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Original

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1 **Regional recruitment and differential behaviour of motor units during postural control in**
2 **older adults**

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30 **Abstract**

31 Ageing is associated with neuromuscular system changes that may have implications for the
32 recruitment and firing behaviours of motor units (MUs). In previous studies, we observed that
33 young adults recruit sub-populations of triceps surae MUs during tasks that involved leaning in
34 five directions: common units that were active during different leaning directions, and unique
35 units that were active in only one leaning direction. Further, the MU sub-population firing
36 behaviours (average firing rate (AFR), coefficient of variation (CoV_{ISI}) and intermittent firing)
37 modulated with leaning direction. The purpose of this study was to examine whether older adults
38 exhibited this regional recruitment of MUs and firing behaviours. Seventeen older adults (aged
39 74.8 ± 5.3 years) stood on a force platform and maintained their center of pressure leaning in 5
40 directions. High-density surface electromyography recordings from the triceps surae were
41 decomposed into single MU action potentials. A MU tracking analysis identified groups of MUs
42 as being common or unique across the leaning directions. While leaning in different directions
43 did not affect the AFR and CoV_{ISI} of common units ($p > 0.05$), the unique units responded to the
44 leaning directions by increasing AFR and CoV_{ISI} , albeit modestly ($F = 18.51, p < 0.001$). The
45 unique units increased their intermittency with forward leaning ($F = 9.22, p = 0.003$). The medio-
46 lateral barycenter positions of MU activity in both sub-populations were found in similar
47 locations for all leaning directions ($p > 0.05$). These neuromuscular changes may contribute to
48 the reduced balance performance seen in older adults.

49 **New and Noteworthy**

50 In this study, we observed differences in motor unit recruitment and firing behaviours of distinct
51 sub-populations of motor units in the older adult triceps surae muscle from those observed in the
52 young adult. Our results suggest that the older adult central nervous system may partially lose the
53 ability to regionally recruit and differentially control motor units. This finding may be an
54 underlying cause in older adults balance difficulties during directionally challenging leaning
55 tasks.

56

57 **Keywords**

58 High-density surface electromyography, ageing, motor units, firing rates, postural control

59

60 **Introduction**

61 The capacity to control standing balance is a requirement for performing many daily life
62 activities and underlies the ability to maintain a mobile and independent lifestyle. Shifting the
63 body weight in different directions during standing is fundamental for more complex motor
64 tasks, and requires contributions from the musculoskeletal system, specifically the medial and
65 lateral gastrocnemius (MG and LG) as well as the soleus (SOL) muscles (1-3).

66 In healthy young adults, the performance of multi-directional leaning tasks in standing is
67 accomplished with minimal difficulty but with a large degree of complexity with regard to the
68 recruitment and firing behaviours the MUs. That is, in our previous studies (4, 5) we found that
69 two distinct two subpopulations of motor units (MUs) were recruited during a multi-directional
70 leaning task: *common* MUs were recruited from the MG and SOL, presumably to meet the
71 baseline torque requirements and as the force requirements for body stabilization changes,

72 *unique* MUs in separate locations were recruited in the MG and SOL muscles. In addition to the
73 recruitment location of the MUs, the distinct MU subpopulations exhibited different firing
74 behaviours, suggesting that the CNS may have the capability to control these subpopulations
75 separately. *Common* units were defined as MUs with waveforms that were indistinguishable
76 from each other when matched across at least two different leaning directions. In contrast, *unique*
77 units were MUs that could not be matched across leaning directions and had distinguishable
78 waveforms (4-6).

79 However, with ageing, there is an age-related loss of muscle mass (7) and associated
80 reduction in maximal muscle strength, power, and physical function, known as sarcopenia (8, 9),
81 which may influence postural control (10). While the causes of sarcopenia are multifactorial (11,
82 12), one of the primary causes of sarcopenia is the structural changes at the neuromuscular level.
83 During ageing, there is a loss of alpha motor neurons and degeneration of the peripheral nerves
84 (13-16). This process begins gradually, early in life, and accelerates more rapidly from 60 years
85 of age (17, 18), with more rapid declines in muscle function from 70 years of age (19-21). One
86 consequence of the loss of alpha motor neurons is the denervation of the muscle fibres that they
87 previously innervated (22). This can lead to a decreased number of muscle fibres, contributing to
88 the loss of muscle mass. However, the surviving motor neurons possess the ability to reinnervate
89 some of these recently orphaned fibres increasing the size of the innervated MUs proportional to
90 the number of fibres rescued (13, 23, 24). These enlarged MUs are considered to be a positive
91 adaptation as they compensate for declining motoneuron numbers and prevent muscle fibre loss
92 (25). However, a consequence of this phenomenon may be altered MU recruitment in the aged
93 neuromuscular system (26, 27). For example, recruitment thresholds are lowered and the
94 dependency of firing rate on recruitment rank (i.e. the onion skin behaviour) is compromised in

95 older adults (28). However, it remains unknown whether the MU remodelling process influences
96 the regionally-specific recruitment pattern previously observed in young adults (4). Regional
97 recruitment of MUs necessarily requires the muscle fibres belonging to a single MU to be
98 localized within a small region (29). It is plausible that during the remodelling process, the new
99 fibers reinnervated by the motor neuron are outside of the spatially localized region. If such a
100 pattern occurred, regionally specific recruitment of MUs may be attenuated or even lost.

101 The recruitment of MUs is also influenced by whether the synaptic current distributes
102 either uniformly or non-uniformly across the motoneuron pools of the triceps surae muscles. In
103 young adults, non-uniform activation of the motoneuron pool has been demonstrated during
104 standing balance (4, 30). In older adults, it has been observed that there is less localized activity
105 in the gastrocnemius during quiet stance, which may suggest a reduced ability to flexibly recruit
106 MUs in specific areas (31). Using coherence analyses, Semmler et al. (32) observed an increase
107 of common drive in older than younger adults, suggesting that older adults have decreased
108 flexibility in MU control. Flexible MU control is the ability of motor neurons to comply with
109 various task constraints. This includes selective voluntary activation of single motor units (33,
110 34), exceptions to the size principle (35, 36) in addition to differential firing behaviours of
111 distinct motoneuron populations (5). An overview of flexible MU control has been recently
112 published by Hug et al. (37). As a flexible control strategy is important for the independent
113 control of spinal motoneurons (37), it is possible that the decreased flexible control observed in
114 older adults may influence the MU firing behaviour observed during directional leaning.

115 It appears that, in the young adult, the central nervous system (CNS) acquires and uses
116 directionally specific force information to control balance in standing postural tasks. What
117 remains to be investigated is how older adults respond to directional force challenges during

118 standing balance. The present investigation, therefore, has two main goals: 1) to determine if
119 older adults regionally recruit MUs, and 2) to evaluate whether the MU firing behaviour in the
120 plantarflexor muscles of older adults modulated during a multi-directional leaning task. Due to
121 the denervation and reinnervation of MUs, we hypothesized that there would be less regional
122 activation of MUs in older adults than that seen previously in young adults. In addition, due to
123 the decrease in flexible neural control, we expected that there would be less evidence of
124 differential control of MU firing behaviours in older adults.

125

126 **Methods**

127 *Participants*

128 A total of 17 older adults (8 males and 9 females: mean \pm SD: age 74.8 ± 5.3 years, body
129 mass 75 ± 15.3 kg; height 171 ± 12.2 cm) participated in this study after providing informed
130 written consent. Exclusion criteria included individuals under the age of 70, the presence of co-
131 morbidity (specifically, known neuromuscular, neurological, or balance disorders), or lower limb
132 injuries within the past 6 months. This study conformed to the standards established by the
133 Declaration of Helsinki latest amendment and was approved by the Institutional Ethics
134 Committee.

135 *Experimental Protocol*

136 The experimental protocol has been described in more detail in previous studies (4, 5).
137 Briefly, participants were instructed to stand naturally in bipedal stance on a force platform
138 (OR6-6 Advanced Mechanical Technology, Watertown, MA, USA) in front of a screen
139 displaying their centre of pressure (CoP) (see Figure 1 in (4, 5)). Participants were allowed to
140 stand in their natural bipedal stance, which allowed them to choose their foot position

141 (approximately shoulder width apart). The position of the arms was instructed to remain at their
142 sides during the duration of the trial. First, a 30 s quiet stance trial was recorded for each
143 participant. Following that, each participant's limits of stability (LoS) were established by
144 conducting a 4-way LoS test (38). Five targets at 0°, 30°, 60°, 90°, and 120° (counter-clockwise
145 from lateral-right of the average CoP position computed during the quiet stance trial) were
146 calculated and placed along an ellipse corresponding to 60% LoS (Figure 1A). The same
147 procedure was completed for the 80% LoS. Given that the CoP distance from the ankle joint
148 scales with the ankle torque (39), the 80% LoS condition required a greater contraction intensity.
149 Only one target was displayed on the screen at any time and using visual feedback participants
150 were instructed to move in a smooth and controlled manner until their CoP position reached the
151 circular target and maintain the CoP position for 35s. A rest period of 15 s was provided between
152 different target directions. Figure 1B provides an example of a single participant's CoP trace
153 during the leaning protocol. The order of the target directions and LoS conditions was
154 randomized for each participant. The duration of each trial was approximately 45 s, 5 s towards
155 the target, 35 s within the target, and 5 s back to their natural bipedal stance. If a participant did
156 not maintain the instructed body position, the trial was repeated. Only the trials with correct
157 leaning posture were retained and analyzed. To familiarize themselves with the procedure
158 participants first performed an unrecorded practice trial.

159

160 *EMG Electrode Placement*

161 Using an ultrasound imaging system (LogicScan 64 LT-1T; Telemed, Vilnius, Lithuania)
162 as a guide, the High Density surface Electromyography (HD-sEMG) grids were placed on the
163 MG, LG and SOL (OTBIOELECTRONICA, Torino, Italy). Detection of the medial and lateral edges of

164 the gastrocnemius and SOL, the insertion of the MG and LG on the Achilles tendon, the fascial
165 space between the MG and LG, and the distal edges between the two muscles were identified
166 and marked on the skin with participants lying prone and the ankle in neutral position. The skin
167 was cleaned and abraded prior to grid placement. A single 32-channel semi-disposable grid
168 (interelectrode distance of 10mm) was placed over each of the MG and LG. The gastrocnemius
169 grids were placed in the proximal region of the muscle 10mm above the distal edges of the
170 superficial aponeurosis of the MG and LG. One 64 channel semi-disposable grid with an
171 interelectrode distance of 10 mm was placed over the SOL muscle centered on the Achilles
172 tendon, 10mm below the distal insertion of the MG. Bio-adhesive foam held the grids in place,
173 and conductive paste was used to ensure optimal electrode skin contact. Three reference
174 electrodes were placed on the patella (MG), the fibular head (LG) and the medial malleolus
175 (SOL).

176 In surface EMG, the amount of muscle volume sampled and the information one can
177 obtain depend on the arrangement of muscle fibers in relation to the skin rather than on the
178 muscle architecture itself. We strived to sample from the largest possible fraction of the muscle
179 volume, and thus represent the biggest possible number of motor units for the grid of electrodes
180 we used. Increasing sensitivity in surface EMG requires placing each electrode in the grid on a
181 different group of muscle fibers, ensuring minimal redundancy between different EMGs.
182 Ideally, greater spatial resolution would be obtained from the sampling of the physiological
183 cross-sectional area of a muscle, which is not possible from the skin surface. For in-depth
184 pinnate muscles, greater spatial resolution is ensured by distributing surface electrodes over
185 different locations covering the muscle aponeurosis (cf. Figures 2 and 3 in (40)). Specifically for

186 the soleus muscle, this would mean covering the muscle central region, over the Achilles tendon
 187 (41).

