ORIGINAL RESEARCH

VASTUS LATERALIS MOTOR UNIT RECRUITMENT THRESHOLDS ARE COMPRESSED TOWARDS LOWER FORCES IN OLDER MEN

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Abstract: Background: Aging results in adaptations which may affect the control of motor units. Objective: We sought to determine if younger and older men recruit motor units at similar force levels. Design: Cross-sectional, between-subjects design. Setting: Controlled laboratory setting. Participants: Twelve younger (age = 25 ± 3 years) and twelve older (age = 75 ± 8 years) men. *Measurements:* Participants performed isometric contractions of the dominant knee extensors at a force level corresponding to 50% maximal voluntary contraction (MVC). Bipolar surface electromyographic (EMG) signals were detected from the vastus lateralis. A surface EMG signal decomposition algorithm was used to quantify the recruitment threshold of each motor unit, which was defined as the force level corresponding to the first firing. Recruitment thresholds were expressed in both relative (% MVC) and absolute (N) terms. To further understand age-related differences in motor unit control, we examined the mean firing rate versus recruitment threshold relationship at steady force. Results: MVC force was greater in younger men (p = 0.010, d = 1.15). Older men had lower median recruitment thresholds in both absolute (p = 0.010, d = 0.0100.005, d = 1.29) and relative (p = 0.001, d = 1.53) terms. The absolute recruitment threshold range was larger for younger men (p = 0.020; d = 1.02), though a smaller difference was noted in relative terms (p = 0.235, d = 0.50). These findings were complimented by a generally flatter slope (p = 0.070; d = 0.78) and lower y-intercept (p = 0.070; d = 0.78) 0.009; d = 1.17) of the mean firing rate versus recruitment threshold relationship in older men. Conclusion: Older men tend to recruit more motor units at lower force levels. We speculate that recruitment threshold compression may be a neural adaptation serving to compensate for lower motor unit firing rates and/or denervation and subsequent re-innervation in aged muscle.

Key words: Motor unit, recruitment threshold, aging, electromyography.

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Introduction

In 1977, Tomlinson and Irving observed drastic morphological differences in the spinal cords of older adults, noting that individuals over the age of 60 possessed significantly fewer functioning motor neurons compared to younger adults (1). More recently, age-related changes to the structure and function of the neuromuscular system have garnered significant interest due to the implications for independence and quality of life of older adults (2), a growing percentage of the population (3). Age-related impairments in the neuromuscular system include reductions in maximal motor unit firing rates (4), the inability to voluntarily activate muscle (5), and corticospinal hypoexcitability among weak older adults (6). These changes may be accompanied by neurodegenerative processes, resulting in motor neuron loss, leaving corresponding muscle fibers denervated and orphaned (7). Many of these fibers are subsequently reinnervated by remaining neighboring motor units (8). How motor unit recruitment patterns may be affected by this process is not completely understood.

There is evidence suggesting that the force level at which a motor unit is recruited (i.e., recruitment threshold) provides insight into the control of the neuromuscular system. For Received July 2, 2019

example, motor unit recruitment thresholds progressively decrease as a muscle fatigues as a compensatory mechanism to offset decline in twitch force amplitude (9). To further our understanding of the aging neuromuscular system, we set out to examine differences in motor unit recruitment thresholds for younger versus older men. Using recently developed surface electromyographic (EMG) signal decomposition algorithms [10], we observed that vastus lateralis motor unit recruitment thresholds are compressed towards lower forces in older men. We propose that recruitment threshold compression may be a compensatory strategy used by the central nervous system to control force output following denervation and subsequent reinnervation of aged muscle.

Methods

Participants

Twelve younger (mean \pm SD age = 25 \pm 3 years, BMI = $22.0 \pm 1.9 \text{ kg/m}^2$) and twelve older (age = $75 \pm 8 \text{ years}$, BMI = $25.9 \pm 3.1 \text{ kg/m}^2$) men completed this study. Participants cleared a health screening, medical history, and physical activity readiness questionnaire (PAR-Q) before enrollment. Individuals were excluded if they exceeded a body mass index (BMI) of 30 kg/m², presented metabolic or neuromuscular dysfunction, been diagnosed with knee osteoarthritis, required the use of a walking device, or had lower extremity surgery within the previous year. During the six months prior to enrolling, participants refrained from lower-body resistance training (< three times monthly) or other structured exercise (e.g., jogging) more than 30 minutes per day, three times per week (11). Participants were instructed to maintain their daily pattern of sleep and dietary intake, including caffeine. Participants refrained from physically demanding tasks for ≥ 24 hours prior to data collection. Exclusion due to medication was determined on a case-by-case basis. A physician was consulted prior to enrollment of two older participants. The following medications were permitted, with the number of participants using them in parentheses: Esomeprazole (one), Levothyroxine (three), Finasteride (one), Omeprazole (one), Irbesartan (one), Tamsulosin (one), Allopurinol (one), Propranolol (one), and Atorvastatin (one). The Texas Tech University Human Research Protection Program approved the study (Study ID #504976). All participants read, understood, and signed an informed consent document prior to participation.

