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DIMINISHED FATIGUE AT REDUCED MUSCLE LENGTH IN HUMAN SKELETAL MUSCLE

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Abstract

Understanding muscle fatigue properties at different muscle lengths is essential to improve electrical stimulation applications in which impaired muscle is activated to produce function or to serve as an orthotic assist. This study examined the effects of muscle length on fatigue in human quadriceps muscle. Twelve healthy subjects were tested at short and long muscle lengths (15° and 90° of knee flexion, respectively) before and after a fatigue-producing protocol using low-, high-, and variable-frequency testing trains. Greater fatigue was observed at the longer muscle length, supporting the notion that fatigue is largely dependent upon metabolic factors. Fatigue, however, was characterized by greater attenuation of low- than high-frequency responses (i.e., low-frequency fatigue, LFF) at the long length. This observation, accompanied by the fact that variable-frequency trains produced greater augmentations in force production than comparable low-frequency trains at the longer length, suggests that excitation–contraction coupling impairment is also a contributing factor to fatigue and plays a greater role at the more fatigue-susceptible longer muscle length.

Keywords

catchlike property; excitation–contraction coupling; frequency; functional electrical stimulation; muscle fatigue

Muscle fatigue is the decrease in the force-generating ability of a muscle resulting from recent activation.⁵ Fatigue exhibits task dependency and its evolution and extent depend on the conditions at which the muscle operates. In the late 1960s, Aljure and Borrero¹ used the influence of muscle length as a model to determine whether fatigue in isolated toad sartorius muscle was due either to failure of the contractile machinery resulting from metabolic activity or to impairments in excitation–contraction coupling. More recently, Fitch and McComas¹⁵ conducted a similar study using human ankle dorsiflexor muscles. The premise of both of these studies was that if the amount of fatigue observed was independent of muscle length, then fatigue could be attributed to impairments in excitation–contraction coupling. Conversely, if fatigue resulted from metabolic events associated with crossbridge interactions, then greater fatigue should be observed at optimal length because crossbridge interactions would be maximal. Both studies found less fatigue if the muscles were fatigued at a short suboptimal length and evaluated at a long length than if the muscles were fatigued and evaluated at a long

length.^{1,15} Thus, both groups concluded that the metabolic consequences of contractile activity were the primary mechanism for muscle fatigue and that reduced contractile activity when the muscle was held in the shortened length resulted in the decrease in fatigue. Aljure and Borrero¹ also noted, however, that muscles that were both fatigued and tested at the short length demonstrated greater fatigue than if fatigued and tested at the optimal length.

Sacco et al.²⁸ studied human dorsiflexor muscles and also found contradictory findings, depending on the muscle length at which fatigue was evaluated. These investigators concluded that their data supported both metabolic factors and impairments in excitation–contraction coupling as mechanisms for fatigue. Sacco and colleagues concluded that diminished fatigue observed when the muscle was fatigued at the short length and then tested at the optimum length was due to movement of the leg, which attenuated the fatigue. This conclusion supports the theory that impairment in excitation–contraction coupling is the mechanism for fatigue, resulting from T-tubule action potential propagation failure.

They postulated that greater narrowing or compression of the T-tubule may occur at short muscle lengths and that excitation–contraction coupling failure, due to localized ionic imbalances that impair action potential propagation in the transverse tubules, may have created relatively quiescent regions of muscle fibers during stimulation. Moving the muscle to a longer length would account for the unexpected force recovery if the previously quiescent fibers now contributed to force development. These studies raise important concerns for understanding the mechanisms underlying fatigue.

In spite of several studies on the human ankle dorsiflexor muscles,^{15,28} this model presents various methodological difficulties. Fitch and McComas were unable to monitor changes in dorsiflexion torque when fatiguing the muscles at short lengths. Additionally, Sacco and colleagues did not discuss the contribution of changes in the effective lever arm of the dorsiflexors and its potential effect on myofilament overlap and torque production about the ankle. Thus, the relative contribution that changes in muscle length and lever arm make to the observed decline in force is not known. To overcome these difficulties, we chose to study the human quadriceps femoris muscle.

