$	17.1± 7.1	49.7±12.0$^{\#}$
TOT	3.1± 1.3	33.2± 6.7*	81.7± 6.3^{*+}	37.4±10.0*	79.9± 9.1^{*+}

Number of disrupted fibers (percent of total; mean ± SD) in each category at each biopsy time: (i) pre-exercise (BASELINE); (ii) immediately post exercise: concentric (POST-CON) and eccentric (POST-ECC) arm; (iii) 48 h post exercise: concentric (48H-CON) and eccentric (48H-ECC) arm. FOC = focal disruption, MOD = moderate disruption, EXT = extreme disruption, TOT = total number of disrupted fibers.

* $P \leq 0.05$ compared to BASELINE

$^{+}$ $P \leq 0.05$ between CON and ECC arms at same biopsy time

$^{\#}$ $P \leq 0.05$ between CON and ECC arms across all post-exercise biopsy times (main effect for arm)

Figure 5. Isometric peak torque values in the CON and ECC arms at each measurement time (expressed as percent changes from pre-exercise baseline values; mean ± SE).

* $P \leq 0.05$ from PRE (pre-exercise) value.

+ $P \leq 0.05$ between CON and ECC arms.

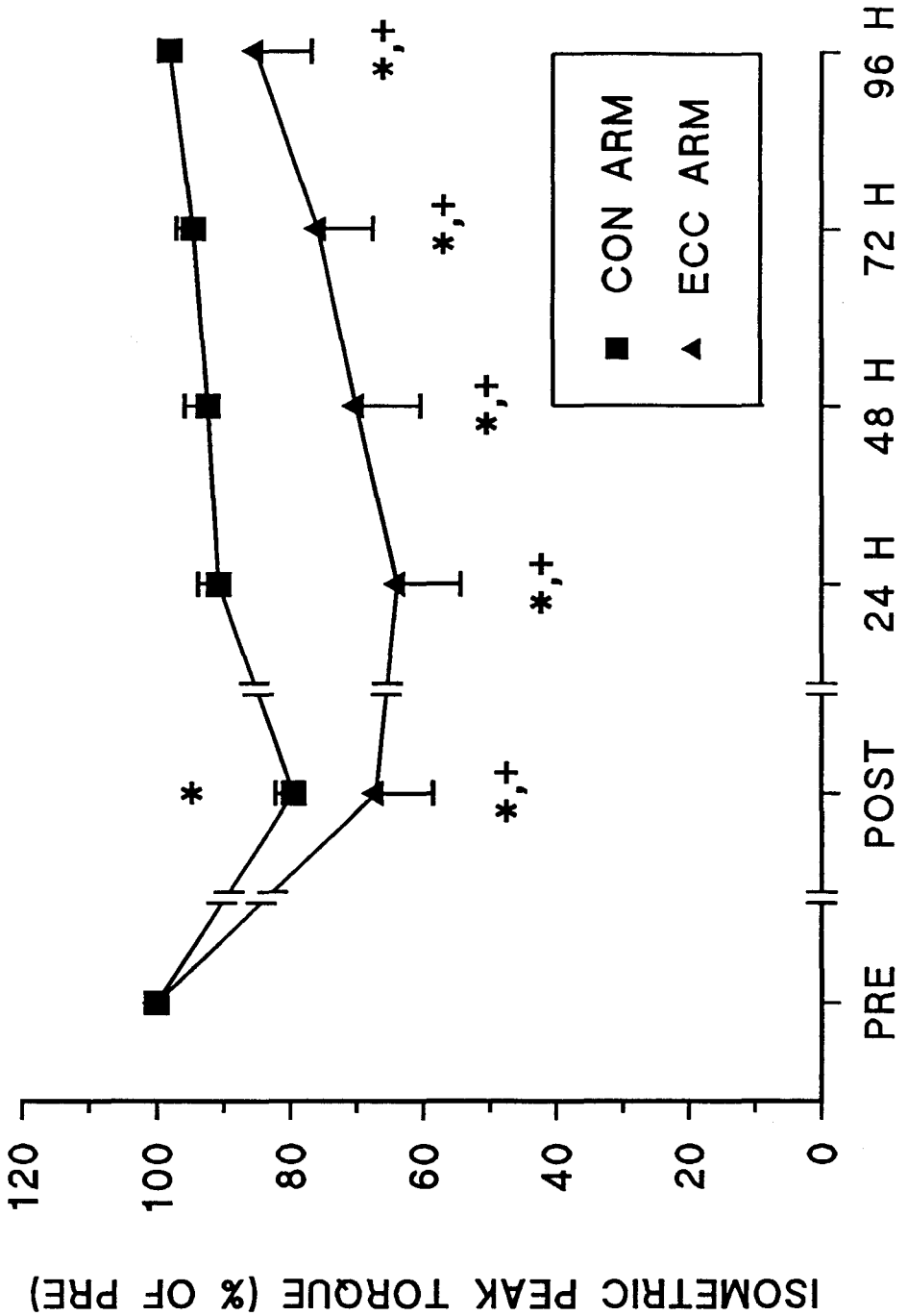

ISOMETRIC PEAK TORQUE (% OF PRE)
120
100
80
60
40
20
0
PRE
POST
24 H
48 H
72 H
96 H
CON ARM
ECC ARM
*
*,+
*,+
*,+
*,+
*,+

Figure 6. Low velocity concentric peak torque values in the CON and ECC arms at each measurement time (expressed as percent changes from pre-exercise baseline values; mean ± SE).

* $P \leq 0.05$ from PRE (pre-exercise) value.

+ $P \leq 0.05$ between CON and ECC arms.

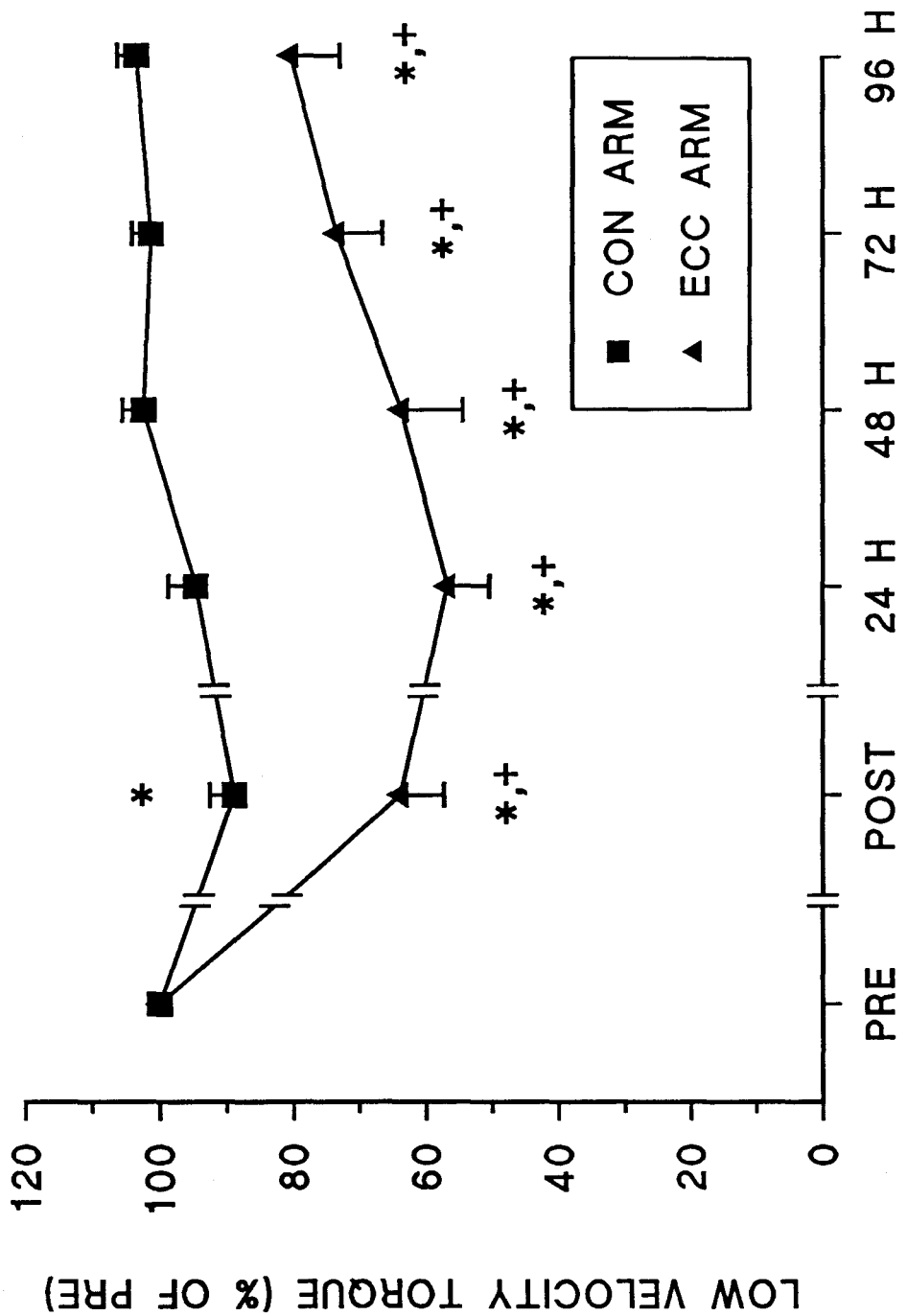

LOW VELOCITY TORQUE (% OF PRE)
120
100
80
60
40
20
0
PRE
POST
24 H
48 H
72 H
96 H
CON ARM
ECC ARM
*
*,+
*,+
*,+
*,+
*,+

Figure 7. High velocity concentric peak torque values in the CON and ECC arms at each measurement time (expressed as percent changes from pre-exercise baseline values; mean ± SE).

* $P \leq 0.05$ from PRE (pre-exercise) value.

+ $P \leq 0.05$ between CON and ECC arms.

