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MOTOR UNIT NUMBER AND TRANSMISSION STABILITY IN OCTOGENARIAN WORLD CLASS ATHLETES: CAN AGE-RELATED DEFICITS BE OUTRUN?

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Abstract (251words)

Our group has shown a greater number of functioning motor units (MU) in a cohort of highly-active older (~65y) masters runners relative to age-matched controls. Owing to the precipitous loss in the number of functioning MUs in the 8th and 9th decade of life it is unknown whether older world class octogenarian masters athletes (MA) would also have greater numbers of functioning MUs (MUNE) compared with age-matched controls. We measured MU numbers and neuromuscular transmission stability in the tibialis anterior of world champion MAs (~80y), and compared the values to healthy age-matched controls (~80y). Decomposition-enhanced spike-triggered averaging was used to collect surface and intramuscular electromyography signals during dorsiflexion at ~25% of maximum voluntary isometric contraction (MVC). Near fibre (NF) MU potential analysis was used to assess neuromuscular transmission stability. For the MAs as compared with age-matched controls; the amount of excitable muscle mass (CMAP) was 14% greater (p<0.05), there was a trend (p=0.07) towards a 27% smaller surface detected motor unit potential – representative of less collateral reinnervation, and 28% more functioning MUs (p<0.05). Additionally, the MAs had greater MU neuromuscular stability than the controls as indicated by lower NF jitter and jiggle values (p<0.05). These results demonstrate that high performing octogenarians better maintain neuromuscular stability of the MU and mitigate the loss of MUs associated with aging well into the later decades of life during which time the loss of muscle mass and strength become functionally relevant. Future studies need to identify the concomitant roles genetics and exercise play in neuroprotection.

New and Noteworthy: World champion master athletes in their 9th decade of life had a greater number of surviving motor units, reduced collateral reinnervation, better neuromuscular transmission stability and a greater amount of excitable muscle mass as compared with age-matched controls. The presumed better maintenance of MUs occurs at a time point when motor unit loss is greatest and the loss of muscle mass becomes functionally relevant, potentially maintaining function and attenuating sarcopenia.

Key Words: Aging, Physical activity, Muscle function, Master athletes, EMG, Sarcopenia, Dynapenia

Introduction

Aging is associated with a loss of functioning motor units (MU) (7, 9, 12, 27, 36). The loss of MUs is concomitant with the denervation of muscle, loss of motor axons and eventual alpha-motoneuron (MN) death (11, 16). With electrophysiological techniques it is possible to estimate the number of functioning MUs and thus make inferences on the number of surviving MNs in old age. Additionally, we can gain insight into neurophysiological changes associated with aging, such as MU neuromuscular transmission instability (17, 31). Age-related alterations to neuromuscular transmission instability may reflect clinical conditions of neuromuscular transmission disturbance and may represent early axonal denervation (2, 11). Moreover, alterations to electrophysiological measures of MU transmission may indicate MU dysfunction preceding the functionally relevant loss of strength and excitable muscle mass known as sarcopenia (16).

From cross-sectional studies it appears there is a gradual reduction in the number of functioning MUs after the third decade of life until the 7^{th} decade followed by a rapid decline into very old age (26, 28, 32). The early adult loss of MUs does not appear to be associated with weakness or functional decline due to the preservation of muscle mass and strength (28), through the process of collateral reinnervation whereby healthy MNs sprout axons which reinnervate those muscle fibers remaining following the death of a MN. The number of functioning MUs can be estimated (MUNE) electrophysiologically by dividing the mean 'electrical' size of surface-detected motor unit potentials (S-MUP) into the corresponding size parameter of the compound muscle action potential (CMAP) [(36) Figure 1]. Age-related reductions of 40-60% in the number of functioning MUs have been reported for several human limb muscles,

including: the biceps brachii (8, 14, 33), extensor digitorum brevis (29), vastus lateralis (31), tibialis anterior (17, 28, 32, 34), soleus (10, 40) and small intrinsic hand muscles (7, 14, 15).

Our group has reported greater MUNE in individuals with high-levels of life-long physical activity in a muscle that typically demonstrates age-related MU loss (34). We found that masters runners in their 7^{th} decade of life had a similar CMAP and a smaller mean S-MUP compared to their age matched counterparts. These parameters likely indicate the masters runners have not undergone the same extent of MU remodelling (i.e. MU loss and subsequent collateral reinnervation) as did their age-matched controls. Moreover, cross-sectional studies indicate that the age-related loss of MUs is exacerbated in healthy older adults who are in their 8^{th} and 9^{th} decades of life (28, 32). Thus, the purpose of this study is to compare cross-sectionally, a cohort of masters athletes (MAs) and age-matched controls to answer the important question: *if functioning motor units are maintained in a group of MAs in their 7^{th} decade of life, can world-class MAs two decades older also show a maintenance of motor units– during a time point in which MU loss is typically greatest, and the loss of muscle mass becomes functionally relevant?* We investigated, electrophysiologically, MU number and MU stability in a cohort of some of the world's most successful agers; octogenarian world class MAs. We hypothesised there will be MU remodeling as indicated by changes in the electrophysiological measures of MU stability, and S-MUPs in both groups, albeit less in the MAs. In addition, the MA will have a higher MUNE compared with age-matched controls owing to more excitable muscle mass (as indicated by a larger CMAP) through improved collateral reinnervation.