188

189 **Data Analysis**

190 *HD-sEMG Signal Analysis*

191 The HD-sEMG signal analysis has been described in detail in previous articles (4, 5).
 192 Briefly, HD-sEMG signals were collected in monopolar configuration. Signals were amplified
 193 1000 times using an amplifier (128-channel EMG-USB with OTbioLab software v.2.05;
 194 OTBioelectronica, Torino, Italy) and digitized at 2048 Hz with a 12-bit analog to digital
 195 converter. After assessing the quality of the signals, monopolar signals were filtered
 196 (Butterworth, 2nd order, 20-350 Hz) and decomposed using the blind source separation method
 197 (42) implemented in the DEMUSE tool software. The decomposition was done for each target
 198 direction and LoS condition separately, over the central 20s of the 35s period when the CoP was
 199 maintained at the target location. The results of the decomposition were inspected visually for
 200 spurious units with propagating potentials, noisy waveforms and those with a pulse to noise ratio
 201 below 28 dB were discarded (43, 44). An example of a retained MU is shown in Figure 2 (top).

202 To determine spatial locations of the decomposed MU, the barycenter was computed for
 203 each MU. The barycenter is quantified from the cluster of channels with an average rectified
 204 value (ARV) above 70% of the maximum ARV (See (4, 30, 45) for more detail). The location of
 205 the peak activity was calculated as:

$$Barycenter = \frac{\sum ARV_{ch} POS_{ch}}{\sum ARV_{ch}}$$

206 With 'ch' being each channel in the cluster of channels with amplitude above 70% of the ARV,
 207 and 'POS' being their position along either rows (medio-lateral) or columns (proximo-distal) on

208 the electrode matrix. The barycenter is the location on the muscle where the MU action
 209 potentials are more clearly represented.

210 The segmented activity maps of the HD-sEMG results in each channel representing a 1
 211 cm² area (Figure 2). The number of channels with an amplitude above 70% ARV for the
 212 barycenter calculation was computed for each MU. This value is indicative of the area of muscle
 213 fibres being activated and is referred as the area of MU from this point.

214 For each retained MU, the average firing rate (AFR) was calculated as the number of
 215 firing instances divided by the time the MU was active. Interspike intervals (ISIs) < 25ms were
 216 excluded as they would have a large impact on the computation of average firing rate. We did
 217 not include ISIs >250 ms in the AFR calculation, as these are considered a de-recruitment of the
 218 MU, which would be captured in the intermittency index (see below). The coefficient of
 219 variation of the ISI (CoV_{ISI}) was calculated as the standard deviation of the ISI divided by the
 220 mean ISI.

221 A previously-established intermittency index (5) was used to determine whether the MUs
 222 were activated intermittently. To standardize the amount of intermittent activity for a single MU,
 223 we calculated the degree of intermittency:

$$\text{Degree of Intermittency} = \frac{\text{Number of intermittent occurrences}}{\text{Time MU was active (s)}}$$

224 Where the “*Number of intermittent occurrences*” is the number of cases in which the MU
 225 firing rate drops below 4 Hz, and the “*Time MU was active*” is the total time the MU was
 226 actively recruited for. This procedure standardized the intermittency of MU activity and allowed
 227 comparisons between different MUs by evaluating the number of intermittent occurrences per
 228 second.

229 *Motor unit tracking*

230 To identify the subgroups of MUs, a MU tracking analysis was conducted separately for
231 the 60% and 80% LoS conditions. Common MU action potentials between different leaning
232 directions were identified. To identify common MUs, the retained MU ARV was calculated
233 across 30ms epochs centered on individual action potentials. The action potentials of each pair of
234 MUs were aligned in time by maximizing their cross-correlation function. The mean square
235 difference was calculated between the two sets of time-aligned action potentials, averaged across
236 channels, and then normalized with respect to the mean ARV of the two sets of MU. Finally,
237 pairs of MU action potentials with a mean square difference smaller than 10% were considered
238 common (4-6). The matching analysis provides a group of MUs that will be referred to as
239 *common units* and a group of MUs not paired across directions which will be called *unique units*
240 from this point on. See Figure 3 in (5) for more details.

241 *Centre of Pressure Analysis*

242 All CoP analyses were performed using MATLAB 2021b (The Mathworks, Inc., Natick,
243 MA, USA). CoP position in the antero-posterior and medio-lateral directions was calculated
244 from the force platform signals filtered with a fourth-order low-pass Butterworth filter (cut-off
245 frequency = 50 Hz) and sampled at 2048 Hz using a 16-bit A/D converter. The mean CoP
246 position, standard deviations in the X (medio-lateral) and Y (antero-posterior) directions, 95%
247 confidence ellipse (CoP ellipse area), and path length were calculated over the central 20s of the
248 35s period where the CoP was maintained at the target location. These parameters were used to
249 evaluate participant's balance performance during each direction of the leaning task.

250 In addition, the CoP was recorded on the HD-sEMG system to synchronize the EMG and
251 CoP signals. Signals were synchronized on the first, rising edge of CoP movement towards the
252 targets.

253

254 **Statistical Analysis**

255 All statistical analyses were conducted on R (v.5.12.1, R Development Core Team,
256 2009). Analysis of the effects of leaning direction, LoS, muscle, MU subgroup (unique or
257 common unit) and age group (young or older adult) on the medio-lateral and proximo-distal
258 barycenter, area of the MU, firing rates, CoV_{ISI} , and degree of intermittency were conducted
259 using separate linear mixed effects regression with the lme4 package (46). Data from the young
260 adults were included from Cohen et al. 2020 and 2022 (4, 5), which used the same experimental
261 protocol. A linear mixed effects approach was used to account for inter-individual differences in
262 the dependent variables. Starting with a maximal random structure, including by-participant and
263 by-item random slopes and intercepts, the structure was reduced to the optimal structure that
264 could be supported by the data. The concatenated models were compared with likelihood ratio
265 tests (LRT) to determine optimal model structure. This led to by-participant random intercepts
266 and random slopes for direction and muscles in all five models. We could not include random
267 slopes of leaning direction or LoS because of singular fit violations (i.e. multiple collinearities).

268 Diagnostic plots of the residuals from all models were inspected for violations of the
269 assumption of normality and homoscedasticity. The AFR and CoV_{ISI} and barycenter values met
270 the assumptions, however, the degree of intermittency did not (violations of homoskedasticity
271 and normality of residuals). The intermittency values were normalized by a $\log(x + 1)$
272 transformation for the data to fit the assumptions of the model.

273 Five linear-mixed effect models were conducted separately for the dependent variables
274 (medio-lateral barycenter, proximo-distal barycenter, AFR, CoV_{ISI} , and degree of intermittency).
275 Statistical significance of fixed effects was determined using type III Wald F tests with

276 Kenward-Rogers degrees of freedom with the ANOVA function from R's *car* package (v.
277 3.0.12). Effect sizes (Cohen's d) were calculated using the *lme.dscore* function from the R
278 *EMAtools* package (v. 0.1.4). We quantified small (Cohen's $d > 0.2$), moderate (Cohen's $d >$
279 0.5), and large effect sizes (Cohen's $d > 0.8$) with the conventional cut-off thresholds (Cohen
280 1988). Post hoc pairwise comparisons (with Bonferroni's correction) were performed using the
281 estimated-means contrast, as employed in R's *emmeans* package (v. 1.7.2). Post hoc comparisons
282 were applied between each target direction (all-pairwise comparisons), and within the same
283 target direction for the LoS condition, muscle group, and MU subgroup. Confidence intervals
284 around the parameter estimates were calculated via parametric bootstrapping with 5000
285 iterations. The post hoc results are reported with the mean estimate difference (M), 95%
286 confidence intervals (CI) and adjusted p -values.

287 To determine if the participant's balance performance during the leaning task differed
288 across the target directions and LoS conditions, the CoP parameters (CoP standard deviations in
289 the medio-lateral and antero-posterior directions, 95% ellipse area, path length, and mean
290 velocity) were analyzed in 5 separate linear mixed effect models. Diagnostic plots determined if
291 the CoP parameters met the assumptions for the linear mixed effects regressions. Using LRT we
292 determined the optimal model structure: using the fixed effect of directions and LoS, with the
293 random intercepts of subject. We could not include random slopes of leaning direction or LoS
294 because of singular fit violations (i.e. multiple collinearities). The same post hoc analysis used
295 for the MU firing parameters determined the differences across directions and LoS.

296

297 **Results**

298 The total number of identified motor units for each muscle per LoS condition in the older
299 adult group can be found in Table 1. Approximately 55% (1756/3175) of the decomposed MUs
300 in the older adults were determined to be common. Whereas the young adults had approximately
301 25% (892/3569) of their decomposed units determined as common (see Table 1 in Cohen et al.,
302 2020). A further breakdown of the proportion of common units decomposed within each muscle
303 by direction in the older adult (*See Figure 3*). To see a comparison between the young and older
304 adult data, please see the supplementary material, <https://doi.org/10.6084/m9.figshare.24352786>.

305 *Barycenter Locations*

306 Within the older adults, the multi-directional leaning task did not have an effect on the location
307 of the barycenter in medio-lateral direction for both unique and common units in all three
308 muscles at any of the leaning directions (p values ranging from 0.27 to 0.69). There was a
309 statistical difference in the proximal-distal position of the barycenters that was dependent on the
310 MU subgroup demonstrated by the interaction of Direction by MU Subgroup ($F = 2.745, p =$
311 0.02 , Cohen's $d = 0.31$). The unique units shifted more proximally during the 30° ($M = 27$ mm),
312 60° ($M = 33$ mm) and 120° ($M = 52$ mm) whereas there was no change in the position of
313 barycenter in the common units (Table 2). Between the older and young adult groups, there was
314 an Ageing effect that was dependent on the MU subgroup and Muscle as indicated by the Age
315 Group x MU Subgroup x Muscle interaction (Proximo-distal: $F = 11.837, p < 0.001$, Cohen's d
316 $= 0.54$; medio-lateral: $F = 3.864, p < 0.04$, Cohen's $d = 0.64$). Post hoc analysis determined that
317 the older adults unique units shifted significantly less than the young adults in both the medio-
318 lateral and proximo-distal directions during the 60° (proximo-distal: $M = 8$ mm; medio-lateral: M
319 $= 15$) and 120° (Proximo-distal: $M = 7.8$ mm; medio-lateral: $M = 25$ mm) directions of the MG
320 and SOL.