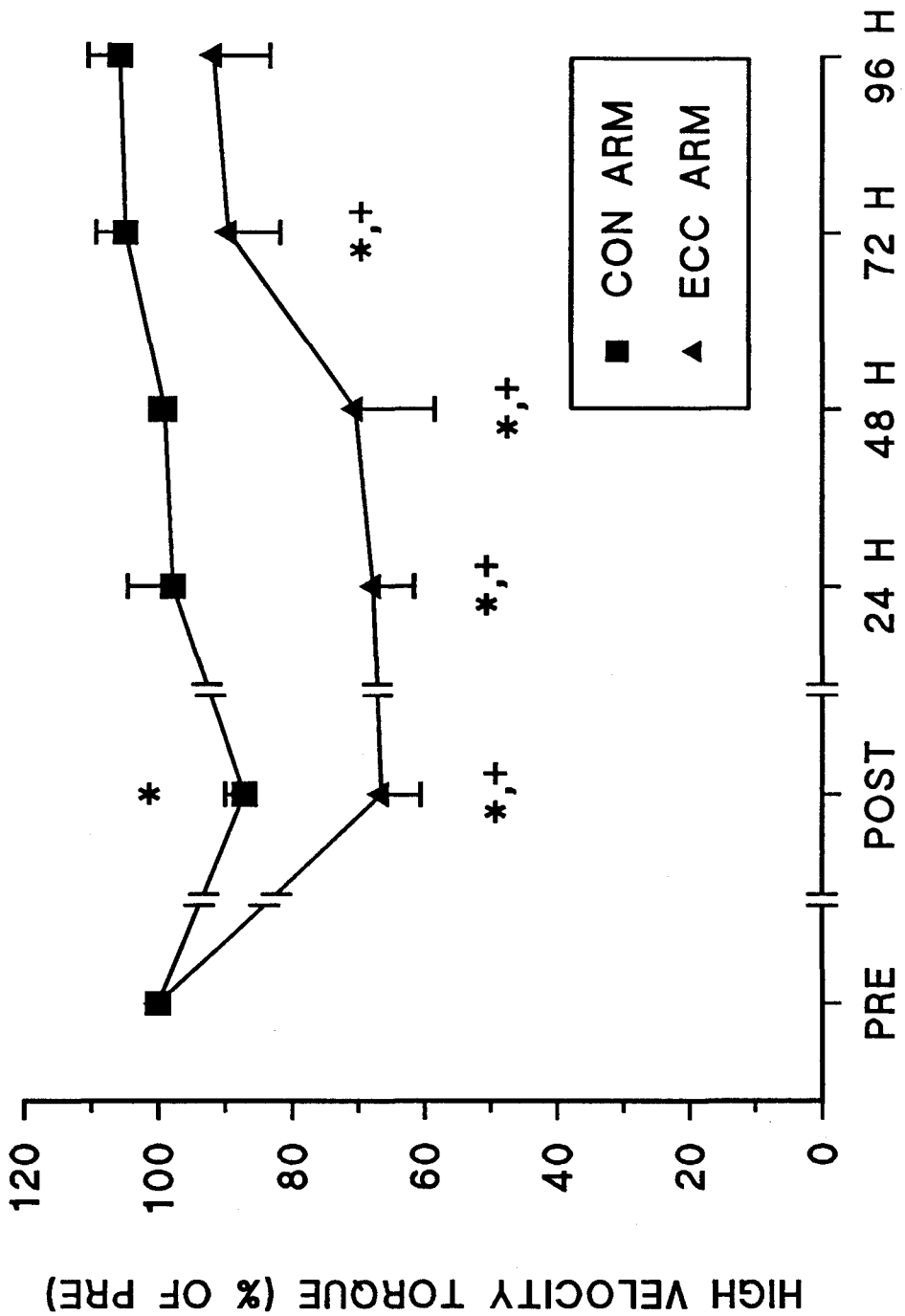

HIGH VELOCITY TORQUE (% OF PRE)
120
100
80
60
40
20
0
PRE
POST
24 H
48 H
72 H
96 H
CON ARM
ECC ARM
*
*,+
*,+
*,+
*,+

Figure 8. Peak twitch torque values in the CON and ECC arms at each measurement time (expressed as percent changes from pre-exercise baseline values; mean ± SE).

* $P \leq 0.05$ from PRE (pre-exercise) value.

+ $P \leq 0.05$ between CON and ECC arms.

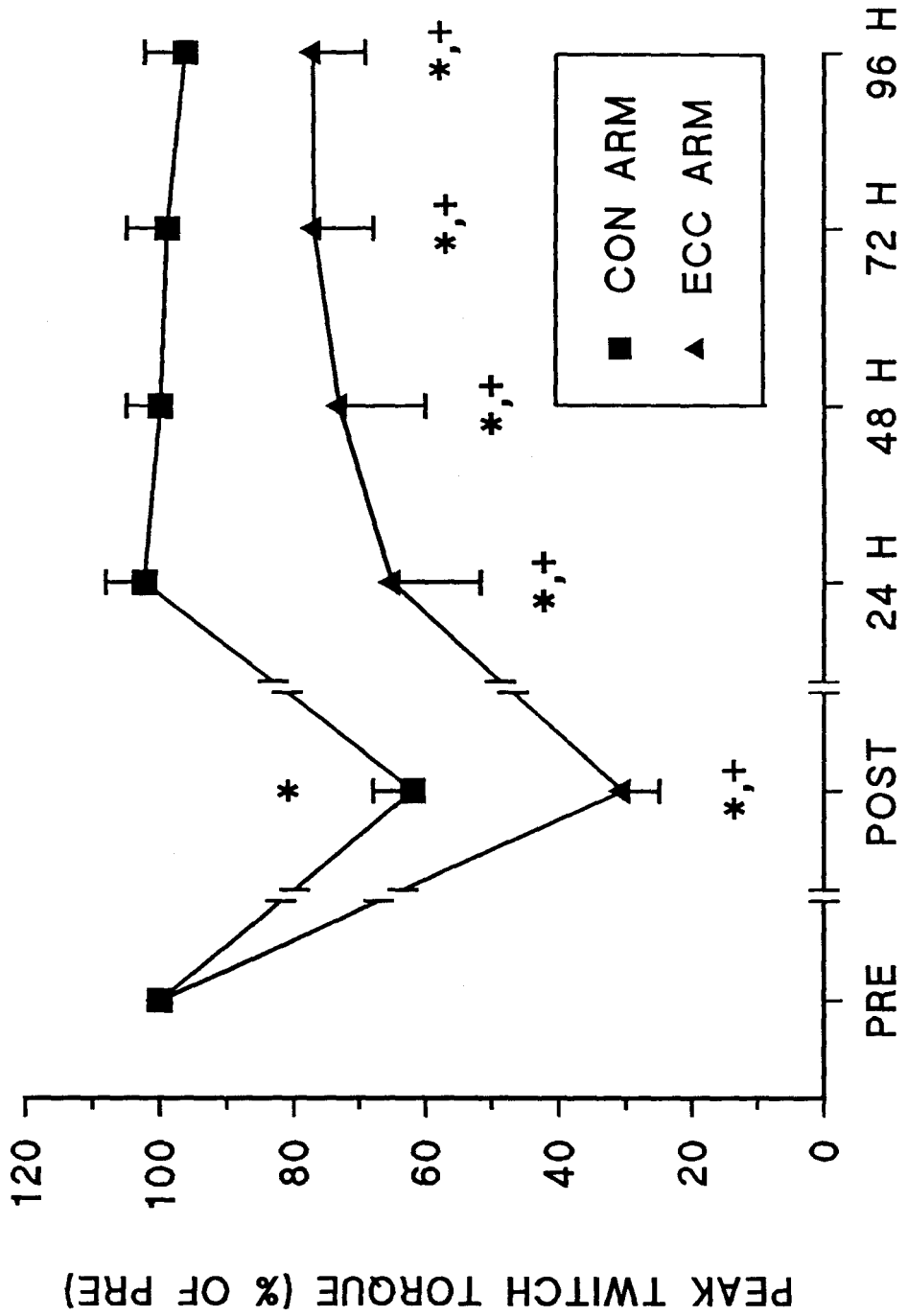

PEAK TWITCH TORQUE (% OF PRE)
120
100
80
60
40
20
0
PRE
POST
24 H
48 H
72 H
96 H
CON ARM
ECC ARM
*
*,+

Figure 9. Number of disrupted fibers observed at each biopsy sampling time (expressed as percent of total; mean ± SE).

* $P \leq 0.05$ compared to BASELINE.

** $P \leq 0.05$ between arms at same biopsy time.

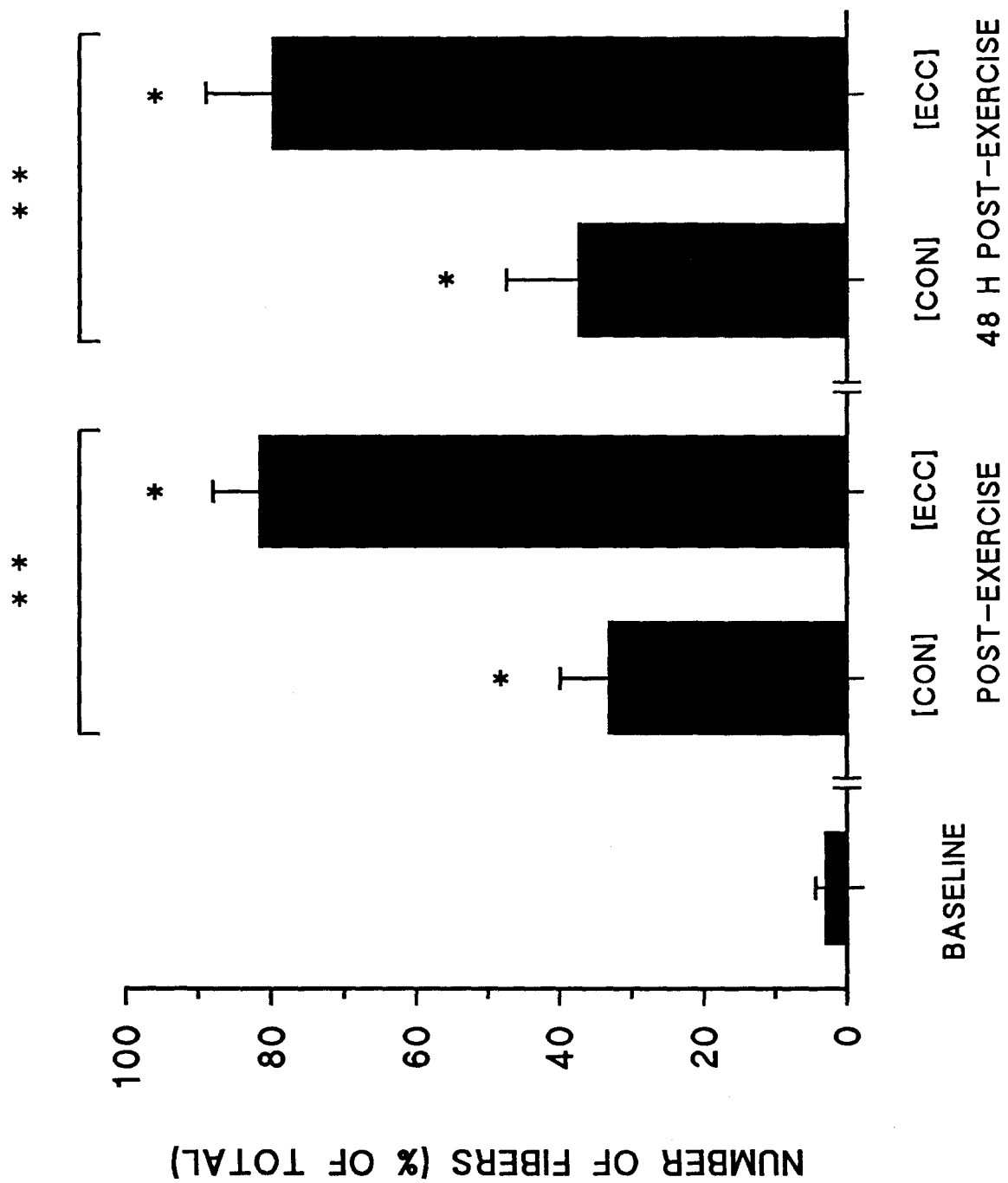

NUMBER OF FIBERS (% OF TOTAL)
100
80
60
40
20
0
BASELINE
[CON]
[ECC]
POST-EXERCISE
[CON]
[ECC]
48 H POST-EXERCISE
* *
* *
*
*
*
*

Figure 10. Number of disrupted fibers within each category observed at each biopsy sampling time (expressed as percent of total; mean ± SE).

