Reduced motoneuron excitability and common synaptic inputs of the aged human vastus lateralis

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Abstract

The age-related loss of neuromuscular function has been comprehensively documented with a disproportionate loss in the quadriceps compared to other muscle groups. Although muscle atrophy may partially account for strength decline, it is further influenced by alterations of the nervous system. The outputs of motoneurons are heavily dependent on their intrinsic excitability and the level of common synaptic inputs they receive. However, these have not yet been described in aged human quadriceps.

High density surface electromyography (HDsEMG) signals were collected from the vastus lateralis of ten young (mean±SD, 24 ± 6.2 y) and ten older (66 ± 7.7 y) men during a 20-s ramp contraction peaking at 20% of maximal voluntary isometric contraction. EMG signals were decomposed to identify individual motor unit discharges, and motoneuron persistent inward current (PIC) amplitude and intramuscular coherence were estimated.

Older participants produced significantly lower peak knee extensor torque ($p<0.001$) and poorer force control ability ($p=0.002$) than young. Older participants also had lower peak discharge rate ($p=0.016$), PIC amplitudes ($p=0.001$) and coherence estimates in the beta frequency band in both ascending ($p=0.026$) and descending phases ($p=0.015$) when compared to young.

These data provide new evidence that intrinsic motoneuron excitability and common synaptic input is lower in the vastus lateralis of older males when compared to young, and highlights the feasibility of this method in the clinically and functionally important

quadriceps, further underpinning age-related loss of force control occurring independent of the loss of muscle mass.

Key Words: Ageing; motor unit; persistent inward current; common synaptic input; vastus lateralis

Key Points

- The age-related loss of muscle mass is exceeded by the loss of function, which is influenced by structural and functional alterations of the nervous system.
- Motoneuronal persistent inward currents and common synaptic inputs play an important role in the activation of motor units and subsequent force generation and control ability.
- Here we show reduced estimates of persistent inward currents and lower common synaptic inputs to older vastus lateralis, potentially contributing to observed lower strength and poorer force tracking.
- These findings highlight decrements of the aged human motor system that act independently of muscle atrophy in functionally relevant muscle groups which should be considered in the application of interventions targeting aged human muscle.

Introduction

Human ageing is characterised by a progressive reduction in muscle size and muscle strength, resulting in an impairment of force generation, force control and physical performance (Manini *et al.*, 2013; Wilkinson *et al.*, 2018; Pethick & Piasecki, 2022), all of which can negatively impact quality of life (Daley & Spinks, 2000; Smee *et al.*, 2012). The loss of muscle size is explicable by the combined effects of muscle fibre atrophy and muscle fibre loss (Wilkinson et al. 2018), and although atrophy may partially account for declines in muscle strength, it is further influenced by structural and functional alterations of the nervous system.

The voluntary contraction of muscle fibres occurs via net excitatory signals to the motor units (MUs); groups of muscle fibres innervated by a single motoneuron (Heckman & Enoka, 2012). The central nervous system regulates muscle force via two primary strategies; varying the number of MUs recruited and varying the discharge rate of each motor neuron, both of which are susceptible to age-related alterations. Substantial motoneuron loss accelerates in the sixth decade of life, resulting in fewer MUs, and a compensatory process of MU remodelling further results in larger MU innervation ratios (Hepple & Rice, 2016; Piasecki *et al*., 2016).

Additionally, although this appears to be muscle specific, an age-related decrease in MU discharge rate has also been documented at normalised contraction levels (Kirk *et al.*, 2021; Orssatto *et al.*, 2022) as well as being associated with circulating hormone levels that are also affected by age (Guo *et al.*, 2022*b*).

Beyond releasing neurotransmitters into postsynaptic ligand-gated channels at the spinal cord level via ionotropic mechanisms, activation of the motor nervous system also relies on neuromodulation (Heckman *et al*., 2005). This is mediated by intracellular second messenger systems that modify the voltage- and ligand-gated channels of neurons and alters several properties from their level of excitability to their pattern of firing in response to a given input (Heckman *et al.*, 2008*b*; Nadim & Bucher, 2014). This process is enhanced by monoamines, e.g. serotonin and norepinephrine released from the brainstem nuclei by stimulating inward-flowing persistent calcium and sodium currents (Heckman *et al*., 2005). As long as the membrane potential remains above the activation threshold, persistent inward currents (PICs) tend to remain activated (Heckman *et al.*, 2005), amplifying the firing behaviours of motoneurons to synaptic inputs by three to five fold (Binder & Powers, 2001) as well as generating long-lasting plateau potentials which results in self-sustaining firing (Gorassini *et al.*, 2002). A reduction in PIC amplitude could result in a reduction of force and an increase in perceived effort due to suppression of MU discharge rates. Even though PIC amplitudes appear to decrease with age in upper (Hassan *et al.*, 2021) and lower limb muscles (Orssatto *et al.*, 2021), PICs can be mediated by local ionotropic inhibitory inputs that may change differently with ageing between muscle groups, particularly those susceptible to functional losses such as the vastus lateralis (VL).