Methods

***Participants*:** Participants consisted of 29 (Table 1.) elderly males and females with no known neurological, musculoskeletal, metabolic or cardiovascular health conditions. The Masters Athletes (MA) consisted of track and field athletes ranked in the top 4 of their respective events at the world masters championships (including 7 current world record holders). Event specialties ranged from sprint and power events to middle and long distance running events (800m up to marathon). The age- and sex-matched controls were living independently and recruited from the local community. All participants were asked to refrain from unaccustomed and strenuous exercise prior to testing. This study was approved by the McGill Faculty of Medicine Institutional Review Board (IRB) for research involving human subjects (A08-M66–12B) and conformed to the Declaration of Helsinki. Informed written consent was obtained from all participants prior to the study.

Experimental Arrangement: All procedures were conducted during a single testing session on a Biodex System 3 dynamometer (Biodex Medical Systems, Shirley, New York, United States) using the isometric mode. The hip and knee angle were maintained at 90° while the participants were seated and reclined comfortably. Ankle angle was positioned to 30° of plantar flexion. The foot of the dominant leg (right) was secured to the footplate with two hook and loop inelastic straps (Velcro USA Inc. Manchester, NH, USA) across the toes and the dorsum of the foot and another strap secured the ankle. The torso of the participant was secured to the Biodex seat back by inelastic straps fastened across the shoulders and waist. To minimize extraneous leg movement, the thigh was supported and stabilized with an inelastic strap. The lateral malleolus was aligned with the dynamometer's axis of rotation.

Tibialis Anterior Electromyographic Data Acquisition. For the present investigation, the tibialis anterior (TA) was selected due to its known loss of MUs with normal adult aging (17, 28, 32, 34) and the role of high-levels of activity related to greater MUNE in masters runners (34). Participants performed 3 dorsiflexion maximal voluntary contractions (MVCs), with at least 3 min of rest between attempts. Each MVC was held for approximately 3 seconds, and participants were provided with real time visual feedback of their torque output and were verbally encouraged.

Voluntary Activation and Torque. Voluntary activation during the second and third MVC attempts was assessed using the interpolated twitch technique [ITT; (3)]. This technique involved supramaximal percutaneous electrical stimulation of the common fibular nerve inferior to the fibular head using a clinical stimulator/EMG system (Neuroscan Comperio system, Neurosoft, El Paso, Texas, USA). The amplitude of the interpolated torque electrically evoked during the plateau of the MVC was compared with a resting twitch evoked ~1s following the MVC. Voluntary activation (VA) was calculated as a percent using the equation: [1-(interpolated twitch /resting twitch)] x 100. The ITT ensured the participants were providing a maximal voluntary effort during MVCs to allow comparison of submaximal contraction levels (see below) between groups. Participants were required to achieve 95% VA or greater before continuing with data collection; and typical for this muscle group (18, 24, 37), this was achieved following familiarisation in all participants. The peak torque of the 3 MVC attempts was taken as the maximal torque amplitude for the participant. All torque signals were collected and sampled online at 500 Hz, and stored on the Biodex computer for additional offline analysis.

Surface EMG signals were recorded from the TA using self-adhering Ag-AgCl electrodes (1 cm × 3 cm). The active electrode was placed over the TA motor point, approximately 7 cm distal to the tibial tuberosity and 2 cm lateral to the anterior border of the TA. This placement was adjusted as needed to maximize TA CMAP amplitude and minimize rise time. The reference electrode was placed over the distal tendon of the TA. A ground electrode was placed over the patella.

Decomposition-based quantitative electromyography (DQEMG) data were acquired using a protocol described in detail elsewhere (13, 39). Intramuscular EMG signals were recorded via a disposable concentric needle electrode (Model N53153; Teca Corp., Hawthorne, NY) inserted into the TA, 5-10 mm distal to the active surface electrode. The surface and intramuscular EMG signals were bandpass filtered at 5 Hz to 1 kHz and 10 Hz to 10 kHz, respectively. Surface EMG signals were sampled at 3 kHz; intramuscular EMG signals were sampled at 30 kHz. To evoke the maximum CMAP a bar electrode held distal to the fibular head provided the delivery of supramaximal electrical stimuli to the common fibular nerve. Subsequently, participants matched a target line of 25% MVC, visible on a computer monitor, for all isometric dorsiflexion contractions while the intramuscular needle electrode was inserted and gently manipulated in the muscle to minimize the rise times of the majority of detected motor unit potentials (MUPs). This contraction intensity has been shown to be the most effective intensity for obtaining a representative MUNE in the tibialis anterior (TA) (28). Surface and intramuscular EMG signals were recorded during ~30 seconds of sustained steady target torque. Between contractions, the concentric needle electrode was repositioned in order to ensure