Isometric Force Assessments

A custom knee extension chair was used for testing. Participants were seated and restrained in the chair with straps secured around their chest, abdomen, and hips. Arms remained crossed throughout testing. An ankle cuff was secured to the dominant ankle joint and attached to a calibrated tension/ compression load cell (Model SSM-AJ-500; Interface, Scottsdale, AZ). All testing was performed at a knee joint angle of 120°. Following a warm-up, participants performed three, five-second maximum voluntary contractions (MVC) separated by three minutes of rest. Strong verbal encouragement was provided. The highest force from the three contractions was used to standardize submaximal testing. Following MVC determination, participants performed trapezoidal isometric contractions by tracing visual templates on a computer monitor. Participants increased isometric force from 0-50% MVC in five seconds (10%/second), held 50% constant for 15 seconds, and decreased isometric force from 50-0% MVC in five seconds (10%/second). All participants demonstrated the ability to gradually and linearly increase force prior to data collection. Multiple attempts were performed, each followed by a three minute rest period.

Surface EMG Signal Recording

Surface EMG signals were recorded from the vastus lateralis during each submaximal contraction via Bagnoli 16-channel Desktop system (Delsys, Inc., Natick, MA). Prior to testing, the skin over the muscle and patella was shaved and cleansed with rubbing alcohol. Oil, debris, and dead skin cells were removed with tape. The sensor was placed over the muscle in accordance with recommendations described by Zaheer et al. (12). A reference electrode was placed over the patella. Signals were detected with a surface array EMG sensor (Delsys, Inc., Natick, MA) that consisted of five pin electrodes (10). Four of the five electrodes are arranged in a square, with the fifth electrode in the center of the square and at an equal distance of 3.6 mm from all other electrodes. Pairwise subtraction of the five electrodes was used to derive four single differential EMG channels. These signals were differentially amplified, filtered with a bandwidth of 20 Hz to 450 Hz, and sampled at 20 kHz. Signal quality (i.e., signal-to-noise ratio > 3.0, baseline noise value \leq 2.0 μ V root-mean-square, line interference < 1.0) was verified for a 20% MVC assessment prior to data acquisition.

Surface EMG Signal Decomposition

The four separate filtered EMG signals served as the input to the Precision Decomposition III algorithm, which was utilized via dEMG Analysis software (version 1.1, Delsys, Inc., Natick, MA). For further information concerning the technical aspects of this algorithm, see the work of De Luca et al. (13) and Nawab et al. (10). Briefly, this algorithm allows investigators to extract motor unit firing rate, recruitment/ derecruitment, and action potential morphology data from surface EMG signals obtained during isometric contractions. Surface EMG signals were decomposed into their constituent motor unit action potential trains. The trains were used to calculate a time-varying firing rate curve for each detected motor unit. All firing rate curves were smoothed with a onesecond Hanning filter. The mean number of pulses per second (pps) for a two-second interval corresponding to the steadiest force (i.e., lowest coefficient of variation) of each motor unit firing rate curve was calculated. Each motor unit's recruitment threshold was calculated as the absolute (N) and relative (% MVC) force level when the first firing occurred (Figure 1). High threshold motor units recruited during the constant-force portion of the contraction were not considered for analysis. Contractions in which motor unit activity was detected prior to the onset of muscle force (i.e., electromechanical delay) were not considered. Similarly, erratic contractions with decreases in force during the ascending portion of the trapezoid were not analyzed. Decompose-Synthesize-Decompose-Compare testing was used to remove motor units with detection accuracy < 93.0% (10). For a contraction to be considered for analysis, the decomposition output must have yielded at least five motor units.