* $P \leq 0.05$ between arms at same biopsy time.

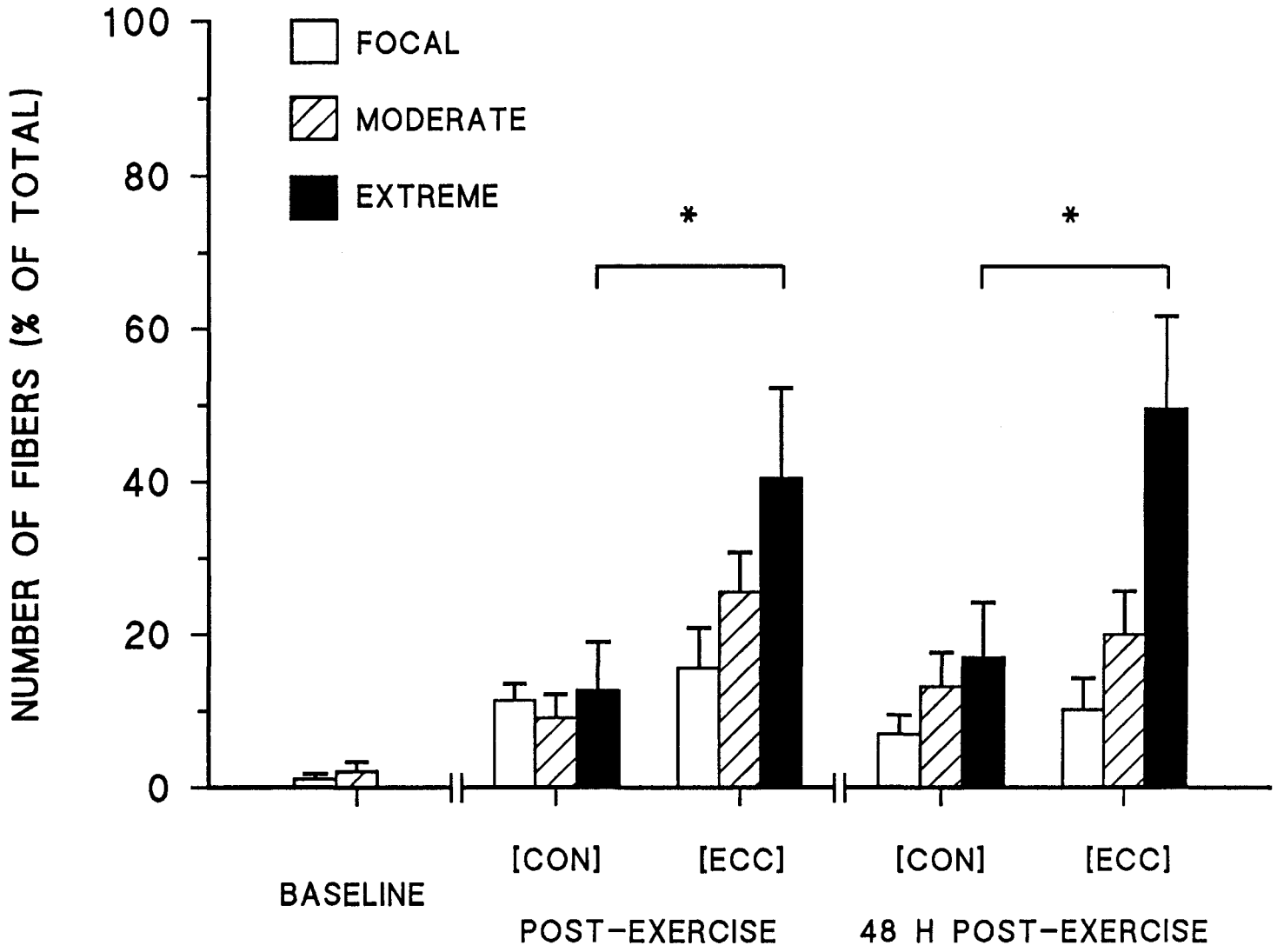

100
80
60
40
20
0
NUMBER OF FIBERS (% OF TOTAL)
FOCAL
MODERATE
EXTREME
*
*
[CON]
[ECC]
[CON]
[ECC]
BASELINE
POST-EXERCISE
48 H POST-EXERCISE

Figure 11. Relation between isometric peak torque (percent of initial) at 48 h in the ECC arm and percentage of fibers showing "extreme" disruption in the 48H-ECC biopsy samples.

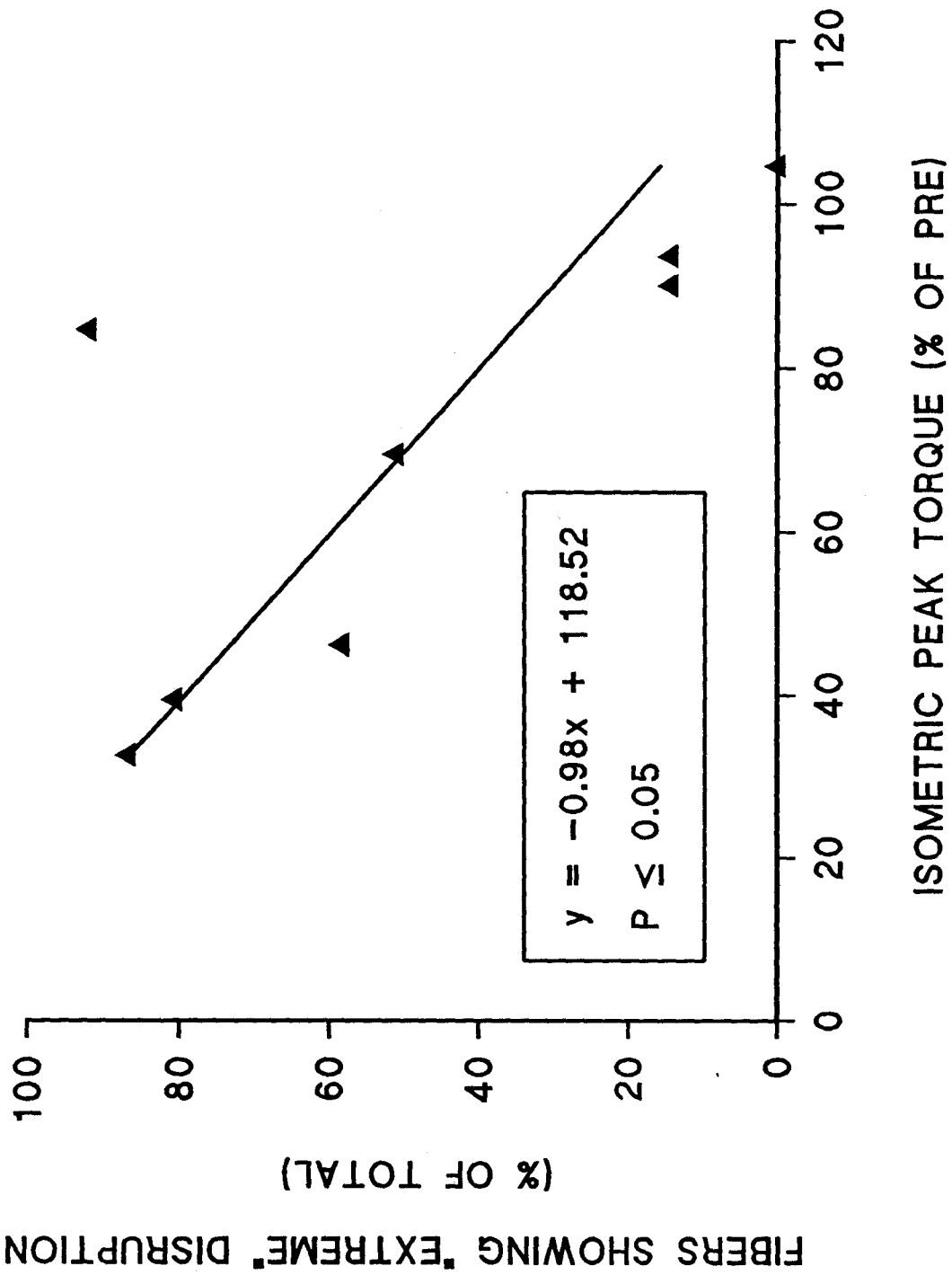
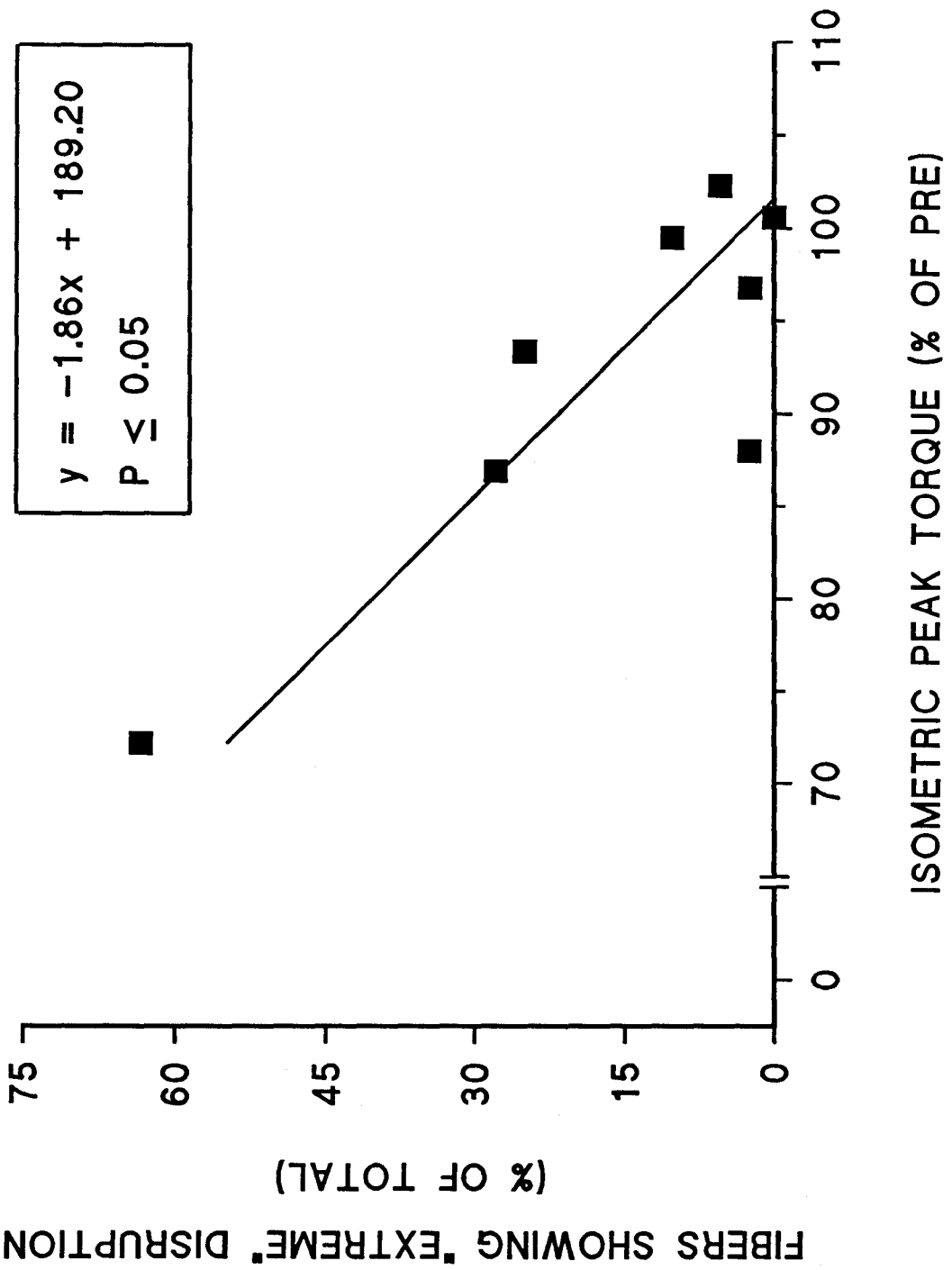