The purpose of this study was to examine the effects of muscle length on fatigue resulting from repetitive activation with short-duration stimulation trains. Because a previous study in our laboratory showed surprising fatigue resistance at short muscle lengths,²² we hypothesized that less muscle fatigue would occur at short than long muscle lengths. The quadriceps femoris was activated at both the same absolute and relative forces at the two muscle lengths to control for the level of force produced at each muscle length. On a subpopulation of subjects, responses to high- and low-frequency trains of stimulation were used to examine excitation–contraction coupling failure at each muscle length. Additionally, because of our laboratory's long-standing interest in using the catchlike property of skeletal muscle to augment muscle forces,^{6–9,23} stimulation trains that elicit a catchlike response were also used to evaluate muscle performance for each fatigue condition. Preliminary results have been presented elsewhere.²²

METHODS

Subjects

Data were obtained from 12 healthy volunteer subjects (6 men, 6 women), ranging in age from 19 to 29 years (mean 22.17, SD $\pm$ 3.38 years), with no history of lower-extremity orthopedic problems. The University of Delaware Review Board approved this study and all subjects signed informed consent forms.

Experimental Arrangement

Subjects were seated on a computer-controlled dynamometer (KinCom II; Chattecx Corporation, Chattanooga, Tennessee) with their hips flexed to 75°. The pelvis, right leg, and thigh were stabilized with Velcro straps. The dynamometer axis was aligned with the knee joint axis and the force transducer pad was positioned against the anterior surface of the leg, approximately 3 cm proximal to the lateral malleolus. The right quadriceps femoris muscle was stimulated using a Grass S8800 stimulator with an SIU8T stimulus isolation unit (Grass Technologies, West Warwick, Rhode Island). All stimulation pulses were 600 μ s in duration. Two self-adhesive, 3 $\times$ 5-inch electrodes were used to stimulate the muscle. With the knee positioned at 90°, the anode was placed proximally over the motor point of the rectus femoris portion and obliquely across the width of the quadriceps femoris muscle. Because the skin over the thigh shifts with respect to the quadriceps as the knee is moved into extension, the knee was then placed at 15° of knee flexion and the cathode was placed distally over the motor point of the vastus medialis portion and obliquely across the width of the quadriceps. Pilot testing showed that this placement of the cathode produced the most stable muscle recruitment when the muscle was stimulated at 15° and 90° of knee flexion. Prior to testing, we adjusted electrode placement to ensure uniform force responses throughout the range of stimulation intensities expected to be used for the individual. A sudden increase or decrease in force either within an individual force response or across successive responses would indicate motor unit recruitment or dropout, respectively. If either phenomenon occurred, the electrodes were repositioned and the force responses observed again. For all testing procedures, the stimulator was driven by a personal computer that controlled all timing parameters of each stimulation protocol. All force data were digitized on-line at a rate of 200 samples per second and stored for subsequent analysis.

Training Sessions

Prior to the commencement of the experimental sessions, all subjects participated in one training session. Subjects were familiarized with the experimental protocol and trained to relax during stimulation of their quadriceps muscle. For each subject, the maximum voluntary isometric contraction (MVIC) was determined by using a burst superimposition technique²⁹ in which a 100-Hz, 10-pulse train at supramaximal stimulation intensity was delivered to the quadriceps muscle during an attempted MVIC. If the stimulation produced a <5% increase in force, the force produced by the subject was determined to be the subject's MVIC. Conversely, if the stimulation produced a >5% increase in force, the subject rested for 5 minutes before attempting another MVIC. For each subject, MVICs were determined at knee joint angles of 15° (short muscle length) and 90° (long muscle length). Pilot work determined that subjects ($n = 8$) produced maximum forces at joint angles ranging from 75° to 90°. All subjects produced MVICs within three trials at each muscle length during the training session. The order of MVIC testing for the two muscle lengths was determined randomly for each subject.