Statistical Analyses

All variables were assessed for normality with Shapiro-Wilk's tests. Mean, median, and range values for recruitment threshold were compared between younger and older men in both absolute (N) and relative (% MVC) terms. For each contraction, the relationship between the mean firing rates and recruitment thresholds of decomposed motor units was examined using linear regression. The resulting linear slope coefficient (pps/% MVC) and y-intercept (pps) values were then quantified. Independent samples t-tests were used to compare differences between age groups. Cohen's d effect size

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The black line represents the ascent of isometric force up to 50% MVC, whereas each colored circle is indicative of the first firing of a motor unit. The blue arrow points to the first active motor unit and the red arrow indicates the last recorded motor unit. Descriptive data from each contraction is provided in the table. The morphology of each motor unit action potential is displayed to the left.

statistics were used to examine differences, with values of 0.20, 0.50, and 0.80 reflecting small, medium, and large effects (14), respectively. An alpha level of $P \le 0.05$ was used to evaluate statistical significance. JASP software (version 0.9.0.1, JASP, Amsterdam, The Netherlands) was used for statistical analysis.

Results

All variables were normally distributed. The mean \pm SD number of motor units detected was 17 \pm 5 for younger and 13 \pm 4 for older men. MVC force was significantly greater in younger vs. older men (709.6 \pm 197.8 vs. 520.8 \pm 121.6 N [p = 0.010; Cohen's d = 1.15]). The relative median recruitment threshold values were significantly greater for younger (26.6 \pm 9.1% MVC) compared to older (15.6 \pm 7.9% MVC [p = 0.005; d = 1.29]) men. Younger men also demonstrated greater median recruitment threshold values when expressed in absolute terms (198.0 \pm 99.2 vs. 81.2 \pm 43.0 N [p = 0.001; d = 1.53]). Similarly, large differences in mean recruitment thresholds

were found when expressed in both relative $(25.9 \pm 7.7 \text{ vs. } 16.2 \pm 7.8\% \text{ MVC} [p = 0.005; d = 1.27])$ and absolute $(191.4 \pm 87.5 \text{ vs. } 85.1 \pm 44.9 \text{ N} [p = 0.001; d = 1.53])$ terms. The relative recruitment threshold range was not significantly different (p = 0.235) between younger $(22.6 \pm 9.5\% \text{ MVC})$ and older $(18.5 \pm 6.4\% \text{ MVC})$ men, but the effect size was moderate (d = 0.50). However, the absolute range was considerably larger for younger $(167.6 \pm 92.4 \text{ N})$ compared to older $(95.7 \pm 36.5 \text{ N} \text{ [p = 0.020; d = 1.02]})$ men. Univariate scatterplots displaying individual participant data have been provided in Figures 2-4.

Regarding the relationship between firing rate and recruitment threshold, the mean y-intercept value was significantly greater for younger (28.7) compared to older men (22.3 [p = 0.009; d = 1.17]). The mean slope coefficients were not significantly different (-0.52 vs. -0.32 [p = 0.070]); however, a moderate effect was observed (d = 0.78). Example relationships for two participants have been displayed in Figure 5.

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Figure 2

Individual participant data depicting median recruitment threshold expressed in relative and absolute terms for both younger and older men. Each data point represents the median recruitment threshold value from all of the successfully decomposed motor units within a contraction



Figure 3

Individual participant data depicting mean recruitment threshold expressed in relative and absolute terms for both younger and older men. Each data point represents the mean recruitment threshold value from all of the successfully decomposed motor units within a contraction



Discussion

This study compared vastus lateralis motor unit recruitment thresholds in younger versus older men. Our findings demonstrated that older men tend to recruit a greater proportion of motor units at lower isometric forces. This conclusion was based on moderate-to-large age-related differences in median recruitment threshold values, and was consistent for both relative and absolute force levels. These results were further supported by age-related shifts in the mean firing rate versus recruitment threshold relationship, which would be suggestive of lower firing rates for low threshold motor units of older men. We propose that recruitment threshold compression towards lower forces serves as a compensatory neural strategy that offsets motor unit remodeling in aged skeletal muscle (15). We will attempt to explain the mechanistic contributors to these findings in subsequent paragraphs.