321 *Area of the MUs*

322 Within each of the age groups, the leaning direction did not have an effect on the area of
323 the MUs for both unique and common units in all three muscles (P values ranging from 0.56-
324 0.89). There was a significant effect of MU subgroup (young adults: $F = 3.42$, $p < 0.001$,
325 Cohen's $d = 0.56$; Older adults: $F = 2.76$, $p < 0.03$, Cohen's $d = 0.28$), with the unique units
326 occupying larger areas compared to the common units ($M = 4.8$ cm). The between group
327 differences of the older adults and young adults determined an ageing effect that was dependent
328 on the MU subgroup, indicated by the Ageing x MU subgroup interaction ($F = 5.67$, $p < 0.036$,
329 Cohen's $d = 0.55$). Post hoc results determined that the older adults common units occupied
330 larger areas compared to the younger adults in the MG ($M = 2.3$ mm), LG ($M = 3.4$ mm), and
331 SOL ($M = 3.2$ mm) muscles (Table 2). No other statistical differences were identified.

332 *Average Firing Rate*

333 Individual MU Average Firing Rates (AFRs) and associated MU distributions for the
334 MG, LG and SOL during the 5 leaning directions are depicted in Figure 3. The AFRs of the
335 common units for all plantarflexors are distributed unimodally (with a rightward skew) with a
336 peak of 8 - 10 Hz. Similarly, the AFRs of the SOL unique units were distributed unimodally
337 (with a rightward skew) but with a larger number of units firing at higher rates (15 - 25 Hz)
338 compared to the common units. The AFRs of the gastrocnemii unique units, conversely, had a
339 relatively uniform distributions across firing rates (10 - 25 Hz) without very distinctive peaks.

340 The estimates of the AFR for each muscle are reported in Figure 4. The AFR increased
341 across directions ($F = 18.51$, $p < 0.001$, Cohen's $d = 0.16$) dependent on the MU subgroup, as
342 indicated by the Direction x MU Subgroup interaction ($F = 5.61$, $p < 0.001$, Cohen's $d = 0.51$).
343 There were no directional effects in the common units for any of the three muscles. Post hoc tests

344 of the within age group differences (within the older adults) determined that the AFR increased
345 in the MG, LG and SOL unique units during the 30° (from 0°: M = 0.92 Hz, and from 120° =
346 2.79 Hz) leaning direction whereas the common units remained relatively constant (Figure 4).
347 The AFR of the unique units was significantly higher than the common units ($F = 42.63$, $p <$
348 0.001 , Cohen's $d = 0.99$) among all directions in all 3 muscles (0°: M = 4.31 Hz, 30°: M = 5.13
349 Hz, 60°: M = 4.72 Hz, 90°: M = 4.09 Hz, 120°: M = 3.44 Hz). The muscle effect ($F = 15.70$, $p <$
350 0.001) determined higher firing rates in the gastrocnemii compared to the SOL (MG: M = 2.54
351 Hz, LG: M = 2.57 Hz). There were no statistical differences between the LoS conditions ($p =$
352 0.19). There was a main effect of age on the AFRs, that was dependent on the Muscle and MU
353 Subgroup as indicated by the Muscle x MU Subgroup x Age Group interaction ($F = 3.35$, $p =$
354 0.009 , Cohen's $d = 0.33$). Post hoc tests between the older and younger age groups determined that
355 older adults unique and common units firing rates were significantly lower than the young adults
356 across all directions in the MG (M = 2.3 Hz) and SOL (M = 3.5 Hz). In addition, the common
357 units of the older adults did not modulate across directions, whereas the young adults did (M =
358 2.1 Hz).

359 *Coefficient of Variation*

360 The estimates of the CoV_{ISI} are presented in Figure 5. The CoV_{ISI} values increased across
361 leaning directions ($F = 10.35$, $p < 0.001$, Cohen's $d = 0.09$). Similar to the AFR, the directional
362 effect was dependent on the MU subgroup, indicated by the interaction between Direction and
363 MU Subgroup ($F = 8.48$, $p < 0.001$, Cohen's $d = 0.92$). There were no directional effects in the
364 common units for any of the three muscles. Post hoc tests determined that the CoV_{ISI} increased
365 in the MG, LG and SOL unique units during the 30° (from 0°: M = 3.2%, 90°: M = 5.5%, and
366 120°: M = 5.5%) and 60° (from 0°: M = 2.2%, 90°: M = 4.3% and 120°: M = 4.3%) leaning

367 directions. The CoV_{ISI} of the unique units was significantly higher than the common units ($F =$
368 $42.63, p < 0.001$) among all directions in the MG ($M = 12.3\%$), LG ($M = 11.3\%$) and SOL ($M =$
369 13.5%). The muscle effect ($F = 15.70, p < 0.001$, Cohen's $d = 2.52$) determined higher CoV_{ISI}
370 values in the gastrocnemii compared to the SOL (MG: $M = 9.5\%$, LG: $M = 8.4\%$). There was a
371 main effect of age on the CoV_{ISI} , that was dependent on the MU subgroup and the Muscle type
372 indicated by Muscle x MU subgroup x Age Group interaction ($F = 6.23, p < 0.001$, Cohen's $d =$
373 0.82). Post hoc analysis between the older and younger age groups determined that the CoV_{ISI} of
374 the unique units of the older adults were higher across all leaning directions in the MG ($M =$
375 5.3%) and LG ($M = 4.8\%$).

376 *Degree of intermittency*

377 The estimates of the degree of intermittency are displayed in Figure 6. The degree of
378 intermittency changed dependent on the MU subgroup across direction, indicated by the
379 Direction x MU Subgroup interaction ($F = 9.22, p = 0.003$, Cohen's $d = 0.32$). Interestingly, the
380 unique units increased their intermittency from the 0° and 120° directions to the 30° ($0^\circ: M =$
381 $0.084, 120^\circ: M = 0.07$) and the 60° ($0^\circ: M = 0.098, 120^\circ: M = 0.089$) whereas the common units
382 showed a decrease in their intermittency from those comparators. In addition, the unique units
383 had significantly more intermittent activity across all directions compared to the common units
384 ($M = 0.11 - 0.19$). Additionally, there was a main effect of muscle ($F = 9.23, p < 0.001$, Cohen's
385 $d = 0.47$) with the MG MUs ($M = 0.29-0.37$) having more intermittent activity compared to the
386 SOL. There was a main effect of age on the degree of intermittency, that was dependent on the
387 MU subgroup and Direction as indicated by MU Subgroup x Direction x Age Group interaction
388 ($F = 6.23, p < 0.001$, Cohen's $d = 0.53$). Post hoc tests between the older and younger age group

389 determined that the unique units of the older adults increased during the 30° (M = 0.244) and 60°
390 (M = 0.278) leaning directions, whereas the unique units of the young adults decreased.

391 *Balance Performance*

392 The estimated means and CIs calculated from the linear mixed effect models for the CoP
393 outcome measures are displayed in Table 3. There was a main effect of direction for both CoP
394 path length and mean velocity (path length: $F = 5.40$, $p = 0.0003$; mean velocity: $F = 7.54$, $p <$
395 0.0001). Post hoc tests determined that there was higher CoP path length and mean velocity
396 during the 60° (M = 6.75 cm; M = 0.39 cm/s), and 90° (M = 9.08 cm; M = 0.51 cm/s) leaning
397 directions as compared to the 0° leaning direction. Additionally, there was a main effect of LoS
398 for both CoP path length and mean velocity (path length: $F = 6.13$, $p = 0.014$; mean velocity: $F =$
399 10.49 , $p = 0.0014$), however, after post hoc corrections were applied the effect was found to be
400 not statistically significant ($p = 0.064$ and $p = 0.065$ for the path length and velocity,
401 respectively). No other CoP parameter had statistically significant differences between leaning
402 directions or LoS conditions (p values range 0.23-0.91).

403

404 **Discussion**

405 The ability of older adults to differentially recruit and control MUs was evaluated during
406 a multi-directional leaning task. The main findings were that a majority of the MUs decomposed
407 in the older adults in this study were considered common (55.4%). In addition, the common units
408 in the older adults occupied significantly larger muscle volumes compared to the young adults
409 with moderate effect sizes. Second, the medio-lateral positions of the barycenter in older adults
410 of both unique and common units were found in similar locations for all leaning directions;
411 while, the unique units shifted in different locations in the proximo-distal aspect of the muscle

412 with direction, albeit with small effect sizes. However, a moderate effect size was found between
413 the two age groups MU barycenter's with the older adults barycenter's shifting significantly less
414 than the young adults. While leaning in different directions did not affect the AFR and CoV_{ISI} of
415 common units, the unique units firing behaviour responded to the leaning directions with
416 increasing AFR (albeit in only the 30° leaning direction with a small effect size), and CoV_{ISI} .
417 The CoV_{ISI} of the older adults was significantly higher than the young adults across all directions
418 and MU subgroups, with large effect sizes. Additionally, while the unique units were observed to
419 increase their intermittency during diagonal leaning directions, the common units were observed
420 to decrease it in the older adults. This is in contrast to those in the young adults, where common
421 units are observed to decrease their intermittent behaviour during diagonal and forward leaning
422 directions. Finally, older adults had higher CoP mean velocity and an increase in the CoP total
423 path length in the 60° and 90° directions than during the 0° . Taken together, this study suggests
424 that older adults may have difficulty controlling directionally-specific leaning tasks in standing.

425

426 *Relative constant AFRs across leaning directions*

427 The unique and common units in older adults were observed to behave differently from
428 each other. Across the leaning directions, the unique units discharged at higher AFRs compared
429 to the common units (Figure 4) with moderate effect sizes. Our previous study in young adults
430 also found similar findings regarding the separation between the AFR of unique and common
431 units (5).

432 However, contrary to the findings in young adults, the modulation of the AFRs of older
433 adults across directions is modest. The common units maintained a constant AFR across all
434 leaning directions, and the unique units had a statistically significant increase but only in one

435 leaning direction (30°) with minimal practical significance (Cohen's $d < 0.2$). This contrasts
436 findings from young adults, where modulation occurs in both common and unique units across
437 the diagonal and forward-leaning directions (30°-90°). Previous studies have observed older
438 adults exhibiting a reduced capacity for rate coding in a variety of force tasks (28, 47-49).
439 Multiple mechanisms contribute to the decreased rate coding seen in older adults (50-53). Due to
440 the decreased capacity to modulate the firing frequencies of MUs, older adults may utilize MU
441 recruitment for force production; however, recruitment may also be impaired because of the
442 smaller proportion of unique units (compared to young adults) and the difficulty to maintain their
443 activity (see below).

444 The decreased modulation in older adults observed in this study may also indicate a
445 decreased ability to flexibly control MUs with selective neural signals. Several studies have
446 demonstrated that there is increased common synaptic input with ageing. Semmler et al., 2003
447 (32) observed that older adults demonstrate greater strength of coherent MU activity, and
448 therefore, a high amplitude of common input, compared to young adults during low-intensity
449 contractions of the first dorsal interosseous. Furthermore, it has been shown that older adults
450 have a greater degree of variation in force fluctuations in the wrist extensors in voluntary than
451 evoked contractions, indicating that the variance in synaptic input is greater for older adults than
452 for young adults (54). In the lower limb, Castronovo et al. 2018 (77) found that declines in force
453 steadiness during isometric contractions of the tibialis anterior seen in older adults were
454 associated with an increase in the power of the common input to motor neurons. Taken together,
455 the greater strength of the common modulation may diminish the ability to flexibly control MUs.
456 With a larger common unit signal, there may be decreased independent synaptic inputs to the
457 different MU subgroups.

458 The consequence of the decreased flexible control and/or the utilization of limited MU
459 recruitment may be decreased force output leading to an increase in the CoP parameters seen
460 during the 60°-90° directions, where force requirements are highest.