192 sampling of different MUs. These procedures were repeated until at least 20 suitable MUP trains
193 and their respective surface-motor unit potentials (S-MUPs) were acquired.
194
195 ***Tibialis Anterior Decomposition-based Quantitative Electromyography Analysis.***
196 Decomposed intramuscular EMG signals were reviewed off-line to determine the acceptability
197 of the extracted MUP trains and their corresponding S-MUPs. MUP trains were inspected
198 visually to ensure that their MUP occurrence patterns were consistent with the expected activity
199 of a single motor unit (consistent firing pattern and inter-discharge coefficient of variation of <
200 0.3). Invalid MUP trains and their associated S-MUPs were excluded from further analyses.
201 The DQEMG algorithms estimate a MUP and S-MUP template waveform and automatically
202 place markers related to onset, end, negative peak and positive peak positions, with respect to the
203 MUP template; and onset, negative peak onset, end, negative peak, positive positions with
204 respect to the S-MUP template. All MUP and S-MUP markers were subsequently reviewed
205 visually by the same operator. A MUNE was derived by dividing the negative-peak amplitude of
206 the maximal CMAP by the negative peak amplitude of the mean S-MUP.
207
208 For the assessment of MUP stability, used to reflect the stability of neuromuscular
209 transmission, MUPs that represent the isolated activity of a single motor unit were automatically
210 selected by the DQEMG algorithms. The sets of automatically selected, isolated MUPs were
211 inspected visually and any MUPs found to be significantly contaminated by the activity of other
212 motor units were removed. The DQEMG technique described has been shown to possess strong
213 test-retest reliability within individuals (5), and high degrees of intra- and inter-rater reliability in

control and clinical populations (2). The investigator was blinded to the status of the participant (Masters Athlete vs. age-matched controls) during off-line analysis.

Near Fibre Motor Unit Potential Parameters: Near Fibre MUP Template. The MUP template provided by the DQEMG algorithms was high-pass filtered using a second ordered low-pass differentiator (39). The second order filter equation is: $[\mathbf{x_t = y_{t+2} - y_{t+1} - y_t + y_{t-1}}]$. Where $\mathbf{y_t}$ is the sampled raw signal and $\mathbf{x_t}$ is the sampled filtered signal. Due to the spatial low-pass filtering properties of volume conduction, the resulting near fibre MUP template waveform, or The NF MUP is used to focus on characteristics of a motor unit's muscle fibres in close proximity (within ~350 μm) to the needle electrode and is defined as a MUP containing contributions from the fibres that are close to the detection surface of the needle electrode. As such, a NF "contribution" is the specific electrophysiological contribution of an individual NF (an individual muscle fibre or small group of motor unit muscle fibres) to a NF MUP. This methodology is particularly useful for studying variables related to neuromuscular transmission stability (i.e. jitter, jiggle) as it allows for the examination of individual MU waveforms with much less contamination from other, more distant MUs as compared to signals collected with traditional Butterworth filtering. This is due to the spatial filtering applied to create NF MUPs, which filters out more distant volume conducted MU activity which could potentially reduce the ability to detect individual muscle fibre activity. Measures of NF MUPs in turn can be used to reflect relative conduction times of muscle fibre action potentials to the electrode detection surface. Further details regarding NF MUP parameters have been described previously (2).

Near Fibre Parameters. The following NF parameters were originally described in clinical populations (1, 2) and some have been used in the investigation of normal adult aging (17). Near fibre area (NF Area) is the sum of the absolute values of the NF MUP between the onset and end positions multiplied by the sampling interval (i.e. 1/ (sampling rate). *Near fibre count* (NF fibre count) is the number of detected NF contributions to the NF MUP. A positive turn detected in the NF MUP with sufficient symmetry and amplitude is considered a distinct NF contribution. The NF fibre count reflects the density of fibres composing a motor unit. The *maximum near fibre interval* (max NF Interval) is the maximum time between consecutive detected NF contributions. Large max NF interval values may indicate long reinnervating axonal sprouts. *Near fibre jiggle* (NF Jiggle – Figure 1) is a statistic that measures the variability in the shape of consecutive isolated NF MUPs of a MUP train. The statistic is the same as originally applied to traditional MUPs (38). The NF MUPs of a MUP train are created by high-pass filtering each MUP using the same second ordered low-pass differentiator used to create the NF MUP template. Isolated NF MUPs are selected as described previously (39). *Near fibre jitter* (NF Jitter) is the mean consecutive difference of time intervals between a pair of distinct NF contributions found consistently within the NF MUPs of a selection of isolated NF MUPs as described by others (2, 38, 39). Suitable NF contribution pair tracking was confirmed by visual inspection.