Experimental Sessions

At both the short and the long muscle lengths, each subject's muscle was activated at two stimulation intensities. One stimulation intensity produced a force equal to 20% of the subject's short muscle length MVIC, and the other produced a force equal to 20% of the subject's long muscle length MVIC. For convenience, the force equal to 20% of the subject's MVIC when the muscle was held at the short length is referred to as "low force" and the force equal to 20% of the subject's MVIC at the long length as "high force." Each subject participated in four experimental sessions. Each experimental session consisted of one of four conditions: low force level and testing with the muscle held at the short (15°) length (low force/short); low force level and testing with the muscle held at the long (90°) length (low force/long); high force level and testing with the muscle held at the short length (high force/short); and high force level and

testing with the muscle held at the long muscle length (high force/long). The order of the four experimental sessions was randomized independently for each subject. Subjects were asked to refrain from strenuous exercise for at least 24 hours prior to each session, and sessions were separated by at least 48 hours. Comparison of low force/short vs. low force/long and comparison of the high force/short vs. high force/long allows comparison of the fatigue produced at the same absolute forces at the two muscle lengths. In contrast, comparison of low force/short vs. high force/long determines the effects of muscle length on fatigue at the same relative force (i.e., 20% of MVIC).

At the beginning of testing for each session, the knee joint angle to be used for that session was set using the computer-controlled dynamometer. For tests at 15° of flexion, the passive force on the load cell with the knee in the test position was recorded and used for gravity correction for subsequent force measures at this angle. At 90° of flexion, the passive force on the load cell was zero. Next, MVIC testing was conducted and the session was continued only if the subject was able to perform an MVIC that was at least 95% of the MVIC produced during the training session at the angle tested. All subjects were able to meet this MVIC standard within three attempts.

After MVIC testing, the subject rested for 5 minutes and then a 14-pulse, 25-ms interpulse interval train (40 pps) was delivered to the muscle once every 5 seconds to potentiate the muscle and set the stimulation intensity. Stimulation intensity was initially adjusted to elicit approximately either the subject's low or high force, depending on the force level being tested. The intensity was then held constant until the force did not increase over three successive trains. This typically required activation of the muscle with 5–10 stimulus trains. Following this force potentiation, the intensity was readjusted to produce the low or high force. The intensity was then kept constant throughout the remainder of the session in an attempt to recruit a consistent population of motor units from each subject's muscle. The muscle was re-potentiated prior to the commencement of each pre-fatigue testing sequence and prior to the commencement of the fatigue-producing sequence (see later).

During each experimental session, subjects received pre-fatigue testing and a fatigue-producing and a fatigue-testing sequence (see Fig. 1). All subjects' pre-fatigue testing sequence included low-frequency testing, which consisted of an alternating order of a low-frequency constant-frequency train [six pulses at 14.3 pps (70-ms interpulse intervals)] and a comparable variable-frequency train (an initial 5-ms interpulse interval followed by four 70-ms interpulse intervals) repeated three times for a total of six trains. Trains were delivered once every 10 seconds to avoid fatiguing the muscle. A subset of 7 subjects also received high-frequency testing, consisting of a high-frequency train [six pulses at 60 pps (16.7-ms interpulse intervals)] and a variable-frequency train. This subset of subjects received the high-frequency testing prior to low-frequency testing and each test was separated by a 5-minute rest (see Fig. 1).

Five minutes after the last pre-fatigue test, the fatigue-producing sequence commenced. The fatiguing sequence was a modification of the fatigue test proposed by Burke and colleagues¹¹ and used a 14-pulse, 40-pps train that was delivered once per second for 180 seconds (i.e., 180 contractions). Immediately (i.e., 1 second) following the fatiguing protocol, all subjects underwent testing with the low-frequency testing trains. To maintain a consistent level of fatigue and to control for prior activation history, trains continued to be delivered once per second. The first testing train was followed by three of the 40-pps trains used to produce fatigue. Successive testing trains were also separated by three 40-pps testing trains. Thus, each testing train was delivered once every 4 seconds. For the subset of subjects who previously received the high-frequency testing protocol, immediately following the low-frequency fatigue testing, the high-frequency fatigue testing was commenced (see Fig. 1).