FIBERS SHOWING "EXTREME" DISRUPTION
(% OF TOTAL)
100
80
60
40
20
0
y = −0.98x + 118.52
P ≤ 0.05
0
20
40
60
80
100
120
ISOMETRIC PEAK TORQUE (% OF PRE)

Figure 12. Relation between isometric peak torque (percent of initial) at 48 h in the CON arm and percentage of fibers showing "extreme" disruption in the 48H-CON biopsy samples.


y = −1.86x + 189.20
P ≤ 0.05
FIBERS SHOWING "EXTREME" DISRUPTION
(% OF TOTAL)
75
60
45
30
15
0
0
70
80
90
100
110
ISOMETRIC PEAK TORQUE (% OF PRE)

2.4 **DISCUSSION**

Ultrastructural evidence of skeletal muscle disruption has been observed following a variety of eccentrically-biased continuous exercise protocols (Fridén et al., 1981, 1983b, 1988; Hikida et al., 1983; Newham et al., 1983). Tissue disruption has also been reported following eccentric resistance exercise (Stauber et al., 1990), but the exercise protocol in that study was unlike those typically performed during resistance training. A scarcity of data exists regarding muscle fiber disruption following traditional (6-10 RM) heavy resistance strength training. Staron et al. (1992) have provided the only direct evidence of myofibrillar disruption (e.g., Z band streaming) following resistance training in humans. Animal investigations are rare (due to the difficulty in simulating the activity), but Giddings et al. (1985) observed muscle fiber necrosis and regeneration in cats trained to perform a concentric weight-lifting exercise for food reward. The present investigation is the first to quantitatively assess myofibrillar disruption immediately following an acute bout of heavy resistance exercises in humans.

The major finding of this study was that a single session of elbow flexor resistance exercise produced extensive myofibrillar disruption of the biceps brachii. We attempted to examine the two distinct phases of the arm curling motion

by having subjects lift the weight with one arm [concentric (CON) muscle action], and lower the weight with the other [eccentric (ECC) muscle action]. An electro-goniometer device was strapped to each arm during the exercise bout, in an effort to maintain limb velocity constant during each phase of the movement. We recognize, however, that controlling limb velocity does not necessarily control internal velocity of changes in individual sarcomere length throughout the fiber (Gordon et al., 1966; Lieber and Baskin, 1983).

Our results indicate that both phases resulted in substantial myofibrillar disruption. Needle biopsies taken immediately post-exercise revealed that significantly more fibers were disrupted in both the concentrically- (POST-CON) and eccentrically-exercised arms (POST-ECC) compared to pre-exercise baseline samples (BASE). A greater number of fibers contained evidence of disruption in the POST-ECC (81.7%) compared to the POST-CON samples (33.2%), confirming a previous observation (Newham et al., 1983a) that ECC muscle action results in greater tissue disruption than CON action. In addition, the disruption was more severe in the eccentrically-exercised arm (40.6% of fibers examined in the POST-ECC samples received an "extreme" disruption rating, compared to only 12.8% in the POST-CON samples).

The extent of fiber disruption observed in the eccentrically-exercised arm in this study was greater than that reported in muscles following continuous dynamic ECC

exercise. Fridén et al. (1983b) reported disturbances affecting 32% of fibers 1 h after a bout of ECC cycle ergometry, while Newham et al. (1983a) observed abnormalities in 40% of fibers examined following ECC bench stepping. The brief, forceful actions characteristic of heavy strength training generate higher tensions within the fibers than repeated, low-resistance actions performed during continuous forms of exercise. [It should be noted that both Fridén (1983b) and Newham (1983a) quantified the extent of fiber disruption using light microscopy, while all analyses in the present investigation were conducted with an electron microscope. It is questionable whether or not the magnification properties of a light microscope are sufficient to detect focal sites of disruption, and thus these studies may have underestimated the actual number of disrupted fibers.].

Direct evidence of myofibrillar disruption in humans following CON exercise has not been previously reported. Newham et al., (1983a) have provided the only other comparison of myofibrillar disruption following CON and ECC muscle action. Following a 20 min stepping exercise, they reported morphological abnormalities in the eccentrically-exercised leg, but no evidence of fiber disruption in the concentrically-exercised leg. It is evident from animal investigations, however, that high tensions generated during CON resistance exercise are sufficient to cause tissue

disruption (Giddings et al., 1985). Accordingly, we attribute the fiber disruption observed in the concentrically-exercised arm to high tensions generated during the CON phase of the movement. An alternative explanation for the disruption observed in the concentrically-exercised arm is that a slight ECC action of the biceps may have occurred when the attendant placed the weight in the palm of the (fully extended) arm. Newham and Jones (1988) observed that force production is reduced to a greater extent when maximal ECC actions of the elbow flexors are performed at long rather than short muscle lengths, ostensibly due to greater fiber disruption. It seems unlikely, however, that such a brief ECC action could disrupt over 30% of the fibers examined.

Contrary to previous investigations which utilized continuous exercise protocols, we did not observe evidence of progressive fiber disruption during the post-exercise period. Biopsies taken 48 h post-exercise from the concentrically- (48H-CON) and eccentrically-exercised (48H-ECC) arms showed virtually the same number of disrupted fibers as in the POST-CON and POST-ECC samples, respectively. Fridén et al. (1983b), however, observed a greater number of fibers were disrupted 3 d compared to 1 h following a bout of ECC cycle ergometry. [In that study, the biopsies 3 d post-exercise were taken from different subjects than those taken 1 h post-exercise. Since large variability exists between subjects regarding the extent of tissue disruption following exercise,

valid comparisons should only be conducted using repeated samples from the same subjects.] Newham et al. (1983a) also reported that a greater number of fibers were disrupted, and to a greater extent, 24-48 h following a bout of ECC bench stepping compared to immediately after the bout. These authors suggested the initial "damage" was mechanically induced, and the "the exacerbation of damage with time could be due to mechanical or chemical factors" (Newham et al., 1983a). Fridén and Lieber (1992) postulated that "following initial injury, further degradation (of the fiber) takes place during the ensuing hours. This process in likely to be initiated by ... activation of the calcium-activated neutral proteases, lysosomal proteases, and other cellular processes that are calcium-mediated." Following other types of injury, the regeneration process appears to be confined to the damaged fiber (Schultz et al., 1986). It remains unknown however, whether the activation of calcium-sensitive enzymes is also confined to the disrupted fiber, or whether *additional* fibers are affected by this process. An alternative possibility is that Fridén et al. (1983b) and Newham et al. (1983a) simply missed focal sites of injury present immediately after the exercise bout (since they quantified damage with the light microscope). Later in the post-exercise period, degradative processes may have enlarged the initial focal areas, and made them more apparent at light microscope magnification.

In addition to myofibrillar disruption, we also observed other fiber abnormalities (e.g., central nuclei; Figure 13), as well as evidence of fiber regeneration (e.g., "satellite-like" cells; Figure 14). The frequency with which these occurred were not examined. The satellite-like cells which we observed were similar to those reported by Giddings et al. (1985) following prolonged weight lifting in the cat. These cells resemble satellite cells, but also appear to contain myofilaments within the surrounding basal lamina.

The voluntary and electrically evoked strength data support the ultrastructural evidence that the ECC phase of the exercise resulted in the greatest fiber disruption. Isometric peak torque, low-velocity concentric peak torque and peak twitch torque remained depressed through 96 h post-exercise, and high-velocity concentric peak torque through 72 h post-exercise, in the ECC arm. All 4 measurements recovered to baseline levels by 24 h in the CON arm. It is not clear why strength recovered faster in the ECC arm at the higher contraction velocity. It has previously been reported that the rate of strength restoration is slowest at the highest angular velocities, ostensibly due to greater Type II fiber disruption (Fridén et al., 1983b). The "high" velocity strength test in the present investigation, however, was performed at 3.14 $rad \cdot s^{-1}$, which is well below the physiological Vmax of the elbow flexors. It is therefore possible that our test was not sensitive enough to disriminate

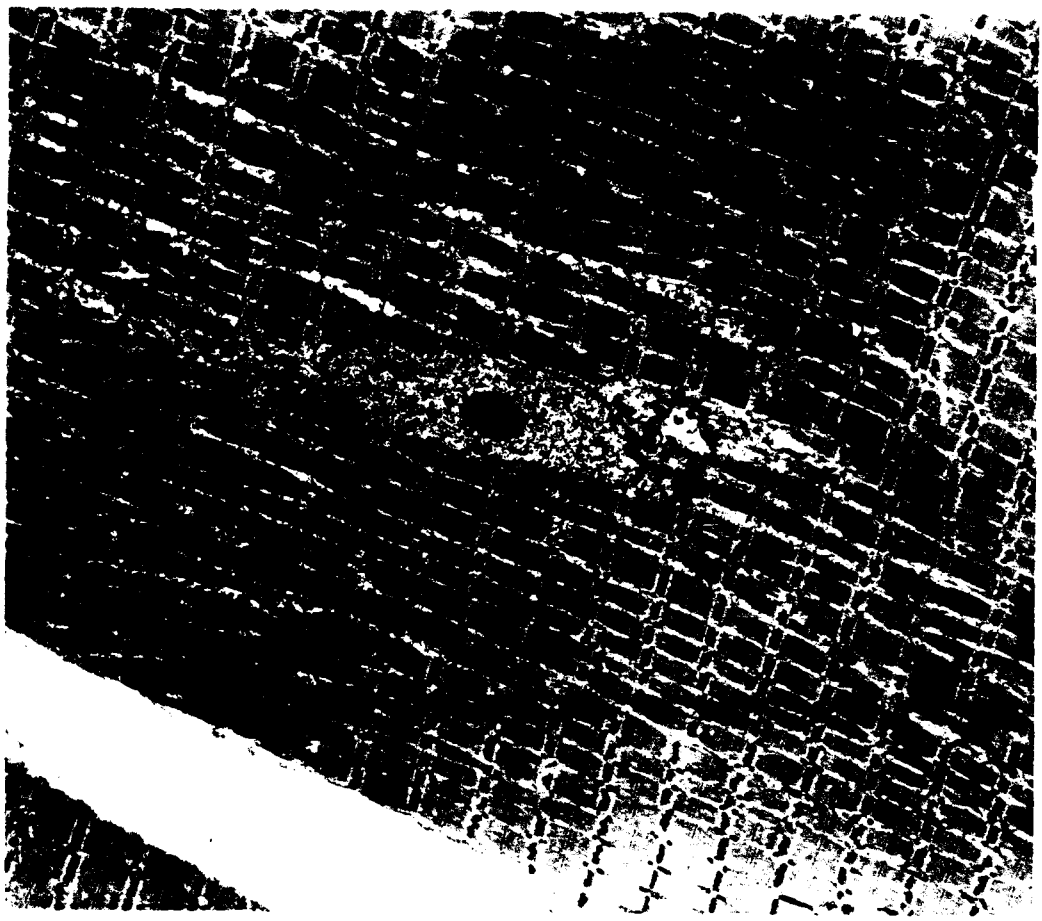


Figure 13. Micrograph of central nucleus (x720).