Statistical analyses of the data were performed with SPSS version 22 (SPSS, Chicago, IL). Unpaired *t*-tests were used to compare participant characteristic values. A two-way analysis of variance (sex × activity status) was used to analyze all electrophysiological data. If no interactions were present for sex and activity status, the data were collapsed for sex and

compared across activity level using unpaired *t*-tests. The level of significance was set at $p \leq 0.05$. To explore the strength of apparent statistical effects, effect sizes (ES) were calculated using Cohen's d. Pearson product correlations (*r*) were implemented to test the strength of independent relationships between the NF MUP and MUNE parameters. Descriptive data in the text and tables are reported as means ± standard deviations; whereas data reported in the figures are means ± standard errors of the mean.

Results

Participant characteristics are presented in Table 1. Voluntary activation, as assessed using the interpolated twitch technique, was > 95% for all groups. There was a sex × activity status interaction for dorsiflexion strength and therefore the strength data were not collapsed across sex. The MA males were 28% stronger as compared with age-matched controls ($p < 0.01$; effect size (ES) = 1.88) and MA females were 22% stronger as compared with age-matched controls ($p < 0.01$; ES = 1.93). For all other variables listed below there were no interactions or main effects for sex. Therefore, data were collapsed across sex and compared for activity status.

Motor Unit Number Estimates. The groups did not differ ($p > 0.05$) in the root mean square (RMS) value of the surface EMG during the targeting contractions (~25% MVC) expressed as a percentage of MVC-RMS (25-30%) and the mean MU discharge rates did not differ ($p > 0.05$) between the groups. The negative peak amplitude of the CMAP was 14% larger in the MAs as compared with age matched controls ($p < 0.05$; ES = 0.98; Figure 2A), likely indicating a greater amount of excitable muscle mass. The negative peak amplitude of the mean S-MUP was not statistically different across groups. Nonetheless, there was a statistical trend (*p*

= 0.07; ES = 0.67) for the MA group to have a 27% smaller value (Figure 2B). This may indicate that collateral reinnervation is occurring in the MAs but the extent to which MUs are being remodelled, as further evidenced by the 20 μV smaller S-MUP, is less then age-matched controls. With a presumed greater amount of excitable muscle mass and less collateral reinnervation, MAs had a 28% greater number of functioning MUs as compared with their age-matched counterparts ($p < 0.01$; ES = 1.14; Figure 2C).

Neuromuscular Transmission Stability. Near fibre MUP parameter values are presented in Table 2. Master Athletes had similar: NF area, duration and maximal NF interval values as compared with age-matched controls, respectively ($p > 0.05$). However, for all other measures of neuromuscular transmission stability, MAs had smaller values as compared with age-matched controls: NF fibre counts (-19%, $p < 0.05$; ES = 0.93), NF jiggle (-21%; $p < 0.01$; ES = 1.21) and NF jitter values (-19%; $p < 0.05$; ES = 1.02), indicating greater neuromuscular transmission stability as compared with their age-matched counterparts. Additionally, across groups there was a significant positive association between increasing S-MUP amplitude and increases in NF jiggle ($r = 0.56$; $p < 0.01$) and NF jitter ($r = 0.52$; $p < 0.01$; Figure 3). Furthermore, as expected, there was a significant negative association with MUNE and NF jiggle ($r = -0.59$; $p < 0.01$) and NF jitter ($r = -0.55$; $p < 0.01$). These associations would indicate that as MUs are being remodeled (collateral reinnervation) with increasing MU size there is less stable neuromuscular transmission in the age-matched controls, but not MAs.

Discussion

The purpose of the present study was to investigate whether the estimated number of functioning motor units (MUNE) was higher in world class octogenarian masters athletes (MA) as compared with age-matched controls. Using electrophysiological techniques we investigated the number of functioning motor units (MU) and neuromuscular transmission stability in a cohort of the world's most successful agers: very old, world class MAs. The hypothesis was confirmed. Despite evidence of MU remodeling (S-MUP; Figure 2B), owing to a greater amount of excitable muscle mass as reflected by a larger CMAP) (Figure 2A), there was a higher MUNE in the MAs as compared with age-matched controls (Figure 2C). Additionally, the MAs had greater neuromuscular transmission stability than the controls as indicated by lower values for: NF jitter and NF jiggle, indicating relatively healthier MUs (Table 2). Previously we showed (34) that lifelong high-intensity physical activity may have the potential to limit the loss of functional MUs associated with natural aging well into the 7^{th} decade of life. Our current findings show this is also evident in world-class athletes nearly two decades older, a critical time-point when MU loss may be a great contributor to the loss of muscle mass (sarcopenia), resulting in substantial strength and functional deficits.