461

462 *Differential Behaviour of the Degree of Intermittency*

463 The unique units of older adults become more intermittent across directions. In stark
464 contrast to young adults (5), the older adults unique units displayed increased intermittency
465 during more forward directions (i.e. 30°-60°) with moderate effects (Cohen's $d > 0.5$). It has
466 previously been shown in young adults that during forward sway MUs in the ankle plantarflexors
467 become less intermittent (5, 55), whereas during backward sway MUs become more intermittent
468 (55). It is presumed that one of the main mechanisms driving intermittent MU activity in young
469 adults is the ability to leverage the pull of gravity to maintain balance, providing an economical
470 solution to standing posture (55). MUs have been shown to de-recruit below a physiological
471 threshold of 4 Hz during backwards translations of the body suggesting that the CNS allows the
472 force of gravity to 'pull' the body to equilibrium rather than actively producing force (55). By
473 allowing the body to essentially fall back to equilibrium, the MUs are not required to be
474 constantly active, thus providing an energy efficient solution to maintaining standing balance.
475 The observation that an opposite outcome was found in older adults may be indicative of two
476 things. First, it may be illustrating that older adults have difficulty recruiting and maintaining
477 activation of the higher threshold unique units. This may derive from the established increased
478 motor unit recruitment thresholds (28, 56) and/or the decreased voluntary activation seen during
479 ageing (57). With a decreased ability to recruit and maintain activation of the unique units, older
480 adults may not be able to achieve the firing rates to maintain continuous activity.

481 A second explanation for the increased intermittency could derive from the deterioration
482 of the aged sensory system resulting in the reduced ability to detect CoP positional changes. This
483 hypothesis fits well with the literature suggesting decreased sensory feedback in older adults (58,
484 59). Previous research has determined intrinsic sensory factors such as decreased cutaneous
485 sensation in the feet (60, 61), and muscle spindle deterioration (62-65) occurring in the older
486 adult which may lead to postural instability (61, 66). One may assume that with decreased
487 sensory feedback, the older adult's CoP positional sense would be less responsive to CoP
488 positional changes. Accordingly, perhaps older adults cannot detect the pull of gravity to the
489 same extent as young adults to provide an economical neural solution to the unstable body,
490 providing additional information regarding older adult's efficiency during postural control.

491 The common units of the older adults became less intermittent across leaning directions.
492 This result is consistent with previous reports on intermittency (55), and is also similar to what
493 has been observed in young adults; a decrease in the common units degree of intermittency was
494 observed but was found to be non-statistically significant (5). Taken together, the intermittent
495 responses of the common units of older adults appear to be similarly sensitive to the positional
496 changes during forward translations as the young adults.

497

498 *Regional recruitment – Barycenter positions*

499 During the leaning protocol, older adults were found to recruit unique units in distinct
500 locations in the proximo-distal direction with small effect sizes (Cohen's $d < 0.5$), but not in the
501 medio-lateral direction (Table 2). The common units were found in similar locations in both
502 proximo-distal and medio-lateral directions. Regional recruitment of MUs creates torques that
503 are most efficient for the specific motor output by increasing the moment arm of the torque

504 generated. While the triceps surae are conventionally thought to exclusively produce plantar
505 flexion torques, it has been demonstrated that inversion and eversion torques are derived from
506 these muscles (4, 30, 67, 68). As the relative degree of inversion and eversion increases at the
507 ankle joint, so does the relative moment arm in the MG and LG. Accordingly, regional activation
508 shifts in medio-lateral direction on the triceps surae can change the length of the moment arms
509 producing more effective inversion and eversion torques required for maintaining balance during
510 leaning. Accordingly, the CNS leverages MUs in the most optimal mechanically advantageous
511 positions for force production (4, 30, 69, 70), by recruiting MUs in the medio-lateral direction
512 for inversion-eversion torques, and proximally for plantar flexion torques. In this study, the older
513 adults MUs were recruited only in the proximal direction, whereas young adults recruit MUs in
514 both directions. In our previous research with young adults (4, 5), we postulated that common
515 units are recruited to produce a baseline tonic level of force production. We inferred that the
516 common MUs were smaller compared to the unique units, and thus were likely recruited first.
517 Once the task exceeded the capabilities of the common units, the recruitment of unique units
518 with fibres grouped in different medio-lateral and proximo-distal spatial locations was utilized to
519 meet the force requirements of the task. The muscle fibres belonging to the unique units,
520 recruited based on task demands, were situated in optimal positions to effectively contribute to
521 the torque requirements. Similar observations in the older adults have been identified in this
522 study: the unique units occupy larger muscle regions than the common units, with lower firing
523 rates in the common units, which could suggest that a similar mechanism of recruitment could be
524 utilized. However, the results of the current study continue to add to this understanding with
525 three critical results. First, there are proportionally fewer identified unique units (compared to
526 young adults) regardless of the spatial direction in the older adult populations. With likely fewer

527 unique units available for recruitment, the ability of the CNS to leverage unique units situated in
528 optimal positions may be suppressed. While speculative, this finding may be due to the MU
529 remodelling process seen during ageing. As motoneurons degenerate, the surviving motoneurons
530 re-innervate orphaned fibres. This process may increase the number of common units, putatively
531 decreasing the number of unique units as the larger motoneurons are believed to degenerate first
532 (23). Indeed, we did find that the area of the common units of the older adults was significantly
533 larger than those in the young adults suggesting that these units have adopted the orphaned
534 fibres. However, while this explanation may explain the proportional increase in common units,
535 it does not provide an excellent explanation for the recruitment patterns. Older adults continued
536 to utilize regionally selective MU recruitment in the proximo-distal direction (Table 2),
537 suggesting there may be an alternative explanation for the regional recruitment observations.

538 Second, a more favourable explanation of the MU recruitment observed may derive from
539 the physiological changes seen in older adults. As mentioned, many sensory deficits can affect
540 postural control in older adults (60-65). If the CNS in older adults cannot identify the directional
541 challenges the task demands, it may no longer utilize regional MU recruitment. Although there
542 are a considerable number of factors that affect postural control performance in the older adult
543 (71-74), the lack of regional recruitment may also contribute to the reduced task performance in
544 this study. The lack of regional recruitment may be an underlying mechanism of motor control
545 that has remained hitherto unexplored within the ageing literature. In this study, we did not
546 observe regionally recruited MUs but we did observe increased CoP path length during the 60°
547 and 90° leaning directions. During these leaning directions, there were significant medio-lateral
548 shifts in young adult barycenter values (4), whereas in the older adults, there are no shifts (Table
549 3). Perhaps the consequence of older adults being unable to recruit MUs in optimal locations to

550 effectively contribute to torque requirements may be the increased CoP path lengths and
551 velocities.

552 A final consideration is that recruiting MUs in distinct locations was not necessarily
553 required in the medio-lateral position of the muscles. The older adults had significantly smaller
554 LoS distances in the medio-lateral direction compared to the young adults (unpublished findings,
555 average distances: young adults = 22.24 cm, older adults = 17.85 cm, two-tailed t -test, $p =$
556 0.036). However, they had similar LoS distances in the anterior-posterior direction (average
557 distances: young adults = 16.92 cm, older adults = 14.90, $p = 0.10$). With smaller medio-lateral
558 LoS distances, there may be a reduced need for spatial recruitment in the medio-lateral position
559 on the muscle. However, with similar anterior-posterior LoS distances achieved, the CNS
560 continues to recruit units in spatial locations based on the highest mechanical advantage as a
561 requirement for necessary force production.

562

563 *Limitations*

564 The older adult experimental groups recruited were relatively healthy, active participants,
565 most of whom are not considered frequent fallers (defined as having had two or more falls within
566 the last year (75, 76) (15/17 participants). Future studies should focus on recruiting specific
567 demographics with frequent fallers to determine if there are further differences in the recruitment
568 and MU control which may relate to falls.

569

570 **References**

571 1. **Masani K, Popovic MR, Nakazawa K, Kouzaki M, and Nozaki D.** Importance of body
572 sway velocity information in controlling ankle extensor activities during quiet stance. *J*
573 *Neurophysiol* 90: 3774-3782, 2003.

- 574 2. **Gatev P, Thomas S, Kepple T, and Hallett M.** Feedforward ankle strategy of balance
575 during quiet stance in adults. *J Physiol* 514 (Pt 3): 915-928, 1999.
- 576 3. **Di Giulio I, Maganaris CN, Baltzopoulos V, and Loram ID.** The proprioceptive and
577 agonist roles of gastrocnemius, soleus and tibialis anterior muscles in maintaining human
578 upright posture. *J Physiol* 587: 2399-2416, 2009.
- 579 4. **Cohen JW, Vieira T, Ivanova TD, Cerone GL, and Garland SJ.** Maintenance of standing
580 posture during multi-directional leaning demands the recruitment of task-specific motor units
581 in the ankle plantarflexors. *Exp Brain Res* 239: 2569-2581, 2021.
- 582 5. **Cohen JW, Vieira T, Ivanova TD, and Garland SJ.** Differential behaviour of distinct
583 motoneuron pools that innervate the triceps surae. *J Neurophysiol* 2022.
- 584 6. **Farina D, Negro F, Gazzoni M, and Enoka RM.** Detecting the unique representation of
585 motor-unit action potentials in the surface electromyogram. *J Neurophysiol* 100: 1223-1233,
586 2008.
- 587 7. **Cao L, and Morley JE.** Sarcopenia Is Recognized as an Independent Condition by an
588 International Classification of Disease, Tenth Revision, Clinical Modification (ICD-10-CM) Code. *J*
589 *Am Med Dir Assoc* 17: 675-677, 2016.
- 590 8. **Cruz-Jentoft AJ, and Sayer AA.** Sarcopenia. *The Lancet* 393: 2636-2646, 2019.
- 591 9. **Hunter SK, Pereira HM, and Keenan KG.** The aging neuromuscular system and motor
592 performance. *J Appl Physiol (1985)* 121: 982-995, 2016.
- 593 10. **Kim AY, Lee JK, Kim SH, Choi J, Song JJ, and Chae SW.** Is postural dysfunction related to
594 sarcopenia? A population-based study. *PloS one* 15: e0232135, 2020.
- 595 11. **Bianchetti A, and Novelli A.** Sarcopenia in the elderly: from clinical aspects to
596 therapeutic options. *Geriatric Care* 5: 2019.
- 597 12. **Liguori I, Russo G, Aran L, Bulli G, Curcio F, Della-Morte D, Gargiulo G, Testa G,**
598 **Cacciatore F, Bonaduce D, and Abete P.** Sarcopenia: assessment of disease burden and
599 strategies to improve outcomes. *Clin Interv Aging* 13: 913-927, 2018.
- 600 13. **Doherty TJ, Vandervoort AA, and Brown WF.** Effects of ageing on the motor unit: a brief
601 review. *Can J Appl Physiol* 18: 331-358, 1993.
- 602 14. **Kawamura Y, O'Brien P, Okazaki H, and Dyck PJ.** Lumbar motoneurons of man II: the
603 number and diameter distribution of large-and intermediate-diameter cytons in "motoneuron
604 columns" of spinal cord of man. *Journal Neuropathol Exp Neurol* 36: 861-870, 1977.
- 605 15. **Kawamura Y, Okazaki H, O'Brien PC, and Dyck PJ.** Lumbar motoneurons of man: I)
606 number and diameter histogram of alpha and gamma axons of ventral root. *J Neuropathol Exp*
607 *Neurol* 36: 853-860, 1977.
- 608 16. **Mittal KR, and Logmani FH.** Age-related reduction in 8th cervical ventral nerve root
609 myelinated fiber diameters and numbers in man. *J Gerontol* 42: 8-10, 1987.
- 610 17. **Campbell MJ, McComas AJ, and Petito F.** Physiological changes in ageing muscles. *J*
611 *Neurol Neurosurg Psychiatry* 36: 174-182, 1973.
- 612 18. **Tomlinson BE, and Irving D.** The numbers of limb motor neurons in the human
613 lumbosacral cord throughout life. *J Neurol Sci* 34: 213-219, 1977.
- 614 19. **Andersen JL.** Muscle fibre type adaptation in the elderly human muscle. *Scand J Med Sci*
615 *Sports* 13: 40-47, 2003.