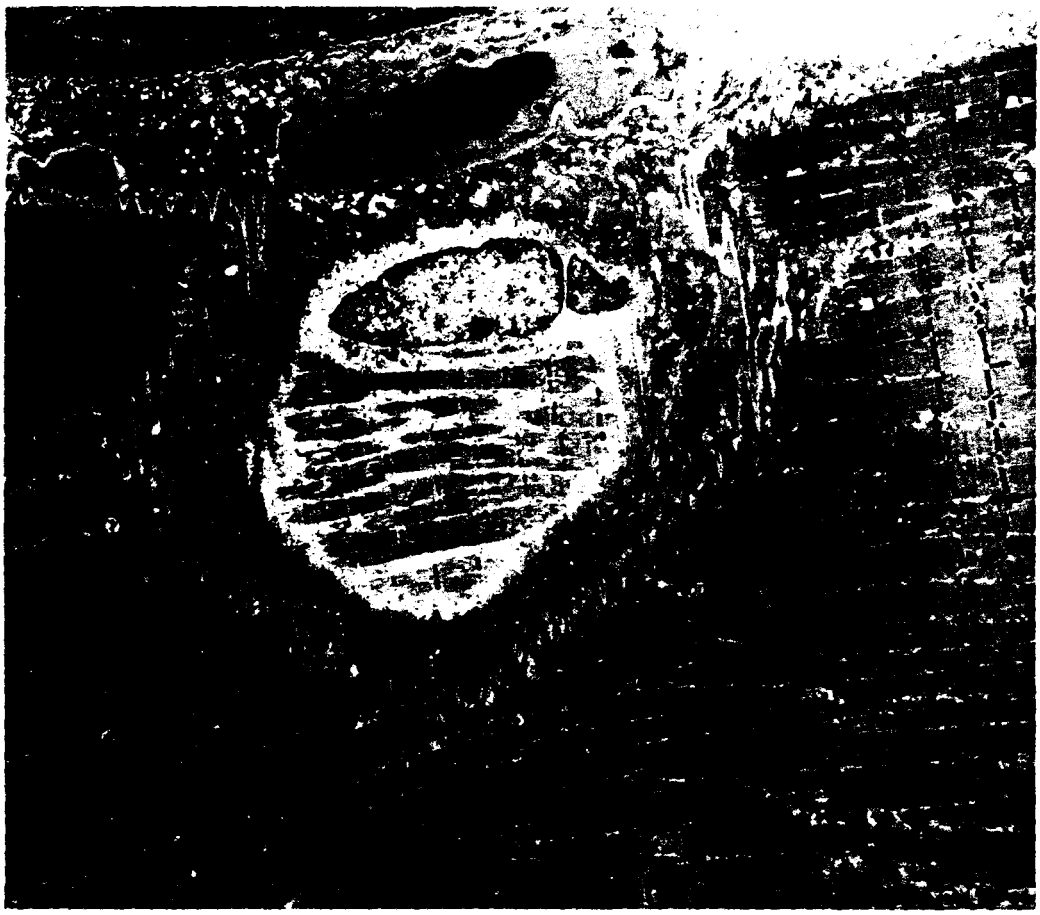


Figure 14. Micrograph of "satellite-like" cell (x550).

between differences in force generation due to specific fiber type involvement.

Prolonged decreases in maximal isometric force generation have been reported previously following eccentrically-biased elbow flexor resistance exercise (Clarkson and Tremblay, 1988; Donnelly et al., 1992; Ebbelling and Clarkson, 1990; Newham et al., 1987; Nosaka et al., 1991), ranging from 45-75% (compared to initial values) 4 d post-exercise. It is important to note, however, that the exercise protocols utilized in these studies were extreme and not typically employed in resistance training programs. Maximal isometric force generation in the present investigation recovered to 85% of baseline levels within 4 d post-exercise.

Our findings suggest that the prolonged decrease in force generation in the eccentrically-exercised arm was due to extensive myofibrillar disruption. The percentage of fibers which showed "extreme" disruption in the 48H-ECC samples significantly correlated with the percent decline in MVC torque at 48 h in the ECC arm ($r = -0.745$, $r^2 = 0.556$, $P \leq 0.05$). A relation between fiber disruption and force generation also existed in the concentrically-exercised arm: the percentage of fibers which showed "extreme" disruption in the 48H-CON samples significantly correlated with the percent decline in MVC torque at 48 h in the CON arm ($r = -0.864$, $r^2 = 0.746$, $P \leq 0.05$). It is recognized that a correlation between 2 variables does not necessarily imply a cause and

effect relationship, but the finding of significant correlations for both arms lends credibility to the hypothesis that strength decrements were related to disruptions in the contractile apparatus.

It is not clear why the strength and contractile measurements were not depressed longer in the concentrically-exercised arm, since approximately one third of the fibers examined in both the POST-CON and 48H-CON samples contained evidence of disruption. The fibers in the concentrically-exercised arm samples were not as severely disrupted as in the POST-ECC and 48H-ECC samples, though. Only 12.8% and 17.1% of the fibers showed "extreme" disruption in the POST-CON and 48H-CON samples, respectively. Significantly more fibers showed "extreme" disruption in the POST-ECC (40.6%) and 48H-ECC (49.7%) samples compared to the those taken from the concentrically-exercised arm. There was a main effect for arm when the post-exercise data was collapsed across measurement time.

We recognize that the prolonged decrease in force production in the ECC arm cannot be solely attributed to myofibrillar disruption. All strength measurements had partially recovered by 48 h post-exercise, despite the fact that the extent of fiber disruption observed at 48 h post-exercise was similar to that seen immediately after the bout. Other potential sites of injury include the sarcolemma, sarcoplasmic reticulum (SR), basal lamina, and surrounding

connective tissue. Sarcomere length is known to vary within active fibers during eccentric muscle action (Colomo et al., 1988), and it is possible that severe lengthening of some sarcomeres could disrupt normal Ca^{2+} cycling by damaging the sarcolemma or SR system (Armstrong, 1990; Armstrong et al., 1991). The inability of the cell to properly sequester Ca^{2+} could cause the fiber to enter a rigor-like state, allowing "shear forces" to develop between fibers (Stauber, 1989). Alternatively, increases in intracellular [Ca^{2+}] could stimulate Ca^{2+}-sensitive enzymes (e.g., proteases, phospholipases) which are active in fiber degeneration (Byrd, 1992). Data from the present investigation, however, do not indicate that the prolonged loss of force was due to disruption of the sarcolemma or SR. We did not observe a prolonged decrease in the ratio of peak twitch torque to voluntary isometric peak torque (PTT/MVC ratio). If the decrease in force production was due to sarcolemmal or SR disruption, we would expect to see a greater decrease in PTT (twitch contraction) compared to MVC (tetanic contraction). Our data therefore appear to rule out disruption of normal excitation-contraction coupling as the reason for the prolonged strength loss.

In addition, the areas of disruption which we observed contained Z bands that were out of alignment, but not "smeared". Z band streaming has been attributed to degradation of the Z disc components by Ca2+-activated

proteases (Busch et al., 1972; Ishiura et al., 1980). Our data would therefore suggest that the disruptions observed were not due to an inability of the fiber to properly sequester Ca^{2+}. These data also rule out the possibility that the disruption may have been due to oxygen free radical attack on the membranous systems of the fiber.

All subjects in the present study reported delayed muscle soreness only in the arm which had acted eccentrically. A specific rating scale was not employed in the study, however, since the results of such a test would have been confounded by the biopsy procedure. It is unlikely that the decrease in force generation observed in the eccentrically-exercised arm was due to the perception of pain. The prolonged depression in peak twitch torque suggests the force decline was due to contractile tissue disruption, since electrically evoked contractions "bypass" the central nervous system (CNS). The CNS may have played a role in suppressing strength immediately after the bout in both arms, however, since motor unit activation (MUA) showed a main effect for measurement time. Post hoc analysis revealed MUA decreased ($P \leq 0.05$) immediately post-exercise, but did not differ from baseline levels at any other measurement time.

The exact cause of myofibrillar disruption following resistance exercise remains unknown, but appears to be associated with the generation of high tensions within active fibers. Most investigators have focused on the mechanical

properties of ECC muscle action to explain tissue disruption, but the results of the present study indicate that disruption can occur following CON muscle action as well. The areas of myofibrillar disruption which we observed were random across the fiber, and the areas frequently interrupted the normal transverse alignment of sarcomeres. This pattern may be due to disruption of the myofibrillar cytoskeleton, particularly the inter Z disc to Z disc connections made of the intermediate filament desmin (Waterman-Storer, 1991). Direct evidence of cytoskeletal disruption in eccentrically-exercised human muscle has been reported previously (Fridén et al., 1984). Disruption of the normal myofibrillar banding pattern could also result from damage to the elastic protein titin, which is responsible for keeping the thick filaments centered between the Z discs during active force generation (Funatsu et al., 1990; Horowits et al., 1986).

REFERENCES

Abbott, B.C., B. Bigland, and J.M. Ritchie. The physiological cost of negative work. J. Physiol. 117:380-390, 1952.

Alberts, B., D. Bray, J. Lewis, M. Raff, K. Roberts, and J.D. Watson. Molecular Biology of the Cell (2nd ed.). New York, NY: Garland Publishing, 1989, p. 802-824.

Armstrong, R.B., Mechanisms of exercise-induced delayed onset muscle soreness: a brief review. Med. Sci. Sports Exerc. 16:529-538, 1984.

Armstrong, R.B., Initial events in exercise-induced muscular injury. Med. Sci. Sports Exerc. 22:429-435, 1990.

Armstrong, R.B., M.H. Laughlin, L. Rome, and C.R. Taylor. Metabolism of rats running up and down an incline. J. Appl. Physiol. 55:518-521, 1983a.

Armstrong, R.B., R.W. Ogilvie, and J.A. Schwayne. Eccentric exercise-induced injury to rat skeletal muscle. J. Appl. Physiol. 54:80-93, 1983b.

Armstrong, R.B., G.L. Warren, and J.A. Warren. Mechanisms of exercise-induced muscle fiber injury. Sports Med. 12:184-207, 1991.

Asmussen, E. Observations on experimental muscular soreness. Acta Rhum. Scand. 2:109-116, 1956.

Asmussen, E. Positive and negative muscular work. Acta Physiol. Scand. 28:364-382, 1953.

Basmajian, J.V. Muscles Alive: Their Functions Revealed By Electromyography (4th ed.). Baltimore, MD: Williams & Wilkins, 1978.

Belanger, A.Y., and A.J. McComas. Extent of motor unit activation during effort. J. Appl. Physiol. 51:160-167, 1981.

Bergstrom, C.J. Muscle electrolytes in man. Scand. J. Clin. Lab. Invest. Suppl. 68: 1-110, 1962.

Bigland-Ritchie, B. and J.J. Woods. Integrated electromyogram and oxygen uptake during positive and negative work. J. Physiol. 260:267-277, 1976.