Motor unit number estimation. Indirect evidence of collateral reinnervation can be found from the size of the negative peak amplitude of the mean S-MUP. In a previous investigation of old masters runners (65 yrs), there was a similar sized S-MUP compared with young adults, whereas age-matched controls had a higher value (34). A higher S-MUP in age-matched controls as compared with masters runners and a similar CMAP indicated that the older runners had not undergone substantial collateral reinnervation, and therefore had higher MUNEs

than age-matched controls, and which were similar to that of young adults. In the present investigation, we showed that MAs in their 9th decade of life did not differ from age-matched controls for S-MUP values. However, presumably owing to a greater amount excitable muscle mass (CMAP) the MAs had a higher number of functioning MUs. It is important to note, there was a trend towards a difference in S-MUP across groups with a ~20μV lower value for the MAs, likely indicating that while there was MN loss and MUs are being remodeled, this process is occurring to a lesser extent in MAs than for age-matched controls (Figure 2B). Through collateral reinnervation muscle mass is maintained but subsequently larger MUs are formed as represented by the S-MUP and near fibre (NF) size parameters (Table 2) and it appears these larger MUs in both groups have different neuromuscular transmission stability properties (see below).

Our current findings in 'very old' age-matched controls are similar to that of McNeil et al. (28) who investigated MUNE in the tibialis anterior across: young, old and very old adults ranging from 23-89 years. They found, while there was no difference in strength between the young and old group, there was a reduction in MUNE. Moreover, in the very old group, there was a significant loss of both strength and MUNE, suggesting that functional significance of MU loss may not occur until after the 7th decade of life. In the present study the male and female MAs both had higher MUNE and higher strength values as compared with sex and age-matched controls (Table 1). Additionally, with advanced aging the capacity of MUs to continue sprouting is potentially less effective (30) and this could be the case in the age-matched controls. A possible explanation is that reinnervation may not be keeping pace with denervation in advanced age which could explain the reduced CMAP. By the nature of cross-sectional designs, selection

bias can be a limitation. Hence, further research is needed to establish longitudinal changes and whether physical activity can prevent or slow the age-related loss of the number of functional MUs.

Near Fibre MUP Parameters. Standard concentric needle electromyography can provide detailed information regarding the denervation-reinnervation process underlying age-related MU loss. The integrity of neuromuscular transmission can be identified through variability in the overall shape of consecutively detected NF MUPs and in the relative timings of their significant NF contributions (2, 38). Two key features related to variability in NF MUP shape and in the relative timings of significant NF contributions are jiggle and jitter, respectively (2, 38). Jiggle refers to the variability in overall NF MUP shape from one MU discharge to the next, and jitter refers to the variability of the time intervals between pairs of significant NF contributions across a set of isolated NF MUPs. Increases in both jiggle and jitter have been reported under clinical conditions of neuromuscular transmission disturbance and can reflect early axonal denervation (2, 11).

Our age-matched control NF jiggle results are consistent with two recent investigations specific to aging which found increased NF jiggle in NF MUPs detected in the tibialis anterior, vastus medialis (77yrs) (17) and vastus lateralis muscles of older adults (71yrs) as compared with young (31). The age-matched controls in the present study had higher values of NF jiggle and NF jitter as compared with MAs and this may be reflective of aberrations in muscle fibre action potential propagation, or neuromuscular transmission instability and could be due to the development of dysfunctional NMJs of newly reinnervated fibres (16). In the present study, NF

area was not different across groups which may be more reflective of intrinsic MU electrophysiological changes than the negative peak area of surface detected potentials (S-MUP) which was trending but was not significantly different between groups. Additionally, we show increased NF fibre count in age-matched controls as compared with MA, which is indicative of reinnervation presumably to compensate for prior denervation (i.e. motor unit remodeling) (6). These electrophysiological measures of grouping are consistent with histological evidence of increased MU homogeneity (i.e., fibre type grouping) in advanced age (16, 21, 25).

It appears that both groups of older adults are experiencing MU remodeling but the MAs exhibit reduced and more effective reinnervation indicated by smaller SMUPs and more stable neuromuscular transmission while the age-matched controls are presenting with more extensive remodeling and less stable neuromuscular transmission. This nature of the remodelling for both groups is further characterized by the positive relationships between NF jiggle, NF jitter and SMUP size, and a negative relationship with MUNE (Figure 3). Thus, in advanced age, collateral reinnervation seems to be resulting in less healthy MUs. Specifically, a decrease in neuromuscular transmission stability is thought to occur in association with partial loss of innervation by damaged motor axons, or incomplete reinnervation occurring between orphaned muscle fibres and their adopted axonal sprout during the process of collateral reinnervation (38). We suggest that the age-matched controls may have developed less stable new axons during reinnervation resulting in increased NF jitter and NF jiggle values.

Maintained Motor Unit Numbers and MU stability. Life-long physical activity has been shown to maintain spinal MN and MU numbers in rats (22) and humans (33-35), respectively.