Data Management

All forces measured at 15° of knee flexion were gravity-corrected for the weight of the subject's limb. Peak forces were calculated from the force responses to each train of the fatigue-producing sequence. The peak forces were then normalized to the maximum response during the fatigue test to compare the fatigue produced during each of the four testing conditions (Fig. 2A). The average peak forces of the final six responses of the fatiguing sequence (i.e., trains 175–180) were used to quantify the percentage of initial force remaining at the end of the fatiguing sequence (Fig. 2B).

The peak forces and force–time integrals produced in response to each testing train were also calculated. The force–time integral is the area under the force trace; it is commonly used to quantify the force augmentation produced by trains that elicit a catchlike response in skeletal muscle.^{4,25,33} The three occurrences of each train were averaged for the pre-fatigue and for the fatigued testing sequences for each testing condition. The percent peak force remaining was calculated for each train type from the ratio of fatigued to pre-fatigue values for each condition to quantify fatigue of the low- and high-frequency train responses. The ratio of the force–time integral produced in response to the variable compared to the constant low-frequency train was used to quantify force augmentation due to stimulation with variable-frequency trains for each testing condition.

Statistical Methods

Two-way repeated measures analyses of variance (ANOVAs) were performed to determine the effects of muscle length and force level on the percentage of the peak forces remaining in response to the fatigue-producing sequence (i.e., 40-pps trains). If a significant main effect for muscle length was observed, two-tailed paired *t*-tests were used to compare data at the same absolute (low force/short vs. low force/long; high force/short vs. high force/long) and relative (low force/short vs. high force/long) force levels across muscle lengths. Likewise, if a significant main effect for force level was observed, two-tailed paired *t*-tests were used to compare force levels within each muscle length (low force/short vs. high force/short; low force/long vs. high force/long). In addition, two-tailed paired *t*-tests were used to compare the relative attenuation in peak forces seen in response to the high- and low-frequency trains within each testing condition. Two-tailed paired *t*-tests were also used to compare the variable- and low-frequency train force–time integral responses within each testing condition. Separate analyses were performed for pre-fatigue and fatigued data. For each analysis, an observation was significant at $P \geq 0.05$.

RESULTS

As anticipated, the MVIC produced by each subject was considerably lower at the short than the long muscle length. The average MVICs with the knee at 15° and 90° of flexion were 335.57 ± 79.72 N and 714.08 ± 285.80 N, respectively.

Fatiguing Protocol

Peak forces in response to each train of the fatiguing protocol during each experimental condition showed a steady decline in force until approximately the 90th contraction, after which consistent forces were produced during subsequent contractions (Fig. 2A). There was a significant effect for muscle length on the peak forces remaining ($F = 9.785$, $P = 0.01$) (Fig. 2B), but no effect for force level was seen. When the low-force condition was used to produce fatigue, 57.9% and 48.0% of the initial peak forces remained when the muscles were fatigued at the short and long muscle lengths, respectively ($P = 0.073$). Similarly, for the high-force condition, 61.3% and 45.9% of the initial peak forces remained at the short and long muscle lengths, respectively ($P \leq 0.05$).

Low- and High-Frequency Testing Train Responses

For both muscle lengths, the responses to the low-frequency trains showed greater attenuation (c.f. low-force/short condition where $T = 1.937$, $P = 0.101$) than the responses to the high-frequency trains (Fig. 3). The peak forces remaining are shown in Figure 3. Ratios of the percent of the pre-fatigue peak forces remaining for the low- and high-frequency trains were 0.802 and 0.648 for the low force/short and high force/short conditions, respectively, and 0.516 and 0.529 for the low-force/long and high-force/long conditions, respectively. Thus, the relative attenuation of low-frequency trains compared with high-frequency trains was greater at long than short muscle lengths. This difference was significant for the same absolute low-force condition (low force/short vs. low force/long) and nearly significant for the same relative force condition (low force/short vs. high force/long).