Rather than controlling the discharge pattern of individual MUs, the central nervous system controls the excitatory inputs to the MU pool, referred to as common synaptic inputs (Farina *et al.*, 2014) which are defined as the proportion of net synaptic input correlated between MUs. Force fluctuations are largely determined by the alterations in low frequency components of common synaptic inputs to MUs, which is reflected in concurrent fluctuations in MU discharge rates from the same MU pool (Farina & Negro, 2015). Accordingly, a decrease in force steadiness associated with ageing is a consequence of an increase in variance of common synaptic inputs (Castronovo *et al.*, 2018; Feeney *et al.*, 2018). However, the variability of common synaptic inputs in

quadriceps has only been explored in healthy young individuals (Avrillon *et al.*, 2020; Rossato *et al.*, 2022), with no available data from older humans.

It is not possible to make intracellular recordings from human spinal motoneurons in order to estimate PICs, so alternative methods are employed which distinguish intrinsic excitability of the MU from its descending synaptic drive by using unit-wise MU analysis technique (Hassan *et al*., 2021). In view of the advances with which MU discharging patterns can be measured, it provides an opportunity to investigate and better understand the physical properties and the underlying mechanisms which may impair motor control in older age. Therefore, the aim of this study was to explore the differences in physical performance as well as the level of neuronal excitability and common synaptic input in the clinically relevant VL between healthy young and older males.

Methods

Participants and ethical approval

26 healthy male participants (11 young and 15 older) provided informed consent to take part in the study, which was approved by the University of Nottingham Faculty of Medicine and Healthy Science Research Ethics Committee (90-0820, 199-0221) and conformed to the Declaration of Helsinki. In order to ensure a sufficient number of MU pairs for PIC amplitude estimation, 20 participants were included; 10 young (mean ± SD; age, 24 ± 6.2 y; body mass index, 24.5 ± 2.8 kg/m^2) and 10 older (age, 66 ± 7.7 y; body mass index, 25.5 ± 2.6 kg/m^2) males. All participants were recruited through advertisements in the local community. Prior to enrolment, all participants completed a comprehensive clinical examination and metabolic screening was conducted at the School of Medicine, Royal Derby Hospital Centre. All recruited participants were recreationally active and free from any metabolic disease, lower limb musculoskeletal abnormalities, neurological disorder, cardiovascular disease and uncontrolled hypertension.

Muscle cross sectional area

Ultrasound was used to measure the cross-sectional area (CSA) of the VL muscle of the right leg, using an ultrasound probe (LA523 probe, B-mode, frequency range 26–32 Hz, and MyLabTM50 scanner, Esaote, Genoa, Italy) at the anatomical mid-point of the right thigh. Images were recorded with the participants laying in a supine

position and the mid-point obtained from the identification of the greater trochanter at the top of the femur and the mid-point of the patella. Panoramic imaging (VPAN) images were obtained in a medial to lateral fashion, beginning and ending the capture of the image at the aponeurosis borders of the VL of the right leg. The produced images were subsequently analysed using ImageJ software (National Institute of Health, USA) through tracing around the contour of the aponeurosis via polygon function to quantify the area of the VL. Each image was analysed three times and the mean value of three images was accepted as CSA.

Muscle strength and force tracking accuracy

The participants were required to attend the lab at 0900 after an overnight fast and abstain from strenuous exercise 24h prior to the testing sessions. Participants were seated in a custom-built chair with hips and knees flexed at ~90 degree. The lower leg was secured to a force dynamometer with noncompliant straps above the medial malleolus. To minimize the movement of the upper trunk during testing, a seat belt was fastened across the pelvis of the participant. After a standardised warm up through several submaximal contractions (Guo *et al.*, 2022*a*), participants were instructed to contract as hard and fast as they could – to the point where the holding of the contraction is mechanically not possible. During the trial, participants were not allowed to hold onto the side of the chair and were asked to cross their arms across the chest. This was performed with real-time visual force feedback on a monitor placed in front of the participant, with verbal encouragement to aid in producing maximal effort. This was repeated two to three times further, giving 60 seconds of rest between attempts standardised recovery. If the difference between the last 2 attempts was < 5%, the highest value recorded in newtons was taken and accepted as the maximal isometric voluntary contraction force (MVC). Peak torque was calculated by multiplying MVC by the lever arm length from the knee to the centre of the ankle strap.

Following a short rest, participants performed four triangular shaped contractions (10s up and 10s down) to 20% of their MVCs with 30s rests. All force targets were set using OTBioLab software (OT Bioelecttronica, Turin, Italy). Ramp contractions to 20% MVC have been widely used for PICs estimation (details below) (Gorassini *et al*., 2002; Hassan *et al*., 2020; Afsharipour *et al*., 2020). To assess force tracking accuracy, all data were exported to Spike2 (version 9.11), where a virtual channel

was created by subtracting the performed path from the requested (target) path and rectifying it. Area under the curve reflects the level of deviation from the target line reflecting muscle force tracking accuracy, with higher values indicating greater deviation.

High-density surface electromyography

A semi-disposable high-density surface electromyography (HDsEMG) array (64 electrodes, 13×5, 8mm, I.E.D., GR08MM1305, OT Bioelettronica, Inc., Turin, Italy) was placed with the orientation of the muscle fibres (proximal to distal) over the muscle belly of right vastus lateralis after skin preparation involving shaving, light abrasion and cleansing with 70% ethanol. Electrodes were secured to the skin using flexible tape. The adhesive grids were attached to the surface of the muscle by disposable bi-adhesive foam layers (SpesMedica, Bettipaglia, Italy). The skin electrode contact was facilitated by filling the cavities of the adhesive layers with conductive paste (AC Cream, SpesMedica). A strap ground electrode (WS2, OTBioelettronica, Turin, Italy) dampened with water was positioned around the ankle of the right leg to optimise the signal quality. HDsEMG signals were acquired in a monopolar configuration, amplified (x256) with filtering set at 10-500 Hz and digitally converted at 2000 Hz by a 16-bit wireless amplifier (Sessantaquattro, OTBioelettronica, Turin, Italy) and transferred to a PC for further offline analysis.

