

Speed and variability of chair stepping and morphological and neural characteristics in community-dwelling older adults

Running head

Chair step and motor unit firing properties

Tetsuya Hirono^{1,2}, Ryosuke Takeda¹, Taichi Nishikawa¹, Masamichi Okudaira^{1,3}, Kohei Watanabe¹

1. Laboratory of Neuromuscular Biomechanics, School of Health and Sport Sciences, Chukyo University
101 Tokodachi, Kaizu-cho, Toyota, Aichi, Japan.
2. Human Health Sciences, Graduate School of Medicine, Kyoto University
53 Kawahara-cho, Shogoin, Sakyo-ku, Kyoto, Japan
3. Faculty of Education, Iwate University, Iwate, Japan
8-18-8, Ueda, Morioka, Iwate, Japan

Corresponding author

Tetsuya Hirono, Ph.D., R.P.T.

Email: hirono.tetsuya.4r@kyoto-u.ac.jp

ORCID: 0000-0002-4337-6249

ABSTRACT

Rapid lower limb coordination is directly related to physical activity skills in older adults and is controlled mainly by the central nervous system. This study aimed to investigate the association between chair stepping skills and neuromuscular properties in older adults. Fifty-eight community-dwelling older adults (72.0 ± 5.0 years) participated. High-density surface electromyography from the vastus lateralis (VL) was recorded and was decomposed into individual motor unit activities. The slope (MU slope) and intercept were calculated from the regression line between the individual motor unit recruitment thresholds and firing rates in each participant. Muscle thickness of the VL and vastus intermedius and echo intensity of the VL were evaluated using ultrasonography. During sitting on a chair, they repeatedly opened and closed their legs to position their feet outside and within 30-cm interval lines rapidly for 20 s. The mean steps were associated with echo intensity ($r=0.388$, $p<0.01$). Step variability was associated with the MU slope ($r=0.324$, $p=0.03$). These results suggest that the rapid lower-limb movement skills would be determined by neuromuscular quality, such as echo intensity and motor unit firing patterns, in older adults. The chair stepping test can be employed as a screening tool as an alternative to specific assessments.

KEYWORDS

Agility, High-density surface electromyography, Physical function test, Motor unit

INTRODUCTION

Older adults' abilities to execute coordinated movements depend to a large extent on the activities they perform in their daily lives, because many daily activities require inter- and intralimb coordination. Aging can influence the decline in abilities (Kang et al., 2022), especially coordination between the lower limbs, which is related to physical activities, such as walking, or the risk of falling (Dite & Temple, 2002; Yamagata et al., 2019). A lack of voluntary coordination is often observed during rapid movements (Barry et al., 2005). Therefore, planning and executing coordinated movements and controlling rapid movements are necessary to maintain and improving physical activities in older adults.

Rapid coordinated movements have been investigated in the upper limbs, hands, and fingers involving various actions (Barry et al., 2005; Park et al., 2012; Temprado et al., 2020), but lower-limb coordination is also important for daily activities in older adults. The chair stepping test, in which older adults perform rapid and repeated opening and closing of their legs, is an easily implemented lower-limb physical function test used to assess agility in older adults (Araki et al., 2016; Kimura et al., 2012). Because the test can be safely performed in a seated position, various populations, including older adults, can perform it, and the number and variability of steps during this test can be used to identify individual characteristics of physical

functions. When bimanual coordination skills are evaluated, less accuracy and greater variability of rapid movements are observed in older adults than in young adults (Temprado et al., 2020), which is associated with an overactivated brain area (Goble et al., 2010) or neural regulations (Zehr et al., 2007). Precise activation of the central nervous system is necessary to coordinate limb movements rapidly and accurately.

Regarding associations with physical function, muscle strength or muscular morphological characteristics have been the focus of research in older adults. Echo intensity, which is used as an index of muscle quality, degenerates with aging (Fukumoto et al., 2018), and is related to the ability to rapidly contract muscles (Hirata et al., 2022; Mota et al., 2018). Thus, physical functions of the lower limbs in older adults are reportedly associated with muscular characteristics (Hill et al., 2021), but it is unclear whether physiological neural components would contribute to physical functions.