Low- and Variable-Frequency Train Responses

There was no significant difference in the force–time integrals produced by the low- and variable-frequency trains before the muscles were fatigued (Fig. 4). In contrast, variable-frequency trains produced significantly greater force–time integrals than the low-frequency trains for all testing conditions when muscles were fatigued; they produced 1.29 and 2.13 times greater force–time integrals than constant-frequency trains in the conditions of low force/short and low force/long length, respectively. The variable-frequency train augmentation was 1.31 and 1.48 for the conditions of high force/short and high force/long length, respectively. Thus, the relative augmentation produced by the variable-frequency trains was greater at long than short muscle lengths if either the same absolute forces (low force/short vs. low force/long; high force/short vs. high force/long), or the same relative forces (low force/short vs. high force/long) were used to fatigue the muscle.

DISCUSSION

The major finding of the present work is that greater fatigue was observed at long than short muscle lengths, regardless of whether the same absolute or relative force levels were used. Also, greater attenuation of low- than high-frequency train responses was observed at long than short muscle lengths. Last, for the fatigued muscles, variable-frequency trains were found to produce greater augmentations in force than comparable low-frequency trains at long or short muscle lengths.

Quadriceps Femoris as a Model for Studying Muscle Length-Dependent Fatigue

Use of the human ankle dorsiflexor muscle presents several methodological difficulties. Fitch and McComas were unable to monitor changes in dorsiflexion torque during their fatiguing procedure when activating the muscles at short muscle lengths due to concurrent activation of the antagonist peroneus longus and brevis muscles.¹⁵ With the quadriceps muscle, however, we were able to control forces and monitor them throughout all experiments. Additionally, although Sacco and colleagues presented a length–force relationship for the dorsiflexors that is consistent with previous investigators,²⁴ the contribution of changes in the effective lever arm of the dorsiflexors about the ankle on this relationship was not discussed. It was presumed that the decline in force at short lengths primarily reflected the reduced overlap between the myofilaments. Thus, the relative contribution that changes in muscle length and lever arm make to the observed decline in force is not known. To overcome these difficulties, we chose to study the human quadriceps femoris muscle. We are able to record forces while electrically activating the quadriceps femoris at long and short muscle lengths without contamination from antagonist muscles. Additionally, the patella glides anteriorly and superiorly as the knee joint extends, and the result is a longer lever arm for the patellar tendon of the quadriceps at 15° than 90°.¹⁷ Thus, by testing the quadriceps muscle, we are certain that the reduction in knee extensor

torque at 15° is due to muscle length changes and not to concomitant shortening of the effective lever arm.

Metabolic Differences and Crossbridge Interactions

Fitch and McComas¹⁵ found that twitch and tetanic responses measured at a long muscle length showed less attenuation if the muscles were fatigued at the short than optimal length and concluded that metabolic factors resulting from crossbridge interactions were the most likely mechanism for fatigue. Although our methods differed (i.e., we did not alter muscle length during testing), our results also showed greater fatigue for muscles activated at long than short muscle lengths if either the same absolute or relative forces were used to activate the muscle. Thus, by using their premise,¹⁵ our results support their conclusion that fatigue largely depends on metabolic factors associated with contractile activity.

Like Fitch and McComas,¹⁵ Sacco and colleagues found that muscles fatigued at short lengths demonstrated less fatigue when tested at optimum angle length.²⁸ However, if muscle length was not altered prior to eliciting fatigue testing responses, they noted greater fatigue when testing at the short length. This latter finding conflicts with our findings. They further suggested that the apparent greater fatigability of muscle activated at optimum angle length was due to greater fatigue recovery by the short muscle as a result of rest or movement to the longer length for testing. We chose to fatigue and evaluate the muscle's force-producing ability without changing muscle lengths. Thus, the lesser fatigability at short lengths observed in our experiments cannot be attributed to a recovery process associated with movement to a different muscle length. Rather, we show that in situ human skeletal muscle is less fatigable at short than long muscle lengths when force levels and percentage activation of the muscle are well controlled. Although stating the contrary in text, Rassier's²⁶ results also show that the human quadriceps femoris is less fatigable at short (30° knee flexion) than long (90° knee flexion) muscle lengths following a fatigue protocol consisting of nine tetanic (50 Hz) 5-second isometric contractions, with a 5-second interval between contractions.