The recorded HDsEMG signals were converted into a MatLab file format with only one contraction in a single data file and then analysed offline using DEMUSE software in MatLab. Each participant performed four ramp contractions; however, a single trial was selected for analysis based on the force tracking accuracy and the quality of MU discharge profiles. Monopolar HDsEMG signals were band-pass filtered at 20-500Hz and then decomposed into MU pulse trains by a convolutive kernel compensation algorithm (Holobar & Zazula, 2007; Holobar *et al.*, 2014). After the decomposition, a trained investigator manually inspected the MU spike trains and edited the discharge patterns of each MU detected. Only MUs with a pulse-to-noise ratio equal or greater than 30 dB were kept for further analysis.

Estimation of persistent inward current amplitude

The instantaneous firing rates of both MUs were calculated as the inverse of the interspike intervals of each MU spike train and smoothed by fitting a fifth order

polynomial function. An estimate of the PIC amplitude can be derived from the well-established unit-wise MU analysis technique (Hassan *et al.*, 2021). The lower threshold unit to be recruited in the ramp contraction is commonly referred to as the "control" unit; the "test" unit is a unit of higher threshold. Delta F (estimates of PIC amplitudes) was calculated as the difference in "control" unit firing rate between the onset and offset of a "test" unit (Gorassini *et al*., 2002). This technique relies on several assumptions: 1) test and control units share the common synaptic drive; 2) PIC is activated before or at the recruitment; 3) firing rate of the control MU closely reflects the depolarizing input to the parent motoneuron; 4) test and control unit process the synaptic inputs in a similar way. The MU pairs for the delta F calculation were selected based on the following criteria: 1) as a measure of common synaptic modulation, rate-to-rate correlation $r \geq 0.7$; 2) to avoid the high variability in delta F calculation due to the initial acceleration phase of MU firings, test units were recruited at least 1s after the control units; 3) to account for the possibility of control unit saturation leading to an underestimation of delta F, pairs in which rate modulation of the control unit fell within 0.5pps were removed from analysis (Gorassini *et al*., 2002; Udina *et al*., 2010; Vandenberk & Kalmar, 2014; Hassan *et al*., 2020). An average delta F was calculated when individual test units were paired with multiple control units (Figure 1). Subsequently, a single delta F was determined from each participant in VL by averaging all the delta F values obtained for all the units from each participant. Peak discharge rates were measured as the highest value in fifth order polynomials for each MU.