Billeter, R., and H. Hoppeler. Muscular Basis of Strength. In: Strength and Power in Sport, edited by P.V. Komi. Oxford: Blackwell Scientific Publications, 1992, pp. 39-63.

Bodine, S.C., R.R. Roy, E. Eldred, and V.R. Edgerton. Maximal force as a function of anatomical features of motor units in the cat tibialis anterior. J. Neurophysiol. 57:1730-1745, 1987.

Burke, R.E. Motor units: anatomy, physiology, and functional organization. In: Handbook of Physiology. Section I, The Nervous System II, edited by V.B.Brooke. Washington, DC: American Physiological Society, 1981, p. 345-422.

Busch, W.A., M.H. Stromer, D.E. Goll, and A. Suzuki. A Ca^{2+}-specific removal of Z lines from rabbit skeletal muscle. J. Cell Biol. 52:367-381, 1972.

Byrd, S.K. Alterations in the sarcoplasmic reticulum: a possible link to exercise-induced muscle damage. Med. Sci. Sports Exerc. 24:531-536, 1992.

Byrd, S.K., A.K. Bode, and G.A. Klug. Effects of exercise of varying duration on sarcoplasmic reticulum function. J. Appl. Physiol. 66:1383-1389, 1989a.

Byrd, S.K., L.J. McCutcheon, D.R. Hodgson, and P.G. Gollnick. Altered sarcoplasmic reticulum function after high-intensity exercise. J. Appl. Physiol. 67:2072-2077, 1989b.

Byrnes, W.C., P.M. Clarkson, J.S. White, S.S. Hsieh, and P.N. Frykman. Delayed onset muscle soreness following repeated bouts of downhill running. J. Appl. Physiol. 59:710-715, 1985.

Carlson, B.M., and J.A. Faulkner. The regeneration of skeletal muscle fibers following injury: a review. Med. Sci. Sports Exerc. 15:187-198, 1983.

Chapman, A.E. The mechanical properties of human muscle. Exerc. Sports Sci. Rev. 13:443-501, 1985.

Clarkson, P.M., W.C. Barnes, K.M. McCormick, L.P. Turcotte, and J.S. White. Muscle soreness and serum creatine kinase activity following isometric, eccentric and concentric exercise. Int. J. Sports Med. 7:152-155, 1986.

Clarkson, P.M., and I. Tremblay. Exercise-induced muscle damage, repair, and adaptation in humans. J. Appl. Physiol. 65:1-6, 1988.

Clarkson, P.M., K. Nosaka, and B. Braun. Muscle function after exercise-induced muscle damage and rapid adaptation. Med. Sci. Sports Exerc. 24:512-520, 1992.

Colliander, E.B., and P.A. Tesch. Effects of eccentric and concentric muscle actions in resistance training. Acta. Phys. Scand. 140:31-39, 1990.

Colomo, F., V. Lambardi, and G. Piazzesi. The mechanisms of force enhancement during constant velocity lengthening in tetanized single fibres of frog muscle. Adv. Exper. Med. Biol. 226:489-502, 1986.

Cooke, P. A periodic cytoskeletal lattice in striated muscle. In: Cell and Muscle Motility, Vol. 6, edited by J.W. Shay. New York: Plenum Press, 1985, pp. 287-313.

Darr, K.C., and E. Schultz. Exercise-induced satellite cell activation in growing and mature skeletal muscle. J. Appl. Physiol. 63:1816-1821, 1987.

Davies, C.T.M., and M.J. White. Muscle weakness following eccentric work in man. Pflügers Arch. 392:168-171, 1981.

Donnelly, A.E., P.M. Clarkson, and R.J. Maughan. Exercise-induced muscle damage: effects of light exercise on damaged muscle. Eur. J. Appl. Physiol. 64:350-353, 1992.

Dudley, G.A., P.A. Tesch, B.J. Miller, and P. Buchanan. Importance of eccentric actions in performance adaptations to resistance training. Aviat. Space Environ. Med. 62:543-550, 1991.

Duncan, C.J. Role of calcium in triggering rapid ultrastructural damage in muscle: a study with chemically skinned fibres. J. Cell Sci. 87:581-594, 1987.

Ebbeling, C.B., and P.M. Clarkson. Exercise-induced muscle damage and adaptation. Sports Med. 7:207-234, 1989.

Ebbeling, C.B., and P.M. Clarkson. Muscle adaptation prior to recovery following eccentric exercise. Eur. J. Appl. Physiol. 60:26-31, 1990.

Eisenberg, B.R. Quantitative ultrastructure of mammalian skeletal muscle. In: Handbook of Physiology, Section 10, Skeletal Muscle, edited by L.D. Peachey, R.H. Adrian, and S.R. Geiger. Baltimore, MD: Waverly Press, 1983, p.73-112.

Evans, W.J., and J.G. Cannon. The metabolic effects of exercise-induced muscle damage. In: Exercise and Sport Sciences Reviews, edited by J. Holloszy. Baltimore, MD: Williams and Wilkens, 1991, vol. 19, p. 99-125.

Evans, W.J., S.D. Pinney, and V.R. Young. Suction applied to a muscle biopsy maximizes sample size. Med. Sci. Sports Exerc. 14:101, 1982.

Fridén. J. Changes in human skeletal muscle induced by long term eccentric exercise. Cell Tissue Res. 236:365-372, 1984a.

Fridén, J., Muscle soreness after exercise: Implications of morphological changes. Int. J. Sports Med. 5:57-66, 1984b.

Fridén, J., and R.L. Lieber. Structural and mechanical basis of exercise-induced muscle injury. Med. Sci. Sports Exerc. 24:521-530, 1992.

Fridén, J., U. Kjörell, and L.E. Thornell. Delayed muscle soreness and cytoskeletal alterations: an immunocytological study in man. Int. J. Sports Med. 5:15-18, 1984.

Fridén, J., J. Seger, and B. Ekbolm. Sublethal muscle fibre injuries after high-tension anaerobic exercise. Eur. J. Appl. Physiol. 57:360-368, 1988.

Fridén, J., J. Seger, M. Sjöström, and B. Ekbolm. Adaptive response in human skeletal muscle subjected to prolonged eccentric training. Int. J. Sports Med. 4:177-183, 1983a.

Fridén, J., M. Sjöström, and B. Ekbolm. A morphological study of delayed muscle soreness. Experimentia. 37:506-507, 1981.

Fridén. J., M. Sjöström, and B. Ekbolm. Myofibrillar damage following intense eccentric exercise in man. Int. J. Sports Med. 4:170-176, 1983b.

Fritz, V.K., and W.T. Stauber. Characterization of muscles injured by forced lengthening. II. Proteoglycans. Med. Sci. Sports Exerc. 20:354-361, 1988.

Funatso, T., H. Higuchi, and S. Ishiwata. Elastic filaments in skeletal muscle revealed by selective removal of thin filaments with plasma gelsolin. J. Cell Biol. 110:53-62, 1990.

Fürst, D.O., M. Osborn, R. Nave and K. Weber. The organization of titin filaments in the half sarcomere revealed by monoclonal antibodies in immunoelectron microscopy: a map of ten nonrepetitive epitopes starting at the Z-line extends close to the M-line. J. Cell Biol. 106:1563-1572, 1988.

Giddings, C.J., W.B. Neaves, and W.J. Gonyea. Muscle fiber necrosis and regeneration induced by prolonged weight-lifting exercise in the cat. Anat. Rec. 211:133-41, 1985.

Goldspink, G. Ultrastructural changes in striated muscle fibres during contraction and growth with particular reference to the mechanism of myofibril splitting. J. Cell Sci. 9:123-138, 1971.

Goldspink, G. Cellular and molecular aspects of adaptation in skeletal muscle. In: Strength and Power in Sport, edited by P.V. Komi. Oxford: Blackwell Scientific Publications, 1992, pp. 211-229.

Gordon, A.M., A.F. Huxley, and F.J. Julian. The variation in isometric tension with sarcomere length in vertebrate muscle fibers. J. Physiol. 184:170-192, 1966.

Hather, B.M., P.A. Tesch, P. Buchanan, and G.A. Dudley. Influence of eccentric actions on skeletal muscle adaptations to resistance training. Acta. Physiol. Scand. 143:177-185, 1991.

Hikida, R.S., R.S. Staron, F.C. Hagerman, M. Leonardi, R. Gilders, J. Falkel, T. Murray, and K. Appell. Serum creatine kinase activity and its changes after a muscle biopsy. Clin. Physiol. 11:51-59, 1991.

Horowits, R., and R.J. Podolsky. Transitional stability of thick filaments in activated skeletal muscle depends on sarcomere length: evidence for the role of titin filaments. J. Cell Biol. 105:2217-2223, 1987.

Horowits, R., E. Kempner, M. Bisher, and R. Podolsky. A physiological role for titin and nebulin in skeletal muscle. Nature. 323:160-164, 1986.

Ishiura, S., H. Sugita, I. Nonaka, and K. Imahori. Calcium-activated neutral protease: its localization in the myofibril, especially at the Z band. J. Biochem. 87:343-346, 1980.

Jenkins, R.R. Free radical chemistry. Sports Med. 5:156-170, 1988.

Jones, D.A., D.J. Newham, J.M. Round, and S.E.J. Tolfree. Experimental human muscle damage: morphological changes in relation to other indices of damage. J. Physiol. 375:435-448, 1986.

Knappeis, G.G., and F. Carlson. The ultrastructure of the Z disc in skeletal muscle. J. Cell Biol. 13:323-335, 1962.

Knuttgen, H.G., and P.V. Komi. Basic definitions for exercise. In: Strength and Power in Sport, edited by P.V. Komi. Oxford: Blackwell Scientific Publications, 1992, p. 4.

Komi, P.V., and E.R. Buskirk. Effect of eccentric and concentric muscle conditioning on tension and electrical activity of human muscle. Ergonomics. 15:417-434, 1972.

Lieber, R.L., and R.J. Baskin. Intersarcomere dynamics of single skeletal muscle fibers during fixed-end tetani. J. Gen. Physiol. 82:347-364, 1983.

Lieber, R.L., and J. Fridén. Selective damage of fast glycolytic muscle fibres with eccentric contractions of the rabbit tibialis anterior. Acta Physiol. Scand. 133:587-588, 1988.

Manfredi, T.G., R.A. Fielding, K.P. O'Reilly, C.N. Meredith, H.Y. Lee, and W.J. Evans. Plasma creatine kinase activity and exercise-induced muscle damage in older men. Med. Sci. Sports Exerc. 23:1028-1034, 1991.

Maruyama, K., T. Yoshioka, H. Higuchi, K. Ohashi, S. Kimura, and R. Natori. Connectin filaments link thick filaments and Z-lines in frog skeletal muscle as revealed by immunoelectron microscopy. J. Cell Biol. 101:2167-2172, 1985.

Maruyama, K., A. Matsuno, H. Higuchi, S. Shimaoka, S. Kimura, and T. Shimizu. Behaviour of connectin (titin) and nebulin in skinned muscle fibers released after extreme stretch as revealed by immunoelectron microscopy. J. Musc. Res. Cell Motil. 10:350-359, 1989.

Maughen, R.J., A.E. Donnelly, M. Gleeson, P.H. Whiting, K.A. Walker, and P.J. Clough. Delayed-onset muscle damage and lipid peroxidation in man after a downhill run. Musc. Nerve. 12:332-336, 1989.