Sacco and colleagues²⁸ used ³¹P-nuclear magnetic resonance spectroscopy to support their claims that metabolic factors associated with contractions at optimum or short muscle lengths are not the cause for the differences in fatigue observed as a function of length. Similar to an earlier report by Baker and colleagues,³ they could not demonstrate significant differences between the metabolic changes associated with contractions at optimum angle lengths compared with short muscle lengths of the tibialis anterior. Hence, they concluded that fatigue differences as a function of muscle length must rely on other factors. Caution must be used, however, in generalizing the results of these two studies. Baker and colleagues³ could not measure force at the short muscle lengths. Interestingly, Sacco and colleagues did show a trend for less adenosine triphosphate turnover occurring at the short than long lengths, which supports Fitch and McComas's contention that fewer crossbridge interactions occur at the short length than at the optimal length and, thus, less fatigue occurs¹⁵; however, because they studied only two subjects, statistical significance was not established. Unfortunately, animal studies do not yield additional insight regarding metabolic consequences of muscle activation as a function of length, because the results are conflicting.^{2,18,30,32}

Excitation–Contraction Coupling

It has been suggested that a breakdown in excitation–contraction coupling may result during fatigue if sarcolemmal action potential propagation is impaired in the transverse tubules.^{19, 28} Narrowing or compression of the transverse tubule was postulated to occur at short muscle lengths, which could exacerbate ion accumulation within the transverse tubular spaces and thus lead to failure of sarcolemmal action potential propagation. Although we agree that impairment in excitation–contraction coupling plays a role in fatigue, our data do not support the conclusion

that excitation–contraction coupling failure was due to T-tubular ionic disturbances. One factor that may explain the discrepancy between our results and those of Sacco et al. is the duration of the stimulation trains used to produce fatigue. With occluded circulation, Sacco and colleagues used six 15-second trains (30 pps) delivered 5 seconds apart to fatigue the muscle and then used 2-second trains (50 pps) to assess the muscle's responses. We kept the circulation intact and used 180 14-pulse trains (40 pps, i.e., 325 ms long) delivered once per second to fatigue the muscle and then used 6-pulse trains (14.3 pps, 60 pps, and variable-frequency trains) to assess the muscle's responses. Because our stimulation trains were brief and intermittent, ionic disturbances in the T-tubule system were unlikely. Additionally, we observed greater attenuation of low- than high-frequency train responses, which suggests the presence of “low-frequency” fatigue and argues against action potential propagation failure, because high-frequency trains have the potential to create greater ionic disturbances in the T-tubular system due to their short interpulse intervals. Interestingly, we observed that the evidence for low-frequency fatigue was greater at long than short muscle lengths.

Although the exact mechanisms for greater attenuation of low- than high-frequency responses are not known, most investigators agree that it is due to disruption in excitation–contraction coupling.^{12,21,31} One postulated explanation for low-frequency fatigue is diminished calcium release from the sarcoplasmic reticulum.^{10,12,13,16,21} Although the action potential is normal during low-frequency fatigue, the calcium transient is lower for a given level of stimulation.³¹ Low-frequency stimulation (i.e., frequencies producing subtetanic responses) produces calcium transients that fall along the steep portion of the force–calcium concentration curve.¹² Thus, during low-frequency stimulation, small changes in calcium release will result in large changes in force. In contrast, for high-frequency stimulation, small changes in calcium concentration will result in negligible changes in force because the muscle is operating on the flat, asymptotic portion of the force–calcium concentration curve. This could explain why less attenuation was seen with high-frequency and variable-frequency trains than with low-frequency trains when the muscles were fatigued.