- 616 20. **Justice JN, Mani D, Pierpoint LA, and Enoka RM.** Fatigability of the dorsiflexors and
617 associations among multiple domains of motor function in young and old adults. *Exp Gerontol*
618 55: 92-101, 2014.
- 619 21. **McNeil CJ, Doherty TJ, Stashuk DW, and Rice CL.** Motor unit number estimates in the
620 tibialis anterior muscle of young, old, and very old men. *Muscle Nerve* 31: 461-467, 2005.
- 621 22. **Holloszy JO, and Larsson L.** Motor units: remodeling in aged animals. *J Gerontol - Biol Sci*
622 *Med Sci* 50: 91-95, 1995.
- 623 23. **Porter MM, Vandervoort AA, and Lexell J.** Aging of human muscle: structure, function
624 and adaptability. *Scand J Med Sci Sports* 5: 129-142, 1995.
- 625 24. **Evans WJ, and Lexell J.** Human aging, muscle mass, and fiber type composition. *J*
626 *Gerontol - Biol Sci Med Sci* 50: 11-16, 1995.
- 627 25. **Piasecki M, Ireland A, Piasecki J, Stashuk DW, Swiecicka A, Rutter MK, Jones DA, and**
628 **McPhee JS.** Failure to expand the motor unit size to compensate for declining motor unit
629 numbers distinguishes sarcopenic from non-sarcopenic older men. *J Physiol* 596: 1627-1637,
630 2018.
- 631 26. **Piasecki M, Ireland A, Coulson J, Stashuk DW, Hamilton-Wright A, Swiecicka A, Rutter**
632 **MK, McPhee JS, and Jones DA.** Motor unit number estimates and neuromuscular transmission
633 in the tibialis anterior of master athletes: evidence that athletic older people are not spared
634 from age-related motor unit remodeling. *Physiol Rep* 4: e12987, 2016.
- 635 27. **Roos MR, Rice CL, and Vandervoort AA.** Age-related changes in motor unit function.
636 *Muscle Nerve* 20: 679-690, 1997.
- 637 28. **Erim Z, Beg MF, Burke DT, and de Luca CJ.** Effects of aging on motor-unit control
638 properties. *J Neurophysiol* 82: 2081-2091, 1999.
- 639 29. **Vieira TM, Loram ID, Muceli S, Merletti R, and Farina D.** Postural activation of the
640 human medial gastrocnemius muscle: are the muscle units spatially localised? *J Physiol* 589:
641 431-443, 2011.
- 642 30. **Cohen JW, Gallina A, Ivanova TD, Vieira T, McAndrew DJ, and Garland SJ.** Regional
643 modulation of the ankle plantarflexor muscles associated with standing external perturbations
644 across different directions. *Exp Brain Res* 238: 39-50, 2020.
- 645 31. **Dos Anjos FV, Pinto TP, Gazzoni M, and Vieira TM.** The Spatial Distribution of Ankle
646 Muscles Activity Discriminates Aged from Young Subjects during Standing. *Front Hum Neurosci*
647 11: 190, 2017.
- 648 32. **Semmler JG, Kornatz KW, and Enoka RM.** Motor-unit coherence during isometric
649 contractions is greater in a hand muscle of older adults. *J Neurophysiol* 90: 1346-1349, 2003.
- 650 33. **Basmajian JV.** Control and training of individual motor units. *Science* 141: 440-441,
651 1963.
- 652 34. **Formento E, Botros P, and Carmena J.** A non-invasive brain-machine interface via
653 independent control of individual motor units. *bioRxiv* 2021.
- 654 35. **Desmedt HE, and Godaux E.** Spinal motoneuron recruitment in man: rank deordering
655 with direction but not with speed of voluntary movement. *Science* 214: 933-936, 1981.
- 656 36. **Herrmann U, and Flanders M.** Directional tuning of single motor units. *J Neurosci* 18:
657 8402-8416, 1998.
- 658 37. **Hug F, Avrillon S, Ibanez J, and Farina D.** Common synaptic input, synergies and size
659 principle: Control of spinal motor neurons for movement generation. *J Physiol* 601: 11-20, 2023.

- 660 38. **Thomsen MH, Stottrup N, Larsen FG, Pedersen ASK, Poulsen AG, and Hirata RP.** Four-
661 way-leaning test shows larger limits of stability than a circular-leaning test. *Gait Posture* 51: 10-
662 13, 2017.
- 663 39. **Morasso PG, and Sanguineti V.** Ankle muscle stiffness alone cannot stabilize balance
664 during quiet standing. *J Neurophysiol* 88: 2157-2162, 2002.
- 665 40. **Vieira TM, and Botter A.** The Accurate Assessment of Muscle Excitation Requires the
666 Detection of Multiple Surface Electromyograms. *Exerc Sport Sci Rev* 49: 23-34, 2021.
- 667 41. **Botter A, and Vieira TM.** Optimization of surface electrodes location for H-reflex
668 recordings in soleus muscle. *J Electromyogr Kines* 34: 14-23, 2017.
- 669 42. **Holobar A, Minetto M, Botter A, and Farina D.** Identification of motor unit discharge
670 patterns from high-density surface EMG during high contraction levels. In: *5th European*
671 *Conference of the International Federation for Medical and Biological Engineering* Springer,
672 2011, p. 1165-1168.
- 673 43. **Holobar A, and Farina D.** Blind source identification from the multichannel surface
674 electromyogram. *Physiol Meas* 35: R143-165, 2014.
- 675 44. **Power KE, Lockyer EJ, Botter A, Vieira T, and Button DC.** Endurance-exercise training
676 adaptations in spinal motoneurons: potential functional relevance to locomotor output and
677 assessment in humans. *Eur J Appl Physiol* 122: 1367-1381, 2022.
- 678 45. **Vieira TM, Merletti R, and Mesin L.** Automatic segmentation of surface EMG images:
679 Improving the estimation of neuromuscular activity. *J Biomech* 43: 2149-2158, 2010.
- 680 46. **Bates D, Machler M, Bolker BM, and Walker SC.** Fitting Linear Mixed-Effects Models
681 Using lme4. *J Stat Softw* 67: 1-48, 2015.
- 682 47. **Kamen G, Sison SV, Du CC, and Patten C.** Motor unit discharge behavior in older adults
683 during maximal-effort contractions. *J Appl Physiol* 79: 1908-1913, 1995.
- 684 48. **Knight CA, and Kamen G.** Modulation of motor unit firing rates during a complex
685 sinusoidal force task in young and older adults. *J Appl Physiol* 102: 122-129, 2007.
- 686 49. **Patten C, Kamen G, and Rowland DM.** Adaptations in maximal motor unit discharge
687 rate to strength training in young and older adults. *Muscle Nerve* 24: 542-550, 2001.
- 688 50. **Larsson L, Degens H, Li M, Salviati L, Lee YI, Thompson W, Kirkland JL, and Sandri M.**
689 Sarcopenia: Aging-Related Loss of Muscle Mass and Function. *Physiol Rev* 99: 427-511, 2019.
- 690 51. **Miller RG.** The effects of aging upon nerve and muscle function and their importance for
691 neurorehabilitation. *J Neurol Rehab* 9: 175-181, 1995.
- 692 52. **Oda K.** Age changes of motor innervation and acetylcholine receptor distribution on
693 human skeletal muscle fibres. *J Neurol Sci* 66: 327-338, 1984.
- 694 53. **Enoka RM, Christou EA, Hunter SK, Kornatz KW, Semmler JG, Taylor AM, and Tracy BL.**
695 Mechanisms that contribute to differences in motor performance between young and old
696 adults. *J Electromyogr Kinesiol* 13: 1-12, 2003.
- 697 54. **Mani D, Feeney DF, and Enoka RM.** The modulation of force steadiness by electrical
698 nerve stimulation applied to the wrist extensors differs for young and older adults. *Eur J Appl*
699 *Physiol* 119: 301-310, 2019.
- 700 55. **Vieira TM, Loram ID, Muceli S, Merletti R, and Farina D.** Recruitment of motor units in
701 the medial gastrocnemius muscle during human quiet standing: is recruitment intermittent?
702 What triggers recruitment? *J Neurophysiol* 107: 666-676, 2012.

- 703 56. **Galganski ME, Fuglevand AJ, and Enoka RM.** Reduced control of motor output in a
704 human hand muscle of elderly subjects during submaximal contractions. *J Neurophysiol* 69:
705 2108-2115, 1993.
- 706 57. **Klass M, Baudry S, and Duchateau J.** Voluntary activation during maximal contraction
707 with advancing age: a brief review. *Eur J Appl Physiol* 100: 543-551, 2007.
- 708 58. **Earles D, Vardaxis V, and Koceja D.** Regulation of motor output between young and
709 elderly subjects. *Clin Neurophysiol* 112: 1273-1279, 2001.
- 710 59. **Scaglioni G, Narici MV, Maffiuletti NA, Pensini M, and Martin A.** Effect of ageing on the
711 electrical and mechanical properties of human soleus motor units activated by the H reflex and
712 M wave. *J Physiol* 548: 649-661, 2003.
- 713 60. **Peters RM, McKeown MD, Carpenter MG, and Inglis JT.** Losing touch: age-related
714 changes in plantar skin sensitivity, lower limb cutaneous reflex strength, and postural stability
715 in older adults. *J Neurophysiol* 116: 1848-1858, 2016.
- 716 61. **Song Q, Zhang X, Mao M, Sun W, Zhang C, Chen Y, and Li L.** Relationship of
717 proprioception, cutaneous sensitivity, and muscle strength with the balance control among
718 older adults. *J Sport Health Sci* 10: 585-593, 2021.
- 719 62. **Burke JR, Schutten MC, Koceja DM, and Kamen G.** Age-dependent effects of muscle
720 vibration and the Jendrassik maneuver on the patellar tendon reflex response. *Arch Phys Med*
721 *Rehabil* 77: 600-604, 1996.
- 722 63. **Kim GH, Suzuki S, and Kanda K.** Age-related physiological and morphological changes of
723 muscle spindles in rats. *J Physiol* 582: 525-538, 2007.
- 724 64. **Miwa T, Miwa Y, and Kanda K.** Dynamic and static sensitivities of muscle spindle
725 primary endings in aged rats to ramp stretch. *Neurosci Lett* 201: 179-182, 1995.
- 726 65. **Liu JX, Eriksson PO, Thornell LE, and Pedrosa-Domellof F.** Fiber content and myosin
727 heavy chain composition of muscle spindles in aged human biceps brachii. *J Histochem*
728 *Cytochem* 53: 445-454, 2005.
- 729 66. **Henry M, and Baudry S.** Age-related changes in leg proprioception: implications for
730 postural control. *J Neurophysiol* 122: 525-538, 2019.
- 731 67. **Lee SS, and Piazza SJ.** Inversion-eversion moment arms of gastrocnemius and tibialis
732 anterior measured in vivo. *J Biomech* 41: 3366-3370, 2008.
- 733 68. **Vieira TM, Minetto MA, Hodson-Tole EF, and Botter A.** How much does the human
734 medial gastrocnemius muscle contribute to ankle torques outside the sagittal plane? *Hum Mov*
735 *Sci* 32: 753-767, 2013.
- 736 69. **Butler TJ, Kilbreath SL, Gorman RB, and Gandevia SC.** Selective recruitment of single
737 motor units in human flexor digitorum superficialis muscle during flexion of individual fingers. *J*
738 *Physiol* 567: 301-309, 2005.
- 739 70. **Butler JE, and Gandevia SC.** The output from human inspiratory motoneurone pools. *J*
740 *Physiol* 586: 1257-1264, 2008.
- 741 71. **Papegaaij S, Taube W, Baudry S, Otten E, and Hortobagyi T.** Aging causes a
742 reorganization of cortical and spinal control of posture. *Front Aging Neurosci* 6: 28, 2014.
- 743 72. **Anton SD, Woods AJ, Ashizawa T, Barb D, Buford TW, Carter CS, Clark DJ, Cohen RA,**
744 **Corbett DB, Cruz-Almeida Y, Dotson V, Ebner N, Efron PA, Fillingim RB, Foster TC,**
745 **Gundermann DM, Joseph AM, Karabetian C, Leeuwenburgh C, Manini TM, Marsiske M,**
746 **Mankowski RT, Mutchie HL, Perri MG, Ranka S, Rashidi P, Sandesara B, Scarpace PJ, Sibille KT,**