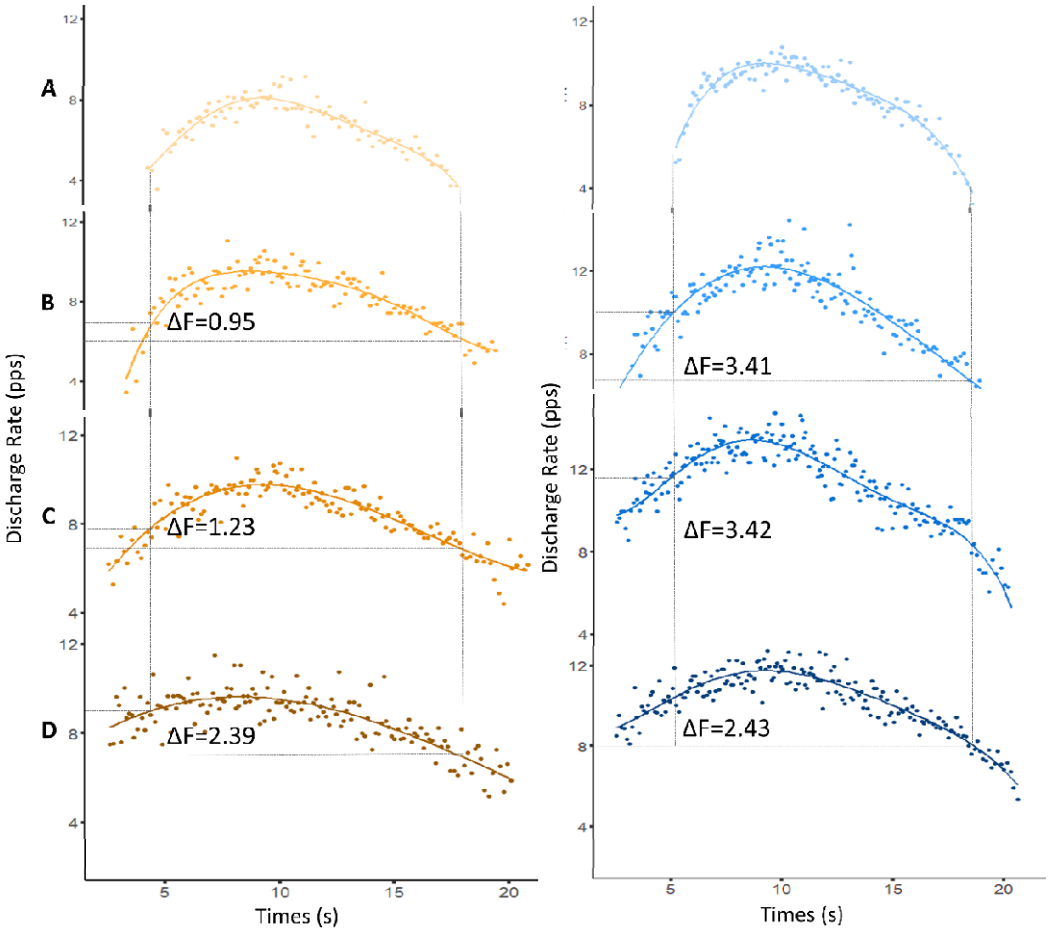

Figure 1. Example data showing delta frequency (ΔF) calculation in vastus lateralis for paired-motor unit technique. Paired motor unit data from a young male is displayed in the left panels (mean ΔF=3.09) and from an older male in the right panels (mean ΔF=1.52). Test units are displayed in panel A and corresponding control units are in B, C and D.

Coherence Estimates

To estimate the level of common synaptic input, intramuscular coherence was calculated during ascending and descending phase of a ramp contraction separately (Figure 2), which represents a frequency domain correlation between cumulative spike trains (CSTs) of the identified motor units. The magnitude-squared coherence was calculated using the Welch's averaged periodogram with non-overlapping Hann windows of 1s at the different frequency bands: delta (0-5 Hz), alpha (5-12 Hz), beta (15-30 Hz) and piper (40-50 Hz) (Raethjen *et al.*, 2007; Laine & Valero-Cuevas, 2017). This analysis was repeated for 60 randomly chosen combinations of two equally sized CSTs calculated as the sum of firing times from three MUs randomly selected from the same set of MUs and then averaged. The Fisher Z-transform was

applied to the coherence estimates to obtain a normally distributed variable for comparisons.

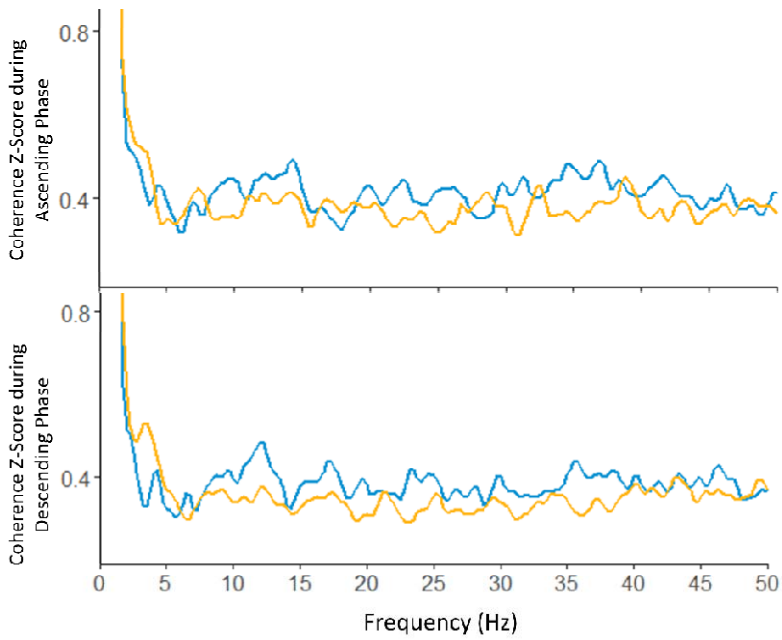

Figure 2. The group mean intramuscular coherence Z-Scores during ascending and descending phase of a ramp contraction at 20% maximal isometric voluntary contraction in young (blue) and older (yellow) participants. The magnitude-squared coherence was calculated from all the combinations (a maximum of 60 random permutations) of two equally sized cumulative spike trains, each composed by three motor units.

Statistical analysis

Data management and analysis were performed using RStudio (Version 2022.07.1). Descriptive data were generated for all variables. *An independent t-test* was conducted to test whether any differences existed between young and older males in torque, force tracking accuracy and coherence estimates. In order to preserve variability within and across participants simultaneously to the greatest extent, *multilevel linear regression models* were generated to compare peak discharge rate, delta F, recruitment threshold and derecruitment threshold between groups using *lme4 package* (Version 1.1-27.1). Recruitment threshold was included as a covariate in the model to explore peak discharge rate and discharge rate range. In the multilevel models, single MU was regarded as the first level; and individual

participant with clustered MUs was considered as the second level. To better show the magnitude of the standardized mean difference between groups, the absolute difference was also calculated by *Hedges' g effect size (ES)*. *Simple linear regression models* were used to explore the relationship between age, peak discharge rate and delta F. For data visualization, individual participant means were displayed in box-and-jitter plots. A *p* value <0.05 was considered significant.

Results

During the ramp contractions peaking at 20% of MVC, a total of 197 MUs were identified from the right VL of 10 young males and 175 MUs from the right VL of 10 older males. For Delta F calculations, there was an average of 5 (SD: 3.68) pairs of test and control units per person in the young and 6 (4.95) pairs in older group, with the mean number of 4 (1.79) test units per person in young and 3 (1.97) in old.