Figure 4

Individual participant data depicting recruitment threshold range expressed in relative and absolute terms for both younger and older men. Each data point represents the recruitment threshold range from all of the successfully decomposed motor units within a contraction. Range was calculated as the difference between the greatest observed recruitment threshold value and the lowest observed value



In the 1970's, Tomlinson and Irving (1) quantified limb motor neurons within the lumbrosacral cord in 47 humans aged 13 to 95 years. They noted considerably fewer motor neurons for those ≥ 60 years, with some lumbrosacral cords displaying counts equal to half of those evident in early adulthood or middle age. Shortly thereafter, investigators began investigating the effects of aging on skeletal muscle. Cross-sectional studies noted reduced muscle size (16), a reduction in fiber size and number (17), and a greater proportion of slow-twitch fibers (18). Given the significant fiber type grouping in aged muscle (19), the aging process likely results in substantial denervation of dormant muscle fibers and subsequent reinnervation by the more active, low threshold motor neurons, thereby increasing motor unit size. This concept is also supported by the presence of extra large motor unit potentials observed in the EMG recordings of older adults. Thus, the notion that voluntary control of motor units may be affected by the aging process is well-grounded in physiology (20).

Figure 5

Individual participant data for the vastus lateralis mean firing rate versus recruitment threshold relationship in one younger and one older man. Note the flatter linear slope coefficient and lower y-intercept for the older participant. The slope and y-intercept values shown in this figure are only slightly different from the means within each age group



Motor unit recruitment compression towards lower forces appears to be consistent with other concepts in the aging literature. Erim et al. (21) reported lower recruitment thresholds in the first dorsal interosseous of older adults, with particularly pronounced differences at 50% MVC. Their findings were accompanied by a number of age-related differences in motor unit behavior, such as reduced common drive, and exceptions to the typically inverse relationship between mean firing rate and recruitment threshold. Second, our findings seem to be in agreement with the work of Fling et al. (22), who noted preservation of the size principle in aged muscle. Fling et al. (22) postulated that if the contractile properties of the reinnervated fibers are suboptimal, a decrease in the net twitch force amplitude could result, despite the motor unit becoming larger. Earlier motor unit recruitment might also be a marker of neuromuscular inefficiency at the onset of an isometric contraction. Kamen and De Luca (23) noted that, despite sufficient familiarization, high threshold motor units displayed prolonged activity during force descent in older adults, which they attributed to excessive antagonistic coactivation. Another explanation for force being controlled by lower threshold motor units may be related to a compensatory mechanism to offset decreased firing rates. This hypothesis is well-supported by our results, as well as the work of Watanabe et al. (24). These authors (24) examined age-related differences in the firing rates of the vastus lateralis at 30% and 70% MVC, finding that the dissimilarity between age groups in firing rates was particularly evident for low threshold motor units. This suggests that, at low forces, older adults must recruit a greater proportion of motor units in order to overcome their slower firing rates. In line with our findings, Kamen and De Luca (23) and Erim et al. (21) also reported a flatter (i.e., less negative) slope for the mean firing rate versus recruitment threshold relationship (21, 23). The results of the current study appear to support these previously observed differences in firing rates, particularly the lower threshold motor units, as well as the less negative slope in older men.

Much of the work in this area has relied on intramuscular recordings, whereas our analysis relied on surface EMG signals. Thus, it is important to be mindful that EMG results are a reflection of the motor units within the vicinity of the recording electrodes. Autopsy studies have demonstrated that vastus lateralis fast-twitch muscle fibers (innervated by large, high threshold motor neurons) are likely to be located near the surface of the skin (25). Therefore, it has been suggested that surface EMG signals may be biased towards high threshold motor neurons become reinnervated by low threshold motor neurons, this might suggest that the surface EMG signal in older adults reflects a greater balance of motor unit types. Potential differences between groups in subcutaneous fat thickness may have also affected these results (12).

In summary, the results of this study revealed important age-related differences in MVC force, absolute and relative median recruitment thresholds, absolute recruitment threshold range, and the mean firing rate versus recruitment threshold relationship. These results imply that older men tend to recruit motor units of the vastus lateralis at lower isometric force levels compared to younger men. This is in line with past observations for the first dorsal interosseous (21) and tibialis anterior muscles (22). We speculate that motor unit recruitment threshold compression towards lower forces is a result of agerelated motor unit remodeling. Future studies should investigate whether this observed behavior is prevalent in women or middle-aged men, as well as whether these properties carry over to dynamic movements.

Conflict of interest: The authors declare no conflicts of interest. *Ethical standards:* This study was carried out in accordance

with the ethical standards of the university Institutional Review Board.

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