- 747 **Solberg LM, Someya S, Uphold C, Wohlgemuth S, Wu SS, and Pahor M.** Successful aging:
 748 Advancing the science of physical independence in older adults. *Ageing Res Rev* 24: 304-327,
 749 2015.
- 750 73. **Chiba R, Takakusaki K, Ota J, Yozu A, and Haga N.** Human upright posture control
 751 models based on multisensory inputs; in fast and slow dynamics. *Neurosci Res* 104: 96-104,
 752 2016.
- 753 74. **Wiesmeier IK, Dalin D, and Maurer C.** Elderly Use Proprioception Rather than Visual and
 754 Vestibular Cues for Postural Motor Control. *Front Aging Neurosci* 7: 97, 2015.
- 755 75. **Quijoux F, Vienne-Jumeau A, Bertin-Hugault F, Zawieja P, Lefevre M, Vidal PP, and**
 756 **Ricard D.** Center of pressure displacement characteristics differentiate fall risk in older people:
 757 A systematic review with meta-analysis. *Ageing Res Rev* 62: 101117, 2020.
- 758 76. **Cho CY, and Kamen G.** Detecting balance deficits in frequent fallers using clinical and
 759 quantitative evaluation tools. *J Am Geriatr Soc* 46: 426-430, 1998.
- 760 77. **Castronovo AM, Marchis SD, Schmid M, Conforto S, and Severini G.** Effect of task
 761 failure on intermuscular coherence measures in synergistic muscles. *Appl Bio and Biomech* 2018
 762

763

764 **Figure Legends**

765

766 **Figure 1**

767 A. A schematic of the experimental set up for the multi-directional leaning task. The grey circles
 768 denote the targets positioned at 0°, 30°, 60°, 90°, and 120° in respect to the position of the
 769 participant's center of pressure before the lean shown with the black circle. B. An example of a
 770 center of pressure trace of a single participant during the multi-directional leaning task.

771

772 **Figure 2**

773 Motor unit action potential and corresponding HD-sEMG motor unit activity map. A: An
 774 example of a lateral gastrocnemius motor unit waveform from a single participant during the 60°
 775 target direction. B: The corresponding averaged rectified value (ARV) activity map and
 776 barycenter location. The black circles indicate the channels included in the barycenter calculation
 777 (i.e. area), the larger gray circle is the resultant barycenter. Note the orientation of the axes:
 778 smaller values of the barycenter position indicate proximal (vertical axis) and medial (horizontal
 779 axis). IED = interelectrode distance.

780

781 **Figure 3**

782 Violin plots for each leaning direction of the average firing rate (AFR) distribution of the motor
 783 units (MUs) for all participants combined, identified during both 60% and 80% limits of stability
 784 conditions overlaid by the MU AFR for the unique (left column; A, B) and common (right
 785 column; C, D) units in the medial and lateral gastrocnemius (MG: white circles, LG: black
 786 circles; A, C) and soleus (SOL: white circles; B, D). The number of MUs for each direction are
 787 indicated under each violin plot. A small amount of swarm was added to the x-axis for clarity of
 788 display. The horizontal location of the individual MU AFR does not have an effect on the
 789 distributions of the violin plots. The proportion of common units in each muscle by direction is

790 the following: *MG*: 0°: 43%, 30°: 44% 60°: 53% 90°: 40% 120°: 43%; *LG*: 0°: 42%, 30°: 36%,
 791 60°: 51%, 90°:42%, 120°: 41%; *SOL*: 0°: 63%, 30°: 66%, 60°: 69%%, 90°: 67%, 120°: 67%

792

793 **Figure 4**

794 Average firing rates (AFR) for unique (circles) and common (triangles) motor units during 60%
 795 (grey) and 80% (black) limits of stability (LoS) reported for the medial gastrocnemius (MG),
 796 lateral gastrocnemius (LG), and soleus (SOL). The data are mean estimates and 95% confidence
 797 intervals calculated from the linear mixed model. * Indicates significantly different AFR from
 798 the 0° and 120° leaning directions for both 60% and 80% LoS conditions ($p < 0.001$). † Indicates
 799 significantly different AFR between unique and common units ($p < 0.001$).

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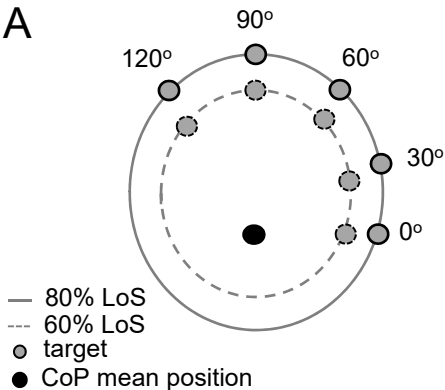
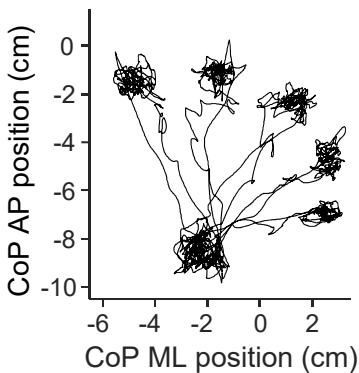
801 **Figure 5**

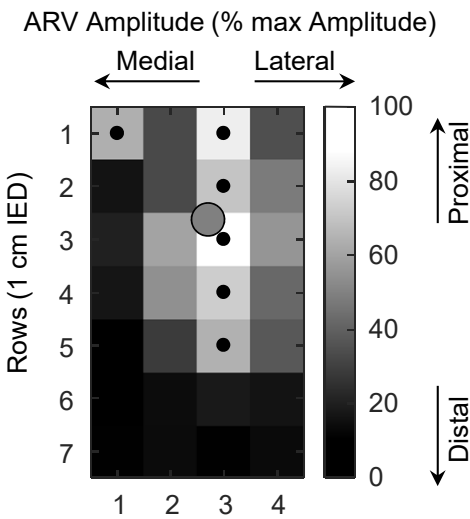
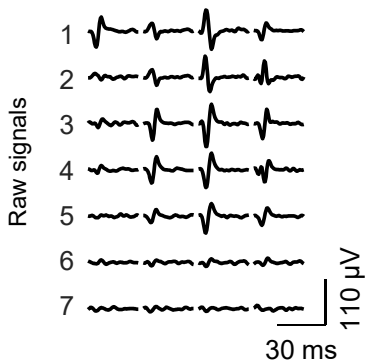
802 Coefficient of variation of the interspike intervals (CoV_{ISI}) for unique (circles) and common
 803 (triangles) motor units during 60% (grey) and 80% (black) limits of stability (LoS) for the medial
 804 gastrocnemius (MG), lateral gastrocnemius (LG), and soleus (SOL). The data are estimated
 805 means and 95% confidence intervals calculated from the linear mixed model. * indicate
 806 significantly different CoV_{ISI} from the 0° and 120° leaning direction for both 60% and 80% LoS.
 807 † Indicates significantly different CoV_{ISI} between unique and common units.