There was no statistical difference in muscle cross sectional area (Y v O: 29.22 ± 4.88 v 23.59 ± 7.88 cm^2, ES = 0.82, p=0.071; Figure 3A), however the younger group had greater muscle strength (279.60 ± 84.77 v 153.50 ± 38.06 Nm, ES = 1.84, p<0.001; Figure 3B) when compared to the older group. There was a significant difference in force tracking accuracy between groups, with the younger group performing better (9.38 ± 3.88 v 17.54 ± 7.01 Ns, ES = -1.38, p=0.005; Figure 3D) during a whole ramp contraction. When contraction phases were explored separately, there was no group difference in the ascending phase (5.13 ± 3.12 v 7.32 ± 3.10 Ns, ES = -0.67, p=0.132; Figure 3E), yet younger participants showed better force control ability than the older participants in the descending phase (4.27 ± 1.03 v 10.26 ± 5.03 Ns, ES = -1.58, p=0.002; Figure 3F).

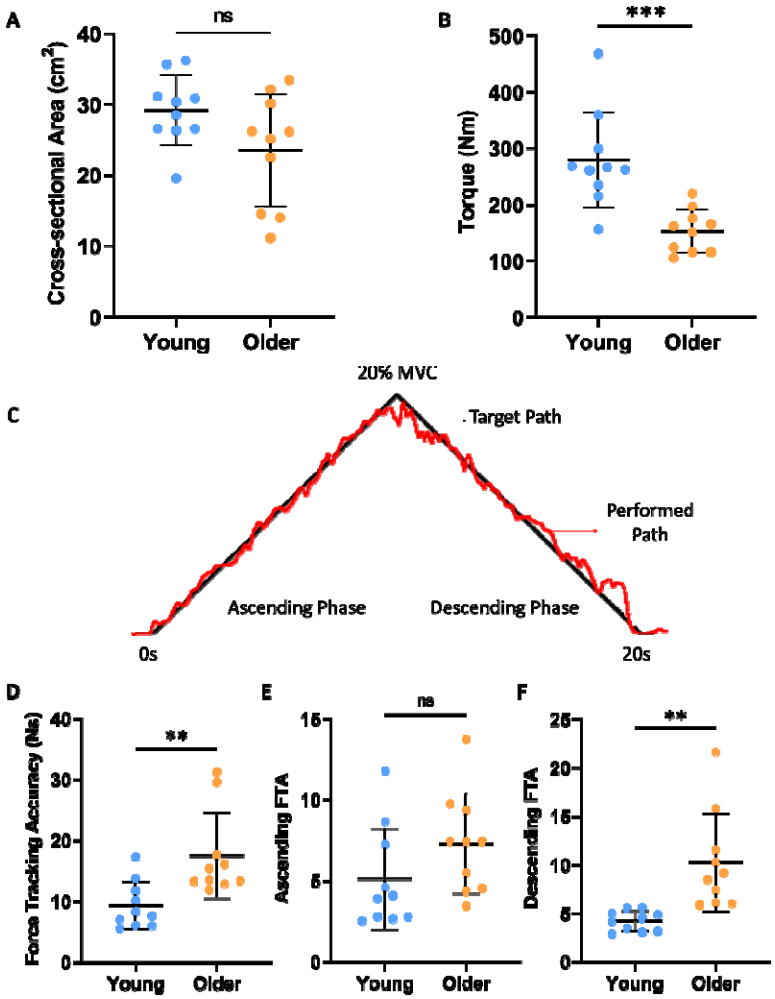

*Figure 3. Scatter plots with means and SDs showing the differences in cross-sectional area (A) and torque (B) between young (blue) and older (yellow) males; during a single ramp contraction at 20% of maximal voluntary contraction (C), the differences in force tracking accuracy during whole contraction (D) as well as during ascending phase (E) and descending phase (F). Note: ***p<0.001, **p<0.010, *p<0.05.*

The younger group had significantly higher peak discharge rates (10.64 ± 1.59 v 9.60 ± 1.34 pps, ES = 0.67, p=0.016; Figure 4A) and delta F values than the old (3.11 ± 0.77 v 1.86 ±0.65 pps, ES = 1.68, p=0.006; Figure 4B). In each group there were no significant relationships between delta F and peak discharge rate (Y: p=0.694; O: p=0.844) or between age and delta F, or between age and peak discharge rate (all p>0.05).

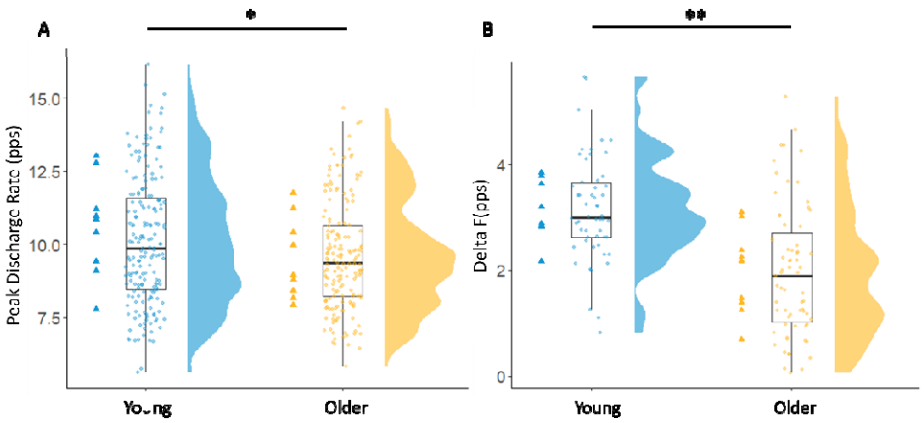

*Figure 4. Box-and-jitter plots of the individual participant mean value and their corresponding rainclouds plots of peak discharge rate (A) and Delta F (B) in young (blue) and older (yellow) males. Note: ***p<0.001, **p<0.010, *p<0.05.*

Younger participants had a greater mean discharge rate range in both ascending (3.57 ± 0.70 v 3.51 ± 0.81 pps, ES = 0.07, p=0.029) and descending (5.38 ± 1.22 v 4.88 ± 0.84 pps, ES = 0.46, p=0.022; Figure 5A) phases of the ramp contraction.