The up-regulation of neurotrophic factors and sensitivity of the MN via increased neuromuscular activity (19, 20, 22), and maintained sympathetic input to the NMJ (23) may offset age-related MN death. As well, chronic activity into advanced age may have a protective effect at the epi-genomic level initiating changes in the methylation landscape of gene promoter pathways associated with MUs (4). Whether thorough exercise training across a lifespan or being born with exceptional genetics is responsible for the MAs exceptional athletic performance, the MAs presented with improved MU survival and reduced and more mature collateral reinnervation with better neuromuscular transmission stability when compared with age-matched controls. The challenge for future studies is to explore these key factors and to identify underlying mechanisms.

Conclusion

Accompanying the substantial loss of MUs is a progressive loss of contractile muscle mass and impaired whole muscle force generation (28, 32). This time-course of degradation is evident in the lower strength values and loss of MUs in the age-matched controls, whereas the MA had higher force production capacity of the ankle dorsiflexors possibly owing to not only a maintenance of MU number but more electrophysiologically stable MUs (32). It seems that MU electrophysiological quality is lower in the age-matched controls as indicated by reduced neuromuscular transmission stability as compared with the MAs – how this may influence strength or function is currently unknown. World champion MAs in their 9th decade of life had a greater number of surviving MUs, reduced collateral reinnervation, better preservation of neuromuscular transmission stability and hence better preservation of excitable muscle mass as compared with age-matched controls. The presumed better maintenance of MUs in MAs occurs

418 at a time point when MUNE loss is greatest and the loss of muscle mass and strength becomes
419 functionally relevant, potentially maintaining function and attenuating sarcopenia in this
420 exceptional cohort of older adults. Future studies on the potential neuroprotective effects of
421 exercise in older humans need to identify the concomitant role genetics and dose-dependence of
422 exercise in maintaining neuromuscular structure and function.
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Figure 3. Relationships between **A.** NF jiggle and **B.** NF jitter and S-MUP, **C.** NF jiggle and **D.** NF jitter and MUNE, between Masters Athletes (open circles) and age-matched controls (grey circles). All relationships were statistically significant ($p<0.01$). The participant (88 yrs) with the largest mean S-MUP had the lowest MUNE count and also had the highest values for NF jiggle and second highest for NF jitter, these data are denoted with a solid black circle. Removing this participant changes the '*r*' values slightly but statistical significance is not changed.

Table 1. Participant Characteristics

	Male Controls (n=9)	Male Athletes (n=7)	Female Controls (n=6)	Female Athletes (n=7)
Age (years)	82.8 ± 4.5	79.4 ± 3.7	79.3 ± 3.8	79.9 ± 6.2
Height (m)	1.71 ± 0.08†	1.74 ± 0.07†	1.53 ± 0.06	1.57 ± 0.06
Mass (Kg)	77.7 ± 10.8†	71.2 ± 10.9†	63.9 ± 11.0	52.9 ± 4.5*
Dorsiflexion Strength (N·m)	22.8 ± 6.8†	31.6 ± 11.1*†	12.8 ± 2.7	16.5 ± 2.5*

Mean ± SD

Table 1. Participant Characteristics. * Significant difference between Masters Athletes and age-matched controls. † = Significant difference between sex.

Table 2. Neuromuscular Transmission Stability and Near Fibre Parameters

Parameter	Masters Athletes	Age-Matched Controls	% Difference
NF Area (kV/s^{s}ms)	7.5 ± 2.6	8.1 ± 3.8	-
NF Duration (ms)	4.3 ± 0.8	4.8 ± 1.3	-
Max NF Interval (ms)	1.3 ± 0.3	1.2 ± 0.3	-
NF Fibre Count (#)	2.2 ± 0.3	2.7 ± 0.7*	*-19%*
NF Jiggle (%)	49.8 ± 8.6	63.2 ± 13.1*	*-21%*
NF Jitter (µs)	47.4 ± 7.8	58.2 ± 13.1*	*-19%*

* Denotes significant difference between groups.

NF – near fibre

Table 2. Neuromuscular Transmission Stability and Near Fibre Parameters. * Significant difference between Masters Athletes and age-matched controls

Figure1.