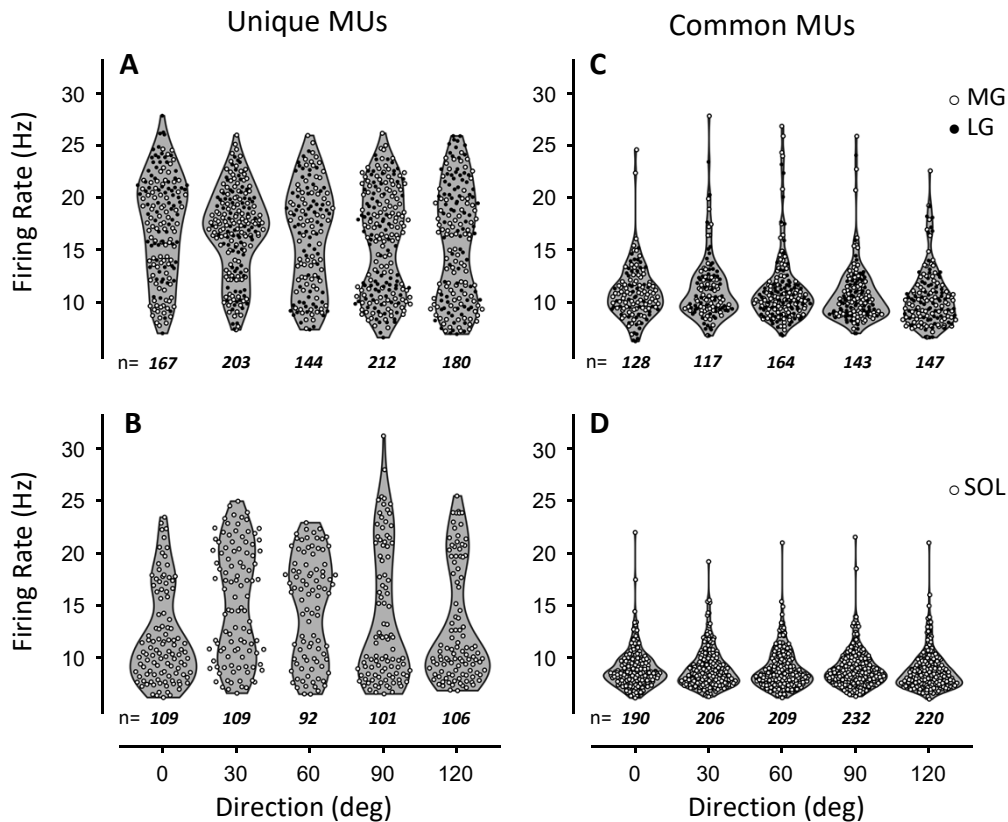
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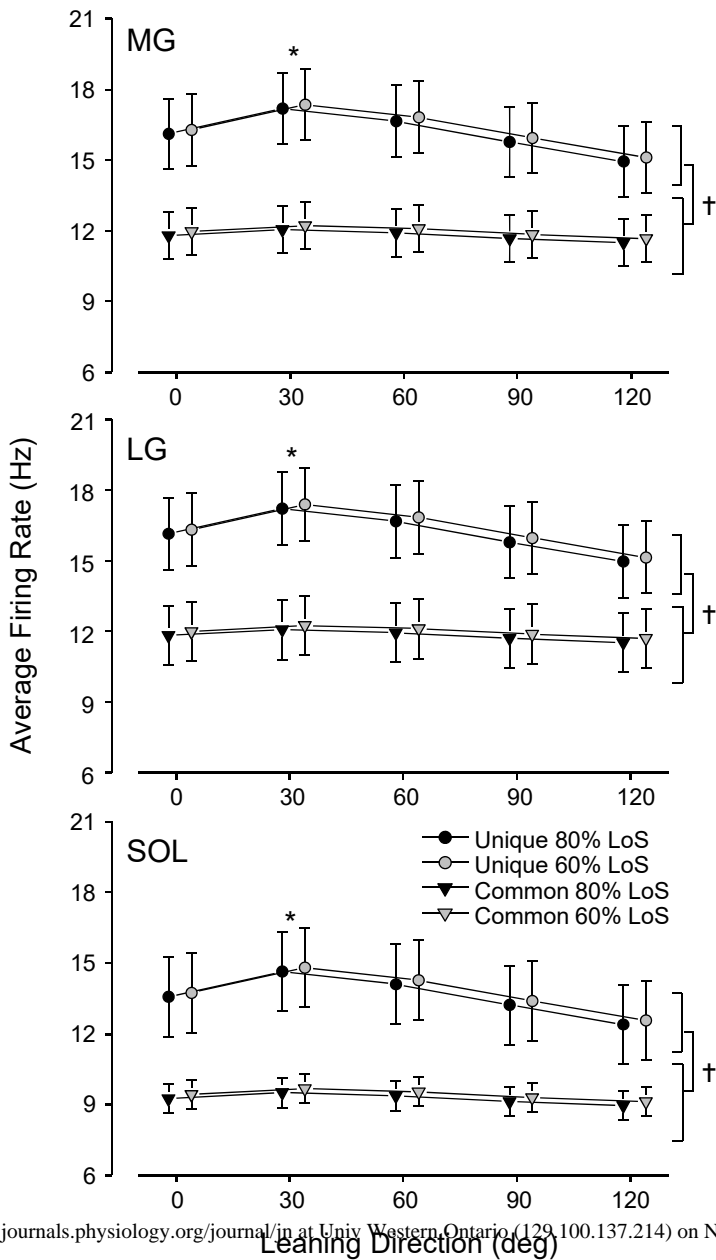
809 **Figure 6**

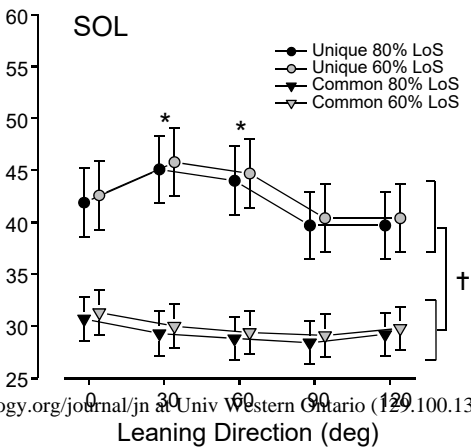
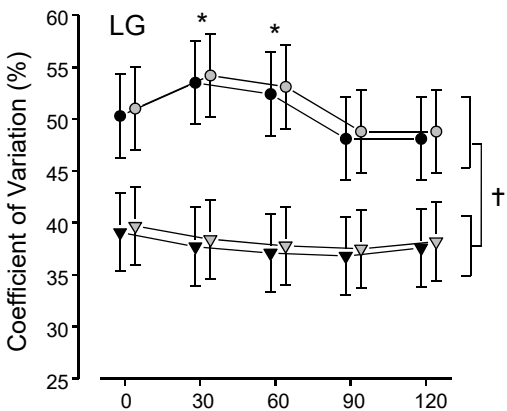
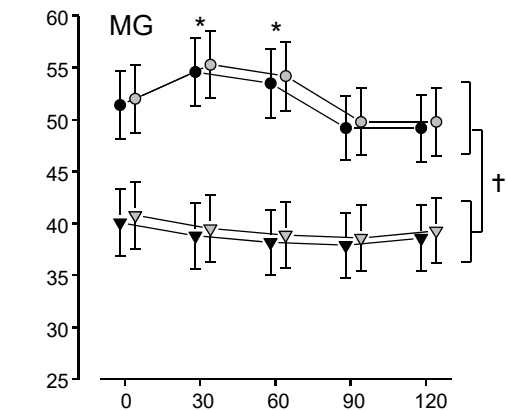
810 Degree of intermittency for unique (top; black symbols) and common (bottom; grey symbols)
 811 motor units. The 60% (grey) limits of stability (LoS) data are depicted for the medial
 812 gastrocnemius (MG), lateral gastrocnemius (LG), and soleus (SOL). The 80% LoS condition had
 813 near identical values and have been omitted for clarity. The data are estimated means and 95%
 814 confidence intervals calculated from the linear mixed model for the log transformed values.
 815 Dotted line rectangles in both unique and common MUs enclose the intermittency values in all 3
 816 muscles that are significantly different from the 0° and 120° leaning directions ($* p < 0.001$). †
 817 Indicates significantly different intermittency between MG and SOL ($p < 0.001$).

A**B**









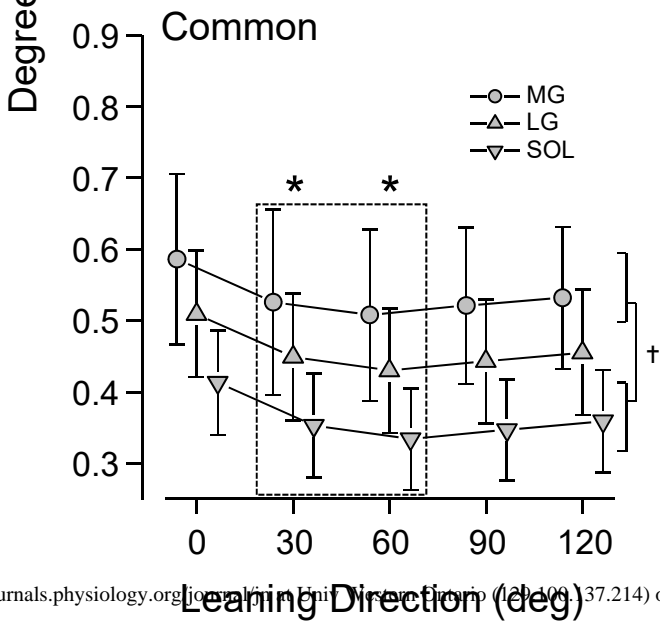
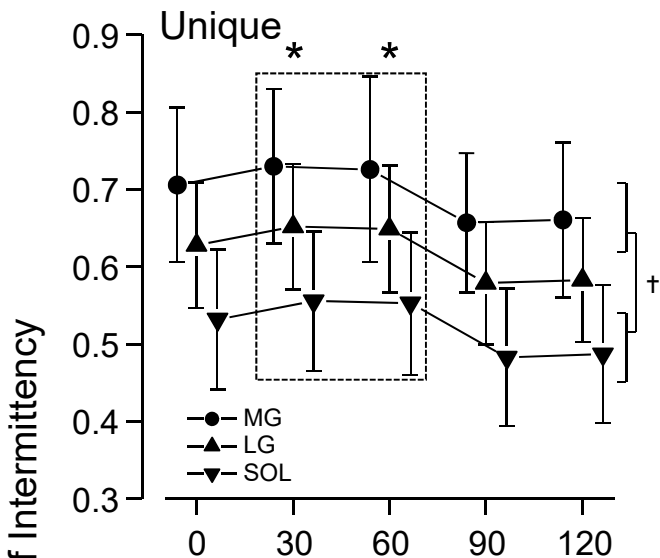


Table 1. Number of identified motor units (MUs) for all leaning directions

MUs	LoS	Medial Gastrocnemius	Lateral Gastrocnemius	Soleus
Total	60%	422 MUs, 25 ± 14 (7-89)	353 MUs, 21 ± 12 (2-48)	759 MUs, 48 ± 33 (12-130)
	80%	485 MUs, 30 ± 21 (6-90)	345 MUs, 20 ± 12 (2-45)	811 MUs, 44 ± 25 (8-145)
Unique	60%	224 MUs, 13 ± 9 (1-30)	188 MUs, 11 ± 9 (1-27)	261 MUs, 15 ± 12 (7-48)
	80%	290 MUs, 17 ± 7 (5-30)	204 MUs, 12 ± 8 (3-23)	252 MUs, 15 ± 10, (4-36)
Common	60%	198 MUs, 12 ± 10 (5-67)	165 MUs, 9 ± 9 (5-36)	498 MUs, 23 ± 20 (5-116)
	80%	195 MUs, 12 ± 9 (5-65)	141 MUs, 8 ± 10 (5-31)	559 MUs, 33 ± 22 (5 – 113)

Values are group totals, mean ± standard deviation and (minimum-maximum) range across participants. 60% and 80% correspond to limit of stability (LoS) condition.