Our findings support the notion that failure of excitation–contraction coupling is secondary to consequences of metabolic activity.¹⁵ Greater fatigue observed at long than short muscle lengths supports the contention that there is a greater metabolic cost to activation at long lengths. Mounting evidence suggests that metabolic byproducts of activity (e.g., increased intracellular lactate,¹³ magnesium,²⁰ and hydrogen peroxide¹⁰ concentrations), changes in counter-ion conductance in the sarcoplasmic reticulum,¹⁴ and elevated calcium concentrations^{12,21} all can cause impairments in excitation–contraction coupling at the level of the calcium-release channel. If any of these impairments caused low-frequency fatigue, we would anticipate that the impairment would be greater at long than short lengths, as we have observed (c.f. Rijkkelijkhuizen et al.²⁷).

In the present study, variable-frequency trains produced approximately 29% to 31% and 48% to 113% augmentation in force–time integrals over the constant-frequency trains during conditions of low force/short, high force/short, high force/long, low force/long length, respectively, when the muscle was fatigued. Parmiggiani and Stein²⁵ noted that one mechanism by which variable-frequency trains augment force occurred when the initial high-frequency portion of the variable-frequency trains released greater Ca^{2+} per pulse than constant-frequency trains. Our results are consistent with this mechanism because augmented Ca^{2+} release would explain the decreased susceptibility of variable-frequency trains to impairments in excitation–contraction coupling as compared with constant-frequency trains, and the observation that augmentation by variable-frequency trains was greatest at long muscle lengths when low-frequency fatigue was the greatest.

The present work has shown that human skeletal muscle is less fatigable when repetitively activated at short than long muscle lengths when using either the same absolute or relative forces at each length. Our findings support the notion that fatigue is largely dependent on metabolic factors associated with contractile activity. However, because we observed greater attenuation of low- than high-frequency response with fatigue and these differences were greater at long muscle lengths, where fatigue was greatest, excitation–contraction coupling impairment must be a contributing factor to fatigue. Further study is needed to identify the cause of this impairment in excitation–contraction coupling.

Abbreviations

ANOVA, analysis of variance; MVIC, maximum voluntary isometric contraction.

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FIGURE 1.

Schematic of an experimental testing session. HFT, high-frequency train (6 pulses, 16.7-ms interpulse intervals); VFT, variable-frequency train (6 pulses, initial doublet followed by four 70-ms interpulse intervals); LFT, low-frequency train (6 pulses, 70-ms interpulse intervals); FT, fatigue-producing train (14 pulses, 40 pps).

FIGURE 2.

(A) Group ($N = 12$) normalized peak force responses to the 180-contraction fatigue producing sequence for each testing condition. (B) Percentage of initial peak force remaining at the end of the fatigue-producing sequence for each testing condition. The muscle was fatigued at long and short muscle lengths and at the same absolute low or high levels of force (low force/short and low force/long; high force/short and high force/long, respectively), or at the same relative force (low force/short and high force/long). Same absolute force comparison: $*P \leq 0.01$. Same relative force comparison: $†P \leq 0.05$.

FIGURE 3.

Group ($N = 7$) percentage of pre-fatigue peak force remaining in response to low-frequency (14.3 pps) and high-frequency (60 pps) testing trains during fatigue. Asterisks indicate a significant difference between the low- and high-frequency responses. $*P \leq 0.01$; $^{\dagger}P \leq 0.001$.

FIGURE 4.

Pre-fatigue and fatigued normalized group ($N = 12$) force–time integrals in response to variable-frequency train (VFT) and constant low-frequency train (CFT) stimulation for the different conditions. VFT responses were normalized to the CFT responses within each testing condition. Asterisks indicate significant difference in force–time integrals produced by the CFTs and VFTs. VFTs produced significant augmentation in force–time integrals for all conditions when the muscle was fatigued. * $P \leq 0.01$; † $P \leq 0.001$.