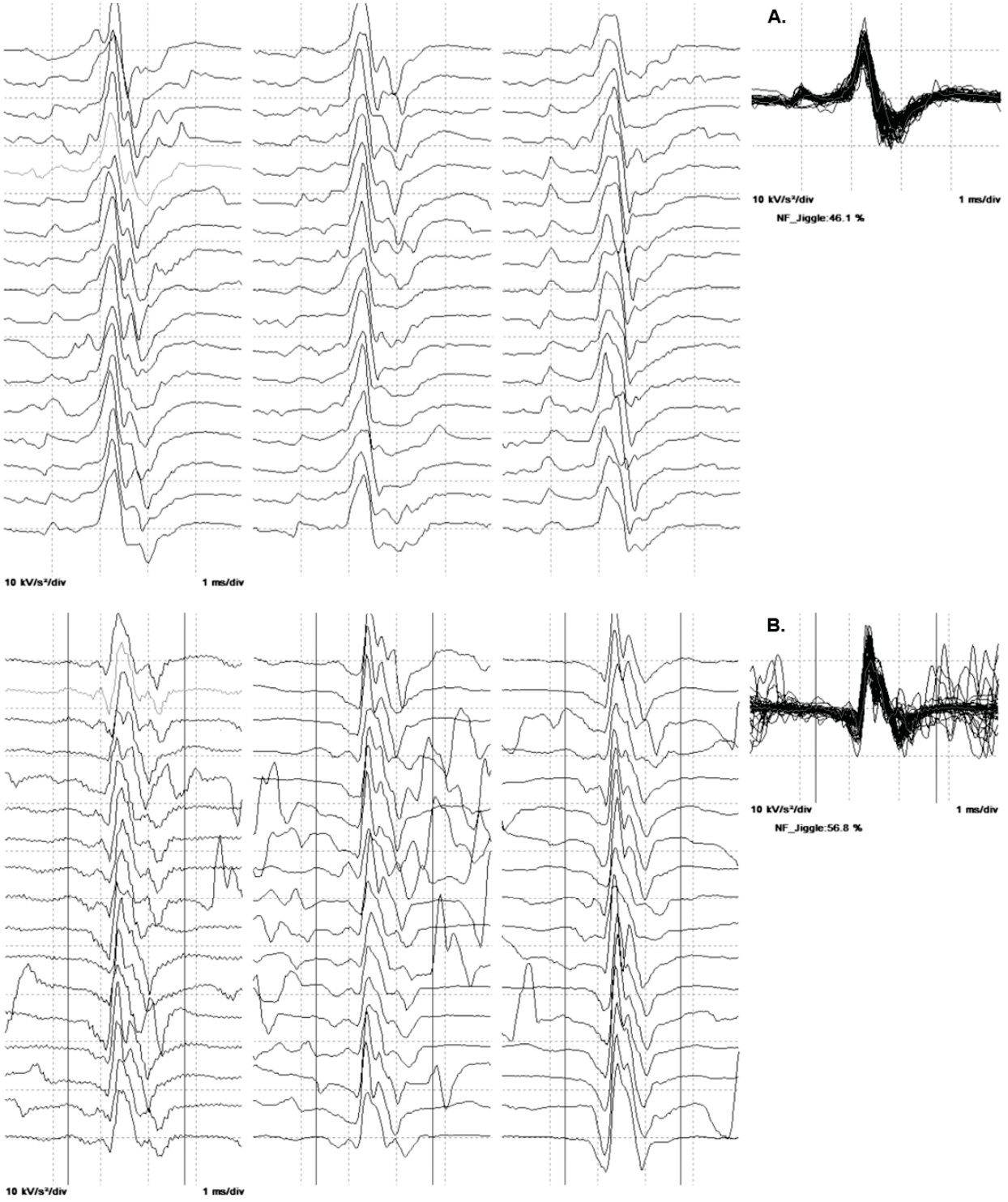

A.
10 kV/s²/div
1 ms/div
NF_Jiggle:46.1 %
10 kV/s²/div
1 ms/div
B.
10 kV/s²/div
1 ms/div
NF_Jiggle:56.8 %
10 kV/s²/div
1 ms/div