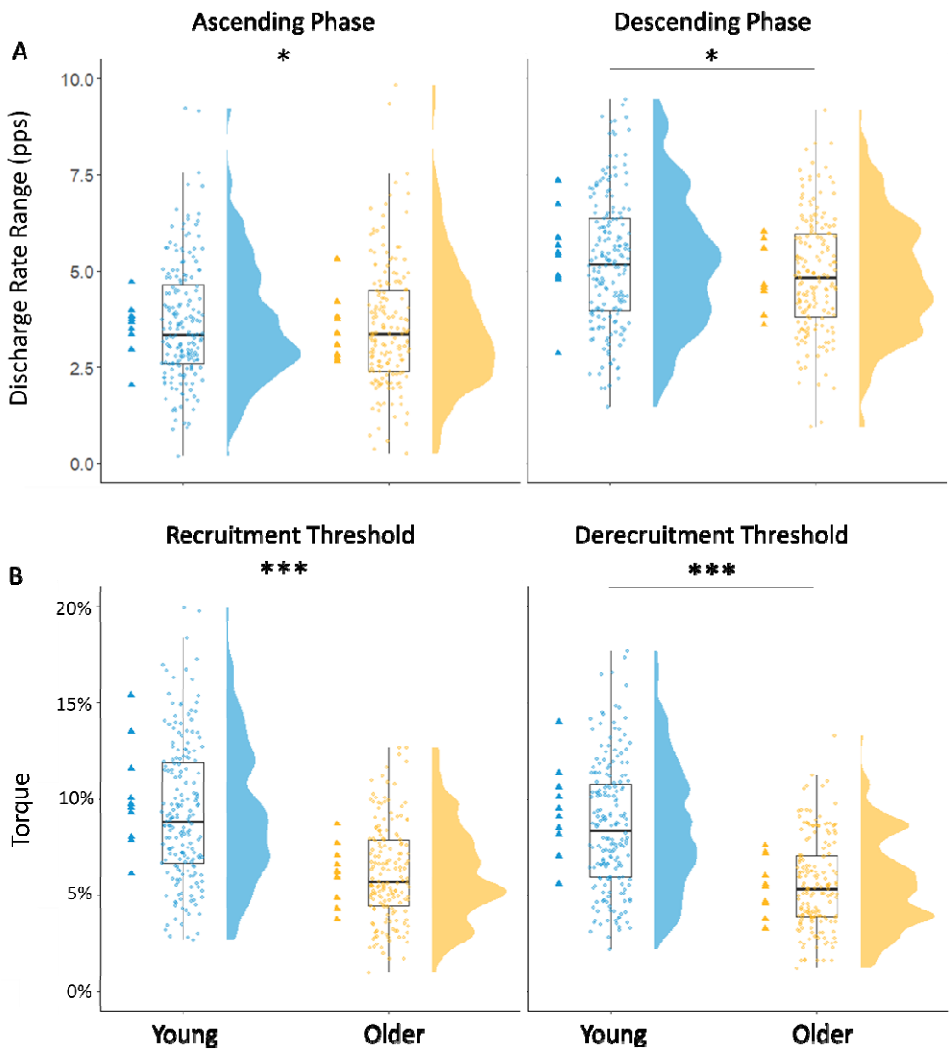

*Figure 5. Box-and-jitter plots of the individual participant mean value and their corresponding rainclouds plots of discharge rate range (A) and recruitment threshold and derecruitment threshold (B) in young (blue) and older (yellow) males. Note: ***p<0.001, **p<0.010, *p<0.05.*

There was a significant difference in MU recruitment threshold between young and older groups (Figure 5B). The mean recruitment threshold for old occurred at 6% MVC, significantly lower that the threshold for young, which was 10% MVC (p=0.001). During the descending phase of the contraction, significantly longer discharging of the activated MUs were observed in older group (p<0.001). The mean derecruitment threshold of the activated MUs in younger group occurred at 9.4% of MVC, with a mean derecuritment threshold at 5.6% of MVC in the older group.

There was no significant age-related difference in the COH Delta, Alpha or Piper bands in either ascending or descending phase of the ramp contraction (all p>0.05; Figure 6). Younger participants had greater coherence estimates in the beta frequency band in both ascending (p=0.026) and descending phases (p=0.015) when compared to older participants.

