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Strain rate effects on the mechanical properties and fracture mode of skeletal muscle



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ABSTRACT

The present study aimed to characterize the mechanical response of beagle sartorius muscle fibers under strain rates that increase logarithmically (0.1 mm/min, 1 mm/min and 10 mm/min), and provide an analysis of the fracture patterns of these tissues via scanning electron microscopy (SEM). Muscle tissue from dogs' sartorius was excised and test specimens were sectioned with a lancet into sections with nominal length, width, and thickness of 7, 2.5 and 0.6 mm, respectively. Trimming of the tissue was done so that the loading would be parallel to the direction of the muscle fiber. Samples were immediately tested following excision and failures were observed under the SEM. No statistically significant difference was observed in strength between the 0.1 mm/min (2.560 \pm 0.37 MPa) and the 1 mm/min (2.702 \pm 0.55 MPa) groups. However, the 10 mm/min group (1.545 \pm 0.50 MPa) had a statistically significant lower strength than both the 1 mm/min group and the 0.1 mm/min group with p < 0.01 in both cases. At the 0.1 mm/min rate the primary fracture mechanism was that of a shear mode failure of the endomysium with a significant relative motion between fibers. At 1 mm/min this continues to be the predominant failure mode. At the 10 mm/min strain rate there is a significant change in the fracture pattern relative to other strain rates, where little to no evidence of endomysial shear failure nor of significant motion between fibers was detected.

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1. Introduction

Soft tissue injuries of the musculoskeletal system can be challenging to treat. A thorough understanding of the underlying mechanical behavior of these tissues is necessary to understand the pathology and for adequate treatment and surgical intervention if necessary. Much work has been done investigating the mechanical properties and the repair of ligaments (i.e. ACL/PCL) and of tendons (rotator cuff). Additionally there can be a significant injury to the muscle in isolation and further characterization of muscle biomechanical behavior and fracture mode is warranted.

Muscle tears and partial muscle tears can occur as a consequence of overexertion of the muscle [1,2], steroid use [3], and trauma [4]. These injuries are often the result of eccentric loading and can take weeks to months to heal with loss of productivity in the short term and possible long term decrease of function [5]. Understanding of the mechanical behavior of this tissue can help us to better formulate preventative

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measures and to potentially develop interventional modalities in high demand individuals such as professional athletes and military personnel, where prolonged removal from activity may have significant professional consequences. An understanding of the dynamic mechanical properties of biological muscle tissue is needed to determine the response of the human body to traumatic loading.

Skeletal muscle tissue is a complex fiber-oriented structure composed of approximately 80% water, 3% fat and 10% collagenous tissues [6]. It can, therefore, be expected to display similar mechanical properties resulting from interactions between these constituents as shown for soft tissues with similar structures, such as ligaments and tendons [7,8]. An understanding of the mechanism of muscle failure can help in understanding the damage mechanisms, designing physical therapy routines, surgical repair techniques, and providing key data for informed design rationale in developing medical devices or protective systems [9–11]. However, this failure mechanism is not well understood for skeletal muscle tissues, especially when tensile loading is considered.

Palevski et al. [12] have noted the sparsity on the viscoelastic properties of skeletal muscle. The published data consists of a few compression and tensile tests, but they lack micrographic explanations as to the mechanism of muscle failure. Bosboom et al. [13]

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conducted compression studies on the tibialis anterior muscles of rats and developed a method to determine the passive transverse properties of skeletal muscle. Best et al. [14] performed live tensile tests on the tibialis anterior and extensor digitorum longus muscles of rabbits and established that the quasi-linear model is the most reliable way to characterize the structural responses of muscle. Zheng et al. [15] conducted ultrasound indentation experiments on human upper and lower limb soft tissues at differing indentation rates (0.75 mm/s to 7.5 mm/s) and found that the extracted young's moduli of limb soft tissues under similar conditions were repeatable. Van Loocke et al. [16] investigated the elastic properties of passive skeletal muscle tissue by means of quasi-static uniaxial unconfined compression tests performed on fresh porcine muscle tissue in vitro. While the results indicated that muscle elastic behavior is nonlinear and transversely isotropic, the study did not detail the fracture pattern and fracture mechanism of the muscle fibers.