Table 2. Estimated means of the barycenter position and area of MUs on the muscles of the older adults during the 60% limit of stability condition

MU group	Barycenter direction	Target Directions				
		0°	30°	60°	90°	120°
<i>Medial Gastrocnemius</i>						
Unique	ML(mm)	24.7 (23.0-26.4)	24.5 (22.8-26.1)	25.5 (23.8 – 27.2)	24.4 (22.8 – 26.1)	24.4 (22.7 – 26.1)
	PD (mm)	35.6 (32.8-38.5)	34.2* (32.7 – 38.4)	33.8* (33.1 – 38.8)	35.7 (32.9 – 38.6)	33.4* (35.0 – 4.02)
	Area (mm)	6.31 (5.89 – 7.82)	7.23 (6.23 – 7.99)	6.89 (5.87 – 7.21)	6.84 (5.33 – 7.45)	6.78 (5.28 – 7.43)
Common	ML(mm)	25.6 (23.7 – 27.5)	25.5 (23.6 – 27.4)	25.5 (23.6 - 27.3)	25.1 (23.2 – 26.9)	25.4 (23.5 – 27.3)
	PD (mm)	34.8 (30.4 – 36.5)	33.5 (31.6 – 37.6)	33.3 (30.5 – 36.5)	35.8 (31.8 – 37.8)	34.8 (31.6 – 36.4)
	Area (mm)	5.32 (4.18 – 6.03)	5.12 (4.23 – 5.91)	5.23 (4.54 – 5.89)	5.18 (4.04 – 5.97)	5.32 (4.67 – 6.01)
<i>Lateral Gastrocnemius</i>						
Unique	ML(mm)	28.6 (26.8 – 3.04)	28.4 (26.6 – 3.01)	29.4 (27.7 – 31.1)	28.4 (26.6 – 30.1)	28.3 (26.5 – 3.01)
	PD (mm)	38.1 (3.65 – 4.24)	36.6* (36.5 – 42.4)	36.3* (35.5 – 43.1)	38.2 (35.3 – 41.1)	35.8* (35.8 – 40.9)
	Area (mm)	6.02 (5.44 – 7.73)	8.02 (6.62 – 8.76)	7.32 (6.59 – 8.03)	7.35 (6.34 – 7.99)	7.02 (5.35 – 7.89)
Common	ML (mm)	29.5 (27.5 – 31.4)	28.4 (27.5 – 31.3)	29.4 (27.5 – 31.3)	29.0 (27.1 – 3.08)	29.3 (27.4 – 31.2)
	PD (mm)	37.2 (34.1 – 40.3)	35.9 (32.8 – 39.0)	35.7 (34.2 – 40.2)	34.4 (32.2-38.2)	35.2 (33.1 – 37.4)
	Area (mm)	5.34 (4.23 – 6.12)	6.33 (4.54 – 6.89)	6.18 (5.32 – 6.74)	6.27 (5.43 – 7.05)	5.89 (5.54 – 6.35)
<i>Soleus</i>						
Unique	ML (mm)	44.3 (42.6 – 46.0)	44.1 (42.4 – 45.8)	45.1 (43.5 – 46.8)	44.1 (42.4 – 45.7_	42.3 (42.4 – 45.7)
	PD (mm)	39.4 (36.6 – 42.3)	38.0* (38.0 – 43.7)	37.6* (36.9 – 42.6)	39.5 (36.7 – 42.4)	37.1* (36.5 – 42.2)
	Area (mm)	6.89 (5.32 – 8.23)	7.67 (6.01 – 8.47)	7.46 (5.99 – 8.65)	7.89 (6.48 – 8.92)	8.04 (6.88 – 8.77)
Common	ML (mm)	45.7 (43.9 – 47.6)	45.7 (43.8 – 47.5)	45.6 (43.8 – 47.4)	45.2 (43.4 – 47.0)	45.5 (43.7 – 47.4)
	PD (mm)	38.6 (35.6 – 41.6)	37.3 (34.3 – 40.3)	37.1 (35.6 – 41.5)	35.8 (33.4 – 38.7)	36.6 (34.2 – 39.1)
	Area (mm)	6.14 (5.18 – 7.03)	6.35 (5.23 – 6.89)	6.18 (5.32 – 6.65)	6.34 (5.13 – 6.92)	6.32 (5.76 – 7.08)

Values are mean estimates (confidence intervals) calculated from the linear mixed effect models. See Figure 2 for the orientation of the proximo-distal (PD) and medio-lateral (ML) axes. Statistical significant ($p < 0.05$) by direction is indicated with an (*) with 0 degrees as the comparator. **Bolded** indicates statistically significant ($p < 0.05$) by age group.

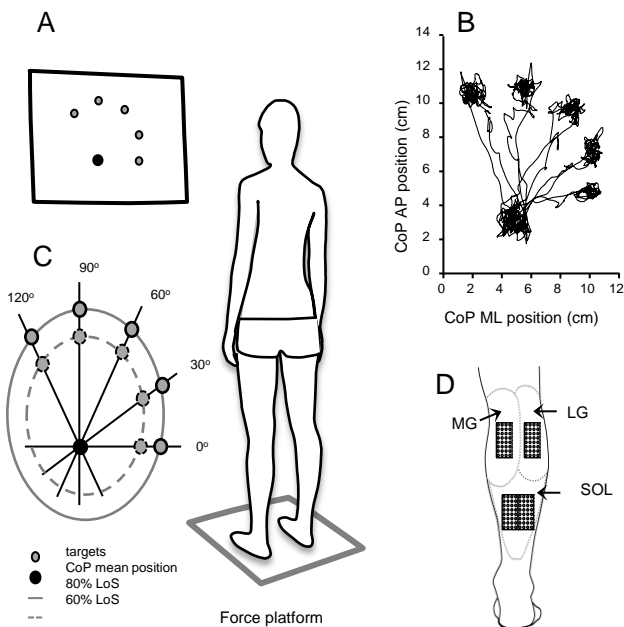
Table 3. CoP outcome measures in each leaning direction and limit of stability condition

CoP measure	Target Directions				
	0°	30°	60°	90°	120°
60% Limit of Stability					
Path Length (cm)	27.0 (21.3 – 32.7)	29.8 (24.1-35.5)	33.7* (28.1 – 39.4)	36.0* (30.3-41.7)	32.8 (27.1 – 38.5)
Mean Velocity (cm/s)	1.27 (0.98 – 1.57)	1.47 (1.18 – 1.76)	1.67* (1.37 – 1.96)	1.78* (1.49 – 2.08)	1.58 (1.26 – 1.87)
95% Ellipse Area (cm ²)	1.47 (0.71 – 2.24)	1.98 (1.21 – 2.75)	1.71 (0.94 – 2.48)	2.04 (1.26 – 2.81)	1.75 (0.98 – 2.52)
SD _{ML} (cm)	0.31 (0.23 – 0.39)	0.34 (0.26 – 0.42)	0.34 (0.25 – 0.41)	0.35 (0.27 – 0.43)	0.32 (0.25 – 0.40)
SD _{AP} (cm)	0.39 (0.32 – 0.47)	0.42 (0.34 – 0.50)	0.38 (0.30-0.45)	0.43 (0.35 – 0. 50)	0.41 (0.34 – 0.49)
80% Limit of Stability					
Path Length (cm)	30.4 (24.6 – 36.1)	33.2 (27.5 – 39.0)	37.1 (31.4 – 42.9)	39.4 (33.7 – 45.2)	36.2 (30.5 – 42.0)
Mean Velocity (cm/s)	1.48 (1.19 – 1.78)	1.68 (1.36 – 1.98)	1.88 (1.58 – 2.17)	1.99 (1.70 – 2.29)	1.79 (1.49 – 2.09)
95% Ellipse Area (cm ²)	1.62 (0.84 – 2.39)	2.13 (1.35 – 2.90)	1.86 (1.08 – 2.63)	2.18 (1.40 – 2.96)	1.89 (1.12 – 2.67)
SD _{ML} (cm)	0.33 (0.25 – 0.41)	0.35 (0.27 – 0.43)	0.34 (0.27 – 0.43)	0.37 (0.29 – 0.45)	0.34 (0.26 – 0.42)
SD _{AP} (cm)	0.41 (0.33 – 0.49)	0.43 (0.36 – 0.51)	0.39 (0.31 – 0.47)	0.44 (0.36 – 0.52)	0.43 (0.35 – 0.51)

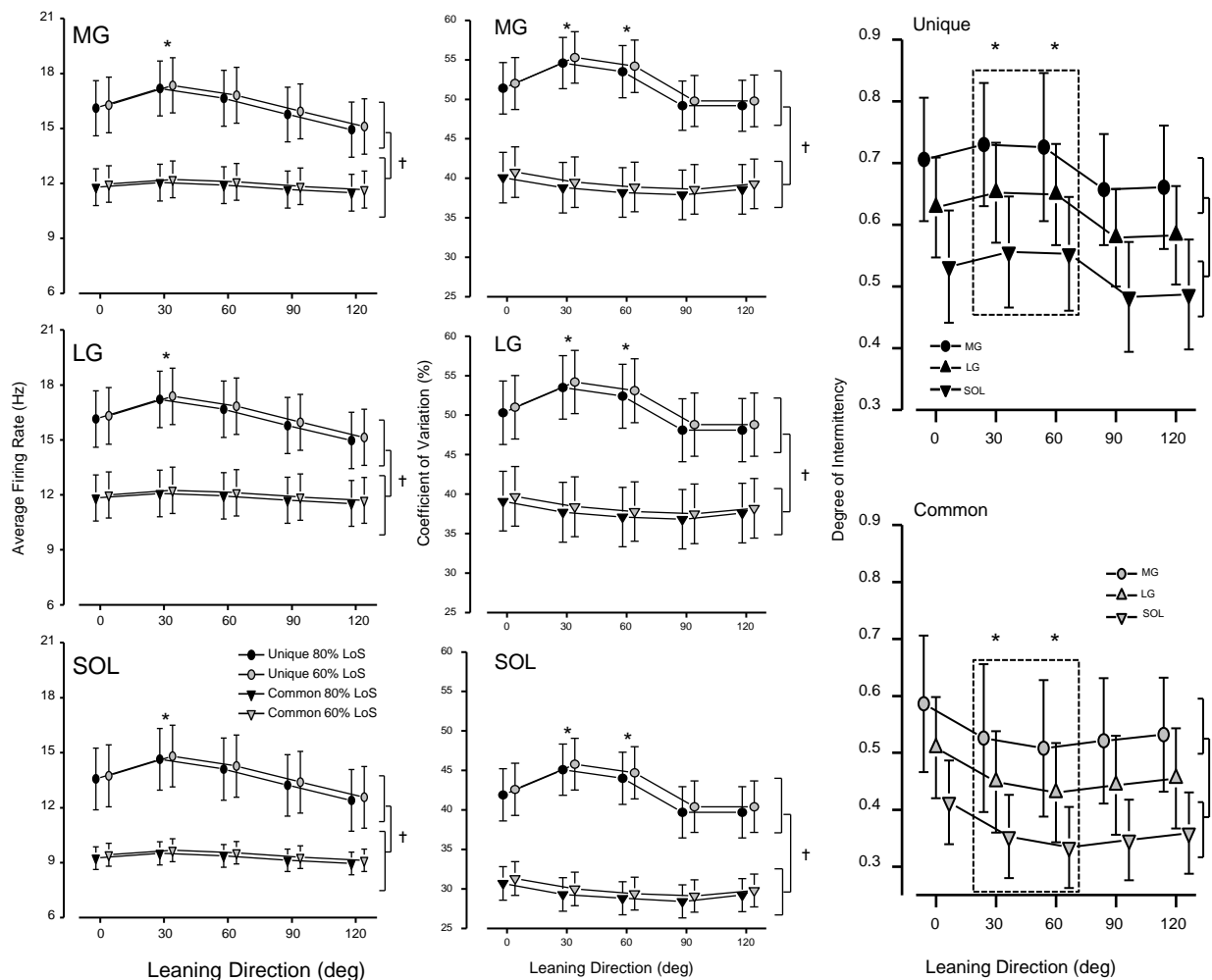
Values are mean estimates (confidence intervals) calculated from the linear mixed effect models. CoP = centre of pressure. SD_{ML} = standard deviation in the medio-lateral direction. SD_{AP} = standard deviation in the antero-posterior direction. * denotes significantly different ($p < 0.05$) from the 0° target direction.

Regional recruitment and differential behaviour of motor units during postural control in older adults

METHODS



RESULTS



CONCLUSION

Older adult's central nervous systems may partially lose the ability to regionally recruit and differentially control motor units. This may be an underlying cause of balance difficulties

Purpose: To determine if older adults regionally recruit MUs, and to evaluate whether the MU firing behaviour in the triceps surae muscle of older adults modulated during a multi-directional leaning task