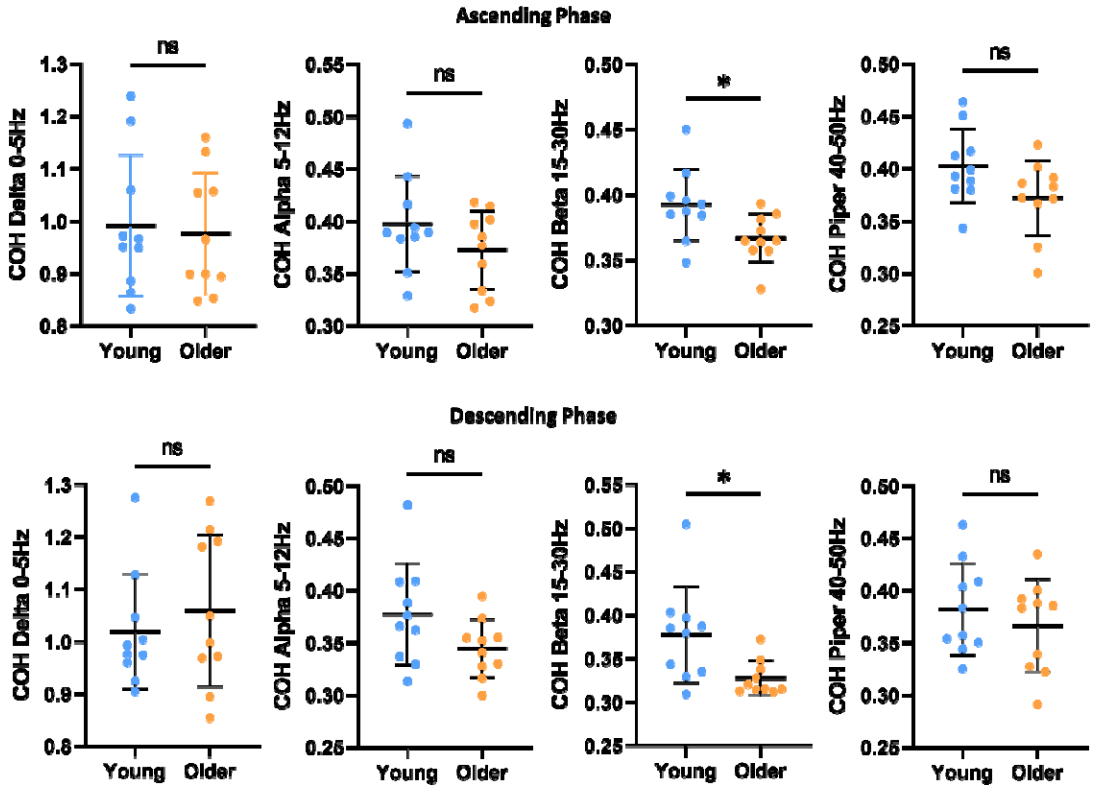

Figure 6. Individual participant means of intramuscular coherence Z-score at different frequency bands during ascending and descending phases of a ramp contraction at 20% maximal voluntary contraction in young (blue) and older (yellow) males.

Discussion

Here we provide the first evidence for reduced neuronal excitability in the aged human VL, a muscle with established susceptibility for age-related functional losses. Our findings indicate that younger individuals have better force control ability during a ramped contraction when compared to older individuals, and notably, this was explained by an inferior tracking ability during the descending phase in the older group. Older participants had lower peak discharge rate, smaller discharge rate range, and lower mean recruitment and derecruitment thresholds compared to young.

We also show age-related differences in common synaptic input to the VL, with a reduced commonality during both ascending and descending phases. These findings highlight important aspects of impaired neural input to older muscle that should be considered in the field of neuromuscular ageing.

The older participants in this cohort were healthy and living independently which may partly explain the lack of a statistically significant (although an ES of 0.87) age-related difference in muscle size, which is not consistent with the wider ageing literature (Piasecki *et al*., 2018). However, the greater difference in muscle strength is consistent with numerous previous studies (e.g. Maden-Wilkinson et al. 2013; Piasecki et al. 2016) and highlights explanatory factors of strength loss beyond total muscle size. Alongside the lower strength and MU discharge rate, reduced force control ability in older participants was also observed during the descending phase of a ramp contraction. Numerous factors associated with advancing age may plausibly explain this poorer force control, including MU remodelling, reduced excitability of the MUs, decreased muscle spindle sensitivity, less common synaptic inputs, and/or impaired cutaneous afferents (Mynark & Koceja, 2001; Shaffer & Harrison, 2007).

Estimates of MU number in the VL are reduced in older age (Piasecki *et al.*, 2016*b*, 2019), and although direct human evidence is lacking, animal models show large MUs are preferentially lost with ageing and denervated fibres are reinnervated by early recruited MUs (Kadhiresan *et al.*, 1996), resulting in expansion of those earlier recruited MUs such as those sampled in the current study (<20% MVC). These MUs with a larger number of fibres, when activated, would have a greater influence on force during their progressive re/derecruitment processes.

The poorer force control ability in the older group could partially be a result of neuronal atrophy. Several studies have reported a decrease of grey matter and white matter throughout the motor cortex with increasing age (Marner *et al.*, 2003; Salat *et al.*, 2004; McGinnis *et al.*, 2011) and a strong correlation between cortical atrophy and fine control capacity (Kennedy & Raz, 2005). A significant decrease in inhibition has also been observed in aged muscles, as well as a greater level of cortical and subcortical area activity during a targeted motor control task (Shinohara *et al*., 2003; Marneweck *et al*., 2011; Plow *et al*., 2013). In the absence of the ability to inhibit appropriate cortical areas during targeted motor tasks, MUs of older people may be activated in an inappropriate manner relative to the task.