Figure 2

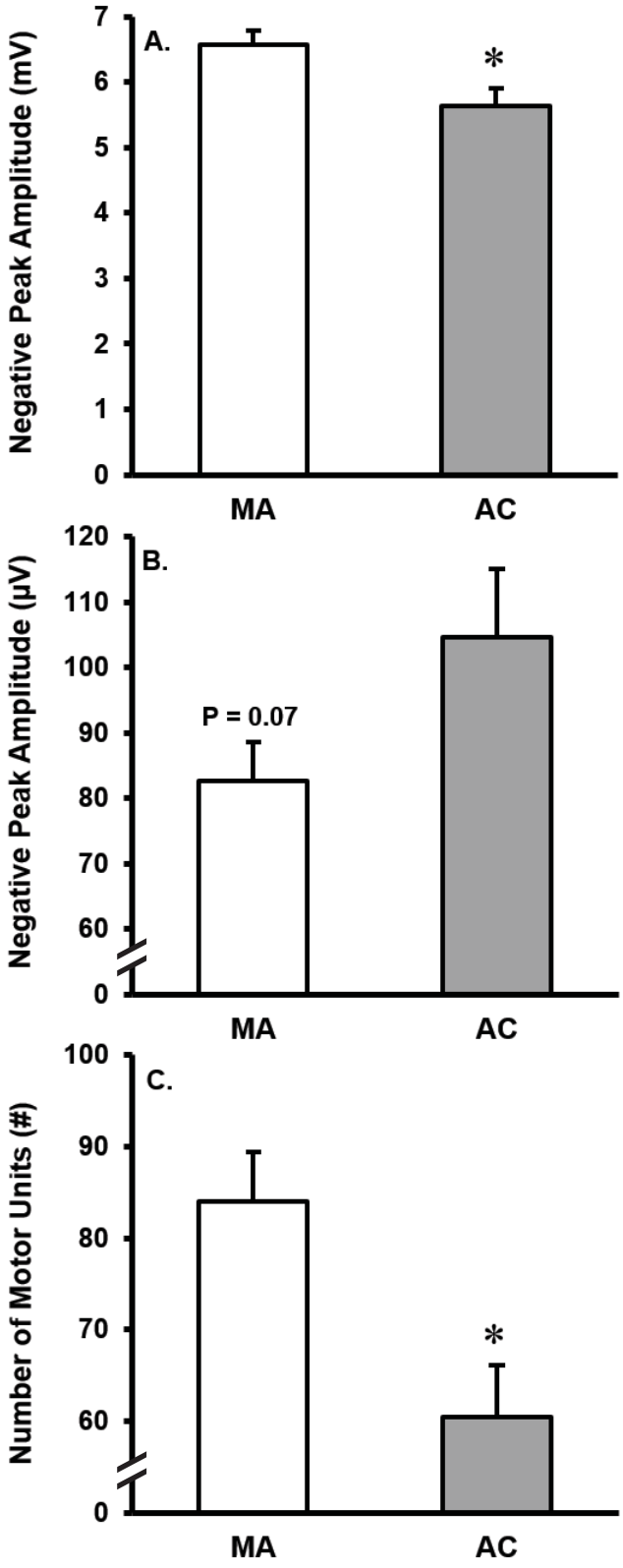
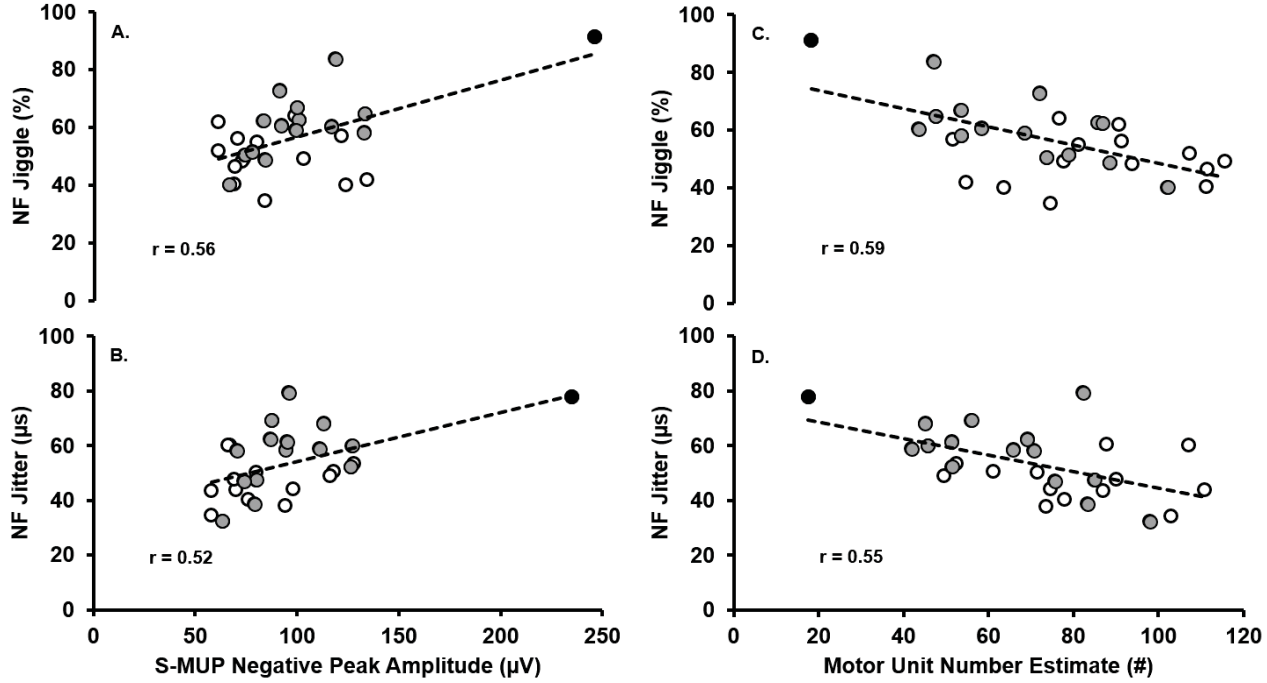

A.
Negative Peak Amplitude (mV)
7
6
5
4
3
2
1
0
*
MA
AC
B.
Negative Peak Amplitude (µV)
120
110
100
90
80
70
60
0
P = 0.07
MA
AC
C.
Number of Motor Units (#)
100
90
80
70
60
0
*
MA
AC

Figure 3


A.
B.
C.
D.
NF Jiggle (%)
NF Jitter (µs)
r = 0.56
r = 0.52
r = 0.59
r = 0.55
S-MUP Negative Peak Amplitude (µV)
Motor Unit Number Estimate (#)