There is little published literature that provides micrographic analysis of the fracture properties of muscle tissue. Carroll et al. [17] investigated the micrographic effects of tension on bovine semitendinosus muscle. They reported that stress applied parallel to the fiber axis resulted in the initial rupture of the muscle fiber-endomysium sheath, while perpendicular stress caused the initial rupture at the endomysium-perimysium junction without disturbing the muscle fibers. However, that study did not consider the effects of various strain rates on tension, nor did it consider the modes of muscle fracture, both of which are detailed in the present study. The present study aimed to characterize the mechanical response of beagle sartorius muscle fibers under strain rates that increase logarithmically (0.1 mm/min, 1 mm/min and 10 mm/min), and provide an analysis of the fracture patterns of these tissues via scanning electron microscopy (SEM).

2. Materials and methods

2.1. Specimen preparation and test materials

All of the experiments were conducted within the biological ethics protocol of NYU and NYU-Poly. Five beagles were euthanized for orthopedic bone related research under the bioethics approval using an overdose of anesthesia; muscle tissue from the sartorius was excised and stored in a 70% ethanol solution at 5–7 °C. The test specimens were sectioned with a lancet into sections with nominal length, width, and thickness of 7, 2.5 and 0.6 mm, respectively. Trimming of the tissue was done so that the loading would be parallel to the direction of the muscle fiber. The initial cross-sectional area was determined by averaging three measurements carried out in correspondence with the central region of the specimen [18]. Samples were immediately tested following excision and then stored in 70% ethanol prior to scanning electron microscopy (SEM).

2.2. Microtensile experimental setup

The machine employed for testing was an Instron 5566 universal test system (Instron, Norwood, MA, USA) equipped with an Intsron 100 N load cell. The specimen ends were bonded to the plates of the machine with cyanoacrylate glue (Scotch Super Glue AD117, 3 M Co., St. Paul, MN, USA). To preserve integrity of the muscle, the muscle strips were moistened with distilled water prior to bonding and throughout the entire test procedure. Tests were conducted at room temperature (25 °C).

Twenty specimens were tested at three different strain rates each, for a total of 60 specimens. Three logarithmically increasing loading rates of 0.1, 1.0, and 10 mm/min were selected for testing. Load–displacement data was obtained using Bluehill® 2.0 software (Instron, Norwood, MA, USA). Tests were conducted until the specimen completely failed. Improperly fractured specimens, such as those fractured at the grips, were excluded from the study. Statistical analysis was performed by

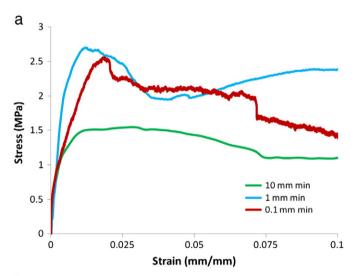
multiple two-tailed heteroscedastic t-tests with statistical significance set to $\alpha=0.05$. The specimens were then examined via SEM for the characterization of failure mode.

2.3. Freeze fracture

Untested samples were freeze-fractured to compare the effects of tensile forces and failure modes due to absolute abrupt fracture on muscle tissue. Untested muscle samples that had previously been placed in a liquid ethanol environment were placed in a liquid nitrogen bath for 75 s, and then fractured at the edges using two pairs of tweezers. The frozen segments were returned to liquid ethanol to thaw before drying at room temperature.

2.4. Characterization of failure mode

A Hitachi S-3500 N variable pressure SEM (Hitachi Ltd, Tokyo, Japan) was used to analyze structural damage caused by the tensile forces. Samples were sputter coated with gold for 60 s prior to SEM examination.



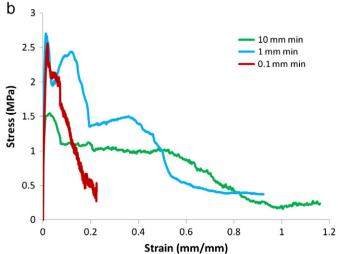


Fig. 1. Representative engineering stress–strain curves of beagle sartorius muscle tested at different strain rates. (a) Initial region of the curves to show the near linear region upon immediate loading, yield, ultimate stress and the initiation of gross failure of the specimen; (b) representative curves in their entirety through complete gross failure.

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