Proprioception is an essential aspect of functional muscle contraction, and again, this is impaired in older age which can negatively impact balance (Ferlinc *et al.*, 2019). Both animal and human studies have revealed a decrease proprioception in aged muscles with the evidence of declined muscle spindle numbers and degeneration of the sensory nerve terminals, leading to the morphological adaptations at a peripheral level and modulation of mechanoreceptor gain at a central level (Winarakwong *et al*., 2004; Macefield & Knellwolf, 2018). Through inhibitory interneurons, inhibitory spinal circuits can affect descending inputs as well as Ia afferents from muscle spindles to spinal motoneurons (Kido *et al.*, 2004). Older participants exhibited a greater loss of Ia afferent feedback (Scaglion*i et al*., 2003) and an impaired ability to modulate Ia presynaptic inhibition (Baudry *et al.*, 2010). Thus, compared to younger individuals, it is probable the older participants were less able to sense force rate changes during the contraction.

Although not directly assessed, the points made thus far are supported by the intramuscular coherence findings during ascending and descending phases. Coherence in alpha band (5-12 Hz) is associated with Ia afferent feedback (Mehrkanoon *et al.*, 2014) and when exploring the differences between ascending and descending phases, a significant lower coherence estimate in alpha band was observed in old only, corresponding to the poorer control ability. Beta band (15-30 Hz) differences between age groups in both ascending and descending phases may reflect the reduced descending inputs from the motor cortex in older group (Walker *et al.*, 2019), suggesting that corticospinal tract function was compromised and there may be neural remodelling process occurring for performance regulation in older participants (Dodd *et al.*, 2017). Finally, age-related deterioration of cutaneous afferent inputs may also contribute to reduced force control ability, which is reduced in older adults (Peters *et al.*, 2016). Therefore, a decreased ability to regulate the force may also be a result of the combination of reduced Ia afferent feedback from muscle spindles and reduced cutaneous afferents.

MUs in older participants showed lower recruitment and derecruitment thresholds; put simply, MUs in older individuals were activated at a lower force level when compared to the younger participants and remained activated for longer. This may be explained by the loss of higher-threshold MUs in older participants, leading to a larger proportion of lower-threshold MUs in the MU pool (Piasecki et al. 2016). It is

widely accepted that in smaller muscles such as those in the hand, all MUs are recruited by 50% MVC with forces above that level achieved via increases in discharge rates (De Luca *et al.*, 1982). In larger muscles, recruitment continues up until approximately 90% MVC and MU discharge rate facilitates force increases beyond that (Masakado, 1994). However, MUs undergo remodelling during ageing, and it is possible that the force regulation strategy is altered with a preferential recruitment of MUs at lower relative contraction levels to compensate.

Compared to younger participants, we report a significantly reduced delta F in the VL of older participants during a ramp contraction. Supportive of studies of other muscles (Hassan *et al.*, 2021; Orssatto *et al.*, 2021), the current findings in VL showed that the intrinsic synaptic excitation was lower in old, which is likely attributable to the reduced activity of the monoaminergic system. Serotonin (5-HT) and norepinephrine (NE) appears primarily to be involved in the modulation of motor control and sensory input, partly influencing the magnitude of PIC amplitude (Lee & Heckman, 1998). Ageing is associated with decreased levels of 5-HT receptors and transporters in human brain (Karrer *et al.*, 2019; Deza-Araujo *et al.*, 2021) as well as impaired NE synthesis and secretion in older participants (Shibata *et al.*, 2006), leading to a reduced monoaminergic neurotransmission in healthy ageing. Additionally, the magnitude of PICs may also be influenced by age-related alterations in Na^{+} and L-type Ca^{2+} ion channels, widely distributed on the dendrites of motoneurons. The deregulation of Ca^{2+} has been reported to have an essential role in the development of ageing and the neurodegeneration process via C. elegans models (Alvarez *et al.*, 2020), which may also be apparent in human spinal motoneurons. A high sensitivity of PIC amplitude to inhibitory input to motoneurons has been reported (Heckmann et al. 2005) and as discussed above, inhibitory inputs significantly decrease with advancing age at both central nervous system and spinal cord levels. The reduced PICs in older participants here may be a result of increased inhibitory input levels. Furthermore, the sex-based differences of PIC magnitude recently observed (Jenz *et al.*, 2023) may be explained by hormonal differences which influences 5-HT, that may similarly differ between young and older males.

Strengths/Limitations

The main strength of this study was this is the first study investigating the common synaptic inputs and intrinsic excitability properties in aged VL between young and older humans. However, there are several limitations. Firstly, the results reported here were from males only which may not directly translate to females, as decreases in estrogen levels may also contribute to the alterations in circulating serotonin and its receptor densities in postmenopausal women (Gonzales & Carrillo, 1993). Secondly, we selected the best contraction among four contractions, which potentially ignored the age-related variability among and within participants. In addition, we used 10s signal length for calculating coherence estimates, possibly resulting in an underestimated effect of ageing on common synaptic inputs and intrinsic properties.

Conclusion

The reduced muscle strength and control ability observed in older males is partially attributable to impaired motoneuron excitability and reduced common synaptic input which occur independently of reductions in muscle size. These findings have important implications in the field of healthy human ageing and should be considered when applying interventions to mitigate age-related functional decrements.

Data Availability Statement

The datasets generated and analysed during the current study are available from the corresponding author upon reasonable request.

Competing interests

The authors have no competing interests to declare.

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