McCartney, N., D. Moroz, S.H. Garner, and A.J. McComas. The effects of strength training in patients with selected neuromuscular disorders. Med. Sci. Sports Exerc. 20: 362-368.

Meltzer, H.Y., R.W. Kuncl, J. Click, and V. Yang. Incidence of Z band streaming and myofibrillar disruptions in skeletal muscle from healthy young people. Neurology. 26:853-857, 1976.

Miller, A.E.J., J.D. MacDougall, M.A. Tarnopolsky, and D.G. Sale. Gender differences in strength and muscle fiber characterisitics. Eur. J. Appl. Physiol. 66:254-262, 1993.

Nadel, E.R., U. Bergh, and B. Saltin. Body temperature during negative work. J. Appl. Physiol. 33:553-558, 1972.

Newham, D.J., and D.A. Jones. Muscle fatigue and pain after eccentric contractions at long and short lengths. Clin. Sci. 74:553-557, 1988.

Newham, D.J., D.A. Jones, and P.M. Clarkson. Repeated high-force eccentric exercise: effects on muscle pain and damage. J. Appl. Physiol. 63:1381-1386, 1987.

Newham, D.J. G. McPhail, K.R. Mills, and R.H.T. Edwards. Ultrastructural changes after concentric and eccentric contractions of human muscle. J. Neurol. Sci. 61:109-122, 1983a.

Newham, D.J., K.R. Mills, B.M. Quigley, and R.H.T. Edwards. Pain and fatigue after concentric and eccentric muscle contractions. Clin. Sci. 64:55-62, 1983b.

Noakes, T.D., Effect of exercise on serum enzyme activities in humans. Sports Med. 4:245-267, 1987.

Nosaka, K., P.M. Clarkson, M.E. McGuiggin, and J.M. Byrne. Time course of muscle adaptation after high force eccentric exercise. Eur. J. Appl. Physiol. 63:70-76, 1991.

Ogilvie, R.W., R.B. Armstrong, K.E. Baird, and C.L. Bottoms. Lesions in the rat soleus muscle following eccentrically biased exercise. Am. J. Anat. 182:335-346, 1988.

O'Hagan, F.T., N. Tsunoda, D.G. Sale, and J.D. MacDougall. Elbow flexion evoked contractile properties in untrained men and women and male bodybuilders. Eur. J. Appl. Physiol. 66:240-245, 1993.

O'Reilly, K.P., M.J. Warhol, R.A. Fielding, W.F. Frontera, C.N. Meredith, and W.J. Evans. Eccentric exercise-induced muscle damage impairs muscle glycogen repletion. J. Appl. Physiol. 63:252-256, 1987.

Paul, G.L., J.P. Delany, J.T. Snook, J.G. Seifert, and T.E. Kirby. Serum and urinary markers of skeletal muscle tissue damage after weight lifting exercise. Eur. J. Appl. Physiol. 58:786-790, 1989.

Pollack, G.H. Muscles and Molecules: Uncovering the Principles of Biological Motion. Seattle, WA: Ebner & Sons Publishers, 1990, pp. 67-69.

Reske-Nielsen, E., and A. Harmsen. Electronmicroscopical study of muscle biopsies from healthy young people. Acta. Path. Microb. Scand. 80:449-467, 1972.

Sale, D.G. Testing strength and power. In: Physiological Testing of the High-Performance Athlete (2nd ed.), edited by J.D. MacDougall, H.A. Wenger, and H.J. Green. Champaign, IL: Human Kinetics, 1991, p. 21-108.

Sale, D.G., J.D. MacDougall, S.E. Alway, and J.R. Sutton. Voluntary strength and muscle characteristics in untrained men and women and in male bodybuilders. J. Appl. Physiol. 62: 1786-1793, 1987.

Schantz, P., E. Randall-Fox, W. Hutchinson, A. Tydén, and P.O. Åstrand. Muscle fibre type distribution, muscle cross-sectional area and maximal voluntary strength in humans. Acta Physiol. Scand. 117:219-226, 1983.

Schultz, E. Satellite cell behaviour during skeletal muscle growth and regeneration. Med. Sci. Sports Exerc. 21:S181-S186, 1989.

Schultz, E., D.L. Jaryszak, M.C. Gibson, and D.J. Albright. Absence of exogenous satellite cell contribution to regeneration of frozen skeletal muscle. J. Muscle Res. Cell Motil. 7:361-367, 1986.

Schwane, J.A., S.R. Johnson, C.B. Vandenakker, and R.B. Armstrong. Delayed-onset muscular soreness and plasma CPK and LDH activities after downhill running. Med. Sci. Sports Exerc. 15:51-56, 1983.

Sheterline, P. Mechanisms of Cell Motility: Molecular Aspects of Contractility. London: Academic Press, 1983, pp. 120-126.

Sjödin, B., Y.H. Westing, and F.S. Apple. Biochemical mechanisms for oxygen free radical formation during exercise. Sports Med. 10:236-254, 1990.

Staron, R.S., R.S. Hikida, T.F. Murray, M.M. Nelson, P. Johnson, and F. Hagerman. Assessment of skeletal muscle damage in successive biopsies from strength-trained and untrained men and women. Eur. J. Appl. Physiol. 65:258-264, 1992.

Staron, R.S., M.J. Leonardi, D.L. Karapondo, E.S. Malicky, J.E. Falkel, F.C. Hagerman, and R.S. Hikida. Strength and skeletal muscle adaptations in heavy-resistance-trained women after detraining and retraining. J. Appl. Physiol. 70:631-640, 1991.

Staron, R.S., E.S. Malicky, M.J. Leonardi, J.E. Falkel, F.C. Hagerman, and G.A. Dudley. Muscle hypertrophy and fast twitch fiber type conversions in heavy resistance-trained women. Eur. J. Appl. Physiol. 60:71-79, 1989.

Stauber, W.T. Eccentric action of muscles: physiology, injury and adaptation. In: Exercise and Sport Sciences Reviews, edited by K. Pandolf. Baltimore, MD: WIlliams and Wilkins, 1989, vol. 17, p. 157-185.

Stauber, W.T., P.M. Clarkson, V.K. Fritz, and W.J. Evans. Extracellular matrix disruption and pain after eccentric muscle action. J. Appl. Physiol. 69:868-874, 1990.

Tokuyasu, K.T., A.H. Datton, and S.J. Singer. Immunoelectron microscopic studies of desmin (skeletin) localization and intermediate filament organization in chicken skeletal muscle. J. Cell Biol. 96:1727-1735, 1983.

Tidball, J.G., T.L. Daniel. Elastic energy storage in rigored skeletal muscle cells under physiological loading conditions. Am. J. Physiol. 250:R56-R64, 1886.

Tsunoda, N., F.T. O'Hagan, D.G. Sale, and J.D. MacDougall. Elbow flexion strength curves in untrained men and women and male bodybuilders. Eur. J. Appl. Physiol. 66:235-239, 1993.

Wang, K. Sarcomere-associated cytoskeletal lattices in striated muscle: review and hypothesis. In: Cell and Muscle Motility, Vol. 6, edited by J.W. Shay. New York: Plenum Press, 1985, pp. 315-364.

Wang, K., and J. Wright. Architecture of the sarcomere matrix of skeletal muscle: immunoelectron microscopic evidence suggests a set of parallel, inextensible nebulin filaments anchored at the Z-line. J. Cell Biol. 107:2199-2212, 1988.

Waterman-Storer, C.M., The cytoskeleton of skeletal muscle: is it affected by exercise? A brief review. Med. Sci. Sports Exerc. 23:1240-1249, 1991.

Whiting, A., J. Wardale, and J. Trinick. Does titin regulate the length of muscle thick filaments? J. Mol. Biol. 205:263-268, 1989.

Wilkie, D.R. Heat, work, and phosphorylcreatine break-down in muscle. J. Physiol. 195:157-183, 1968.

APPENDIX I: ANOVA SUMMARY TABLES

APPENDIX I-A

ANOVA SUMMARY FOR ISOMETRIC PEAK TORQUE

SOURCE	SS	DF	MS	F	P
BLOCKS/SUBJECTS	13951.870	7			
ARM	4735.693	1	4735.693	7.011	.032
ERROR	4728.129	7	675.447		
TIME	4483.756	5	896.751	11.310	<.001
ERROR	2775.183	35	79.291		
ARM TIME	1313.800	5	262.760	4.915	.001
ERROR	1871.200	35	53.463		
TOTAL	33859.631	95			
(RESIDUAL)	9374.512	77			

	PRE	POS	24H	48H	72H	96H	
	n	n	n	n	n	n	
	74.27	58.80	67.64	68.47	70.46	73.00	
CON							68.77
	10.30	8.04	12.74	10.95	11.88	11.17	
	741.91	452.34	1135.87	838.64	987.15	873.42	
	n	n	n	n	n	n	
	71.96	48.81	46.38	43.84	55.43	61.94	
ECC							54.73
	12.61	19.98	21.56	26.00	21.41	21.36	
	1113.34	2795.43	3254.52	4730.57	3208.14	3195.05	
	73.11	53.80	57.01	56.16	62.94	67.47	

APPENDIX I-B

ANOVA SUMMARY FOR LOW VELOCITY CONCENTRIC PEAK TORQUE

SOURCE	SS	DF	MS	F	P
BLOCKS/SUBJECTS	5061.291	7			
ARM	3337.042	1	3337.042	12.618	.009
ERROR	1851.292	7	264.470		
TIME	1578.833	5	315.767	14.375	<.001
ERROR	768.834	35	21.967		
ARM TIME	876.333	5	175.267	9.889	<.001
ERROR	620.333	35	17.724		
TOTAL	14093.958	95			
(RESIDUAL)	3240.458	77			

	PRE	POS	24H	48H	72H	96H	
	8	8	8	8	8	8	
	48.63	43.25	46.25	49.88	49.25	50.25	
CON							47.92
	4.75	6.61	8.68	7.30	5.99	5.68	
	157.87	305.50	527.50	372.87	251.50	225.50	
	8	8	8	8	8	8	
	48.63	31.63	28.25	31.88	36.50	39.88	
ECC							36.13
	7.39	11.33	11.77	15.18	13.13	14.11	
	381.87	897.87	969.50	1612.87	1206.00	1392.87	
	48.63	37.44	37.25	40.88	42.88	45.06	

APPENDIX I-C

ANOVA SUMMARY FOR HIGH VELOCITY CONCENTRIC PEAK TORQUE

SOURCE	SS	DF	MS	F	P
BLOCKS/SUBJECTS	4151.292	7			
ARM	495.042	1	495.042	6.686	.035
ERROR	518.292	7	74.042		
TIME	916.833	5	183.367	7.828	<.001
ERROR	819.833	35	23.424		
ARM TIME	299.833	5	59.967	5.555	<.001
ERROR	377.833	35	10.795		
TOTAL	7578.958	95			
(RESIDUAL)	1715.958	77			

	PRE	POS	24H	48H	72H	96H	
	8	8	8	8	8	8	
	34.13	29.63	33.25	33.75	35.50	35.75	
CON							33.67
	6.08	5.42	8.15	5.97	6.16	5.90	
	258.87	205.87	465.50	249.50	266.00	243.50	
	8	8	8	8	8	8	
	35.63	24.13	24.13	25.75	32.13	33.00	
ECC							29.13
	6.76	8.66	8.15	12.78	10.75	11.44	
	319.87	524.87	464.87	1143.50	808.87	916.00	
	34.88	26.88	28.69	29.75	33.81	34.38	

APPENDIX I-D

ANOVA SUMMARY FOR PEAK TWITCH TORQUE

SOURCE	SS	DF	MS	F	P
BLOCKS/SUBJECTS	375.039	7			
ARM	63.749	1	63.749	5.621	.048
ERROR	79.392	7	11.342		
TIME	213.207	5	42.641	16.521	<.001
ERROR	90.333	35	2.581		
ARM TIME	17.940	5	3.588	4.397	.003
ERROR	28.555	35	.816		
TOTAL	868.216	95			
(RESIDUAL)	198.281	77			

	PRE	POS	24H	48H	72H	96H	
	8	8	8	8	8	8	
	8.62	5.07	8.72	8.37	8.39	8.13	
CON							7.88
	2.32	1.08	2.55	1.48	2.13	2.05	
	37.67	8.14	45.48	15.38	31.71	29.33	
	8	8	8	8	8	8	
	8.60	2.68	5.99	6.62	6.85	6.80	
ECC							6.25
	2.40	1.45	4.02	3.91	3.33	2.75	
	40.16	14.80	113.13	106.94	77.60	52.98	
	8.61	3.87	7.36	7.49	7.62	7.46	

APPENDIX I-E

ANOVA SUMMARY FOR MAXIMUM RATE OF TORQUE DEVELOPMENT

SOURCE	SS	DF	MS	F	P
BLOCKS/SUBJECTS	537610.035	7			
ARM	56863.113	1	56863.113	4.831	.062
ERROR	82401.324	7	11771.618		
TIME	210091.818	5	42018.364	11.973	<.001
ERROR	122825.316	35	3509.295		
ARM TIME	22798.871	5	4559.774	3.254	.016
ERROR	49041.200	35	1401.177		
TOTAL	1081631.680	95			
(RESIDUAL)	254267.841	77			

	PRE	POS	24H	48H	72H	96H	
	8	8	8	8	8	8	
	316.63	199.26	317.73	282.64	281.51	278.06	
CON							279.30
	88.50	62.73	107.85	58.20	80.79	63.25	
	54823.85	27541.36	81424.69	23708.06	45689.01	28007.84	
	8	8	8	8	8	8	
	317.68	122.00	223.15	235.80	248.58	236.58	
ECC							230.63
	100.57	74.21	139.54	126.31	127.98	91.58	
	70800.45	38544.90	136300.03	111674.94	114659.91	58702.83	
	317.15	160.63	270.44	259.22	265.04	257.32	

APPENDIX I-F

ANOVA SUMMARY FOR HALF RELAXATION TIME

SOURCE	SS	DF	MS	F	P
BLOCKS/SUBJECTS	4869.650	7			
ARM	2168.566	1	2168.566	2.728	.140
ERROR	5563.715	7	794.816		
TIME	5821.770	5	1164.354	12.410	<.001
ERROR	3283.748	35	93.821		
ARM TIME	4315.022	5	863.004	16.095	<.001
ERROR	1876.658	35	53.619		
TOTAL	27899.130	95			
(RESIDUAL)	10724.121	77			

	PRE	POS	24H	48H	72H	96H	
	8	8	8	8	8	8	
	67.06	62.34	64.58	68.05	66.75	62.02	
CON							65.13
	11.29	12.27	5.90	7.18	8.09	6.36	
	892.38	1053.46	243.66	360.92	458.10	282.87	
	8	8	8	8	8	8	
	70.00	28.55	47.76	54.75	67.41	65.29	
ECC							55.63
	6.28	14.28	21.46	22.42	15.07	18.00	
	276.04	1427.24	3223.64	3518.52	1588.77	2268.17	
	68.53	45.45	56.17	61.40	67.08	63.65	

APPENDIX I-G

ANOVA SUMMARY FOR MOTOR UNIT ACTIVATION

SOURCE	SS	DF	MS	F	P
BLOCKS/SUBJECTS	2634.811	7			
ARM	24.736	1	24.736	.213	
ERROR	812.376	7	116.054		
TIME	1499.388	5	299.878	3.218	.017
ERROR	3261.617	35	93.189		
ARM TIME	624.250	5	124.850	1.043	.408
ERROR	4190.268	35	119.722		
TOTAL	13047.448	95			
(RESIDUAL)	8264.262	77			

	PRE	POS	24H	48H	72H	96H	
	8	8	8	8	8	8	
	95.35	79.87	90.90	89.58	97.30	93.09	
CON							91.02
	5.55	13.42	8.75	7.22	2.80	6.37	
	215.31	1260.88	535.86	364.65	54.93	284.37	
	8	8	8	8	8	8	
	93.10	87.16	84.27	90.54	89.87	95.06	
ECC							90.00
	5.19	14.84	25.96	10.73	9.89	5.90	
	188.44	1540.69	4719.09	806.24	685.06	243.56	
	94.23	83.51	87.58	90.06	93.59	94.08	

APPENDIX I-H

ANOVA SUMMARY FOR OVERALL PERCENTAGE OF DISRUPTED FIBERS

SOURCE	SS	DF	MS	F	P
BLOCKS/SUBJECTS	5389.874	7			
TIME	35920.802	4	8980.200	21.282	<.001
ERROR	11815.183	28	421.971		
TOTAL	53125.858	39			

BAS	P-C	P-E	48C	48E	
8	8	8	8	8	
3.10	33.24	81.66	37.36	79.85	
4.04	20.16	19.01	30.29	27.51	47.04
114.22	2844.48	2529.40	6421.12	5295.84	

APPENDIX I-I

ANOVA SUMMARY FOR PERCENTAGE OF FIBERS WITH "EXTREME" DISRUPTION

SOURCE	SS	DF	MS	F	P
BLOCKS/SUBJECTS	9295.774	7			
ARM	7284.245	1	7284.245	5.651	.047
ERROR	9023.550	7	1289.079		
TIME	360.461	1	360.461	.560	
ERROR	4505.664	7	643.666		
ARM TIME	46.080	1	46.080	.431	
ERROR	748.255	7	106.894		
TOTAL	31264.029	31			
(RESIDUAL)	14277.469	21			

	POS	48H	
	8	8	
	12.78	17.09	
CON			14.93
	18.84	21.45	
	2485.77	3219.65	
	8	8	
	40.55	49.66	
ECC			45.11
	35.26	36.18	
	8702.40	9165.42	
	26.66	33.38	

APPENDIX II: ETHICS APPROVAL AND SAMPLE CONSENT FORM

McMASTER UNIVERSITY
HAMILTON, ONTARIO, CANADA

COMMITTEE ON
THE ETHICS OF RESEARCH ON HUMAN SUBJECTS

TO: Dr. J. Duncan MacDougall

RE: Martin Gibala's M.Sc. Research

TITLE: The effects of concentric and eccentric resistance exercise on muscle ultrastructure

The above named applicant has submitted an application to the Committee on Ethics of Research on Human Subjects.

The Committee has reviewed this request and finds that it meets our criteria of acceptability on ethical grounds. The review has been conducted with a view toward insuring that the rights and privacy of the subject have been adequately protected; that the risks of the investigation do not outweigh the anticipated gain; and that informed consent will be appropriately obtained.

We concur in all necessary endorsements of the application.

Digby Elliott

Date: Dec 10/92

For the Committee on the Ethics of Research on Human Subjects

C.K. Bart, Associate Professor, Business
T. Beckett, Judge, Unified Family Court
I.M. Begg, Professor, Psychology
B. Donst, Ecumenical Chaplain, Chaplains' Office
D. Elliott, Associate Professor, Physical Education and Athletics (Chair)
R. Howard, Professor, Sociology
T. Kroeker, Lecturer, Religious Studies
R.J. Preston, Professor, Anthropology
J. Synge, Associate Professor, Sociology

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"THE EFFECTS OF CONCENTRIC AND ECCENTRIC RESISTANCE EXERCISE ON MUSCLE ULTRASTRUCTURE"

INFORMATION AND CONSENT FORM

The principal investigators for this project are Dr. Duncan MacDougall and Martin Gibala. They will provide you with a detailed verbal description of the procedures involved in the study. In addition, you are asked to carefully read the following information form and sign it if you wish to be a subject for this study.

A. PURPOSE

The purpose of this study is to examine the changes in muscle ultrastructure, strength and soreness that occur following concentric and eccentric resistance exercise.

B. PROCEDURES

During your first visit to the laboratory, an assessment of your maximal elbow flexor strength will be made, and a needle biopsy sample will be obtained from the biceps muscle of one arm. The biopsy will involve the removal of a small piece of muscle tissue (50-100 mg) by a skilled physician with a sterile hollow needle under local anaesthesia.

Approximately two-weeks later, you will again report to the laboratory on five consecutive days (Monday - Friday) at the same time. On the first day, you will be asked to perform an arm curl exercise with each arm separately. One arm will perform only concentric muscle actions (i.e. muscle shortening), while the other arm will only perform eccentric muscle actions (i.e. muscle lengthening). Approximately 80 repetitions in total will be performed in total with each arm. Immediately following the exercise, an assessment of elbow flexor strength will be made, and a needle biopsy sample will be obtained from the biceps of each arm. Maximal strength measures will similarly be made 24,48,72 and 96 hours following the exercise session. In addition, a second needle biopsy sample will be taken from the biceps of each arm 48 hours following the exercise. Thus, five biopsies will be performed in total (two samples from one arm, and three from the other). These samples will be analyzed for various indices of muscle damage, by comparing the baseline sample with those taken after the concentric and eccentric exercise bouts.

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C. DETAILS OF THE PROCEDURES AND THEIR POSSIBLE RISKS

The needle biopsy procedure involves the local injection of an anaesthetic ("freezing") into the skin of the biceps area, after which a small (4 mm) incision will be made and a small (50-100 mg) piece of muscle will be removed with a special needle. After the procedure a suture will close the skin and pressure will be applied to minimize bruising. Most people report little discomfort with the procedure. It will be performed by a skilled physician who is familiar with the technique.

Complications with the procedure are rare. However, in our experience with athletes, fewer than 1 in 400-500 subjects experience a local skin infection, 1 in 30-40 have a temporary (up to four months) localized loss of sensation in the skin at the site of incision, and a few subjects have mild bruising around the incision for 4-5 days. There is also the very rare (one in a million) chance that you may be allergic to the local anaesthetic.

D. CONFIDENTIALITY OF RESULTS

The data collected will be used in preparation of reports to be published in scientific journals. Subjects will not be identified by name in these reports. You will have access to your own data and the group data when it is available for your own interest.

E. REMUNERATION

You will receive an honorarium of $250.00 for the completion of the study to help compensate for your time commitment.

F. FREEDOM TO WITHDRAW FROM THE STUDY

You are free to withdraw from the study at any time. If, after reading the above information, you are interested in participating as a subject you should read the statement below and sign in the space provided.

I HAVE READ AND UNDERSTAND THE ABOVE EXPLANATION OF THE PURPOSE AND PROCEDURES OF THE PROJECT AND AGREE TO PARTICIPATE AS A SUBJECT.

Signature: _____

Date: _____

Witness: _____