An assessment of motor unit activity indicates the physiological neural factors of the pathway from the central nervous system to the muscle. Each motor unit has a different property, depending on its recruitment threshold. Normally, a motor unit is recruited at relatively low-intensity contraction discharges at high frequency, while a motor unit is recruited at relatively high-intensity contraction discharges at low frequency during submaximal isometric

contractions (Erim et al., 1996), indicating that there is a hierarchical pattern of the motor unit firing rate depending on the recruitment threshold. However, a previous study reported that the motor unit firing pattern in older adults was different from that in young adults, indicating that the hierarchical motor unit firing pattern depending on the recruitment threshold was often absent (Watanabe et al., 2016). Additionally, a correlation between the recruitment threshold and firing rate was not observed in patients with Parkinson's disease (Nishikawa et al., 2021), whose motor coordination was impaired (Park et al., 2012). The hierarchical motor unit firing pattern, which can be assessed by the slope from the regression line between the recruitment threshold and firing rate (Hirono, Takeda, Nishikawa, Okudaira, et al., 2024; Hirono, Takeda, Nishikawa, & Watanabe, 2024; Okudaira et al., 2023), would generate inter-individual differences in the physiological characteristics of the neuromuscular system.

The purpose of this study was to investigate whether muscle morphological and motor unit firing properties are associated with speed and variability during chair stepping in community-dwelling older adults. We hypothesized that stepping speed and variability would be associated with muscle echo intensity and motor unit firing properties.

MATERIALS and METHODS

100 Participants

101 Fifty-eight community-dwelling older adults participated in the present study (20 men and 38
102 women, age: 72.0 ± 5.0 yrs, height: 158.3 ± 8.2 cm, body mass: 56.8 ± 9.1 kg). The participants
103 were recruited from community-dwelling older adults with physically independent and in good
104 health. The inclusion criteria were over 60 years old; no acute disease; without any exercise
105 restrictions imposed by medical doctors; without neurological disorder or orthopedic problems
106 that could affect the performances in the present study. The experimental procedures were
107 explained to the participants before they provided informed written consent to participate in
108 this study. This study was approved by the Research Ethics Committee of Chukyo University
109 (2022-017) and conducted in accordance with the Declaration of Helsinki.

110

111 Neuromuscular Functions

112 Maximum knee extensor strength

113 The participants were seated in a custom-made chair to perform maximum voluntary isometric
114 knee extension with hip and knee angles of 90 degrees, and right knee extension strength was
115 measured. The chair was equipped with a dynamometer (Takei Scientific Instruments Co.,
116 Ltd., Niigata, Japan) that was fixed to a force transducer (LU-100KSE; Kyowa Electronic
117 Instruments, Aichi, Japan). Following a familiarization session consisting of several

submaximal voluntary contractions (50%, 70% and 90% of MVC), maximum voluntary isometric contraction (MVC) of knee extension was performed twice. Third trial was additionally performed if the second MVC value was much higher than first MVC value. A greater value was obtained and used to calculate the maximum torque by multiplying the force value and the force length, which was determined as the distance between the knee joint axis and the force transducer. Furthermore, knee extension torque was divided by individual body mass [Nm/kg].

Motor unit firing properties

Submaximal isometric ramp-up contraction to 50% of MVC, consisting of a 17-sec increasing phase and 10-sec sustained phase at 50% of MVC, was performed. The exerted and target levels were displayed on a monitor in real time as visual feedback. During submaximal contraction, high-density surface electromyography (EMG) was recorded from the right vastus lateralis using an EMG acquisition device (Sessantaquattro, OT Bioelettronica, Torino, Italy), and the force signal was obtained at the same time. A 64-channel electrode grid (13 × 5, with one missing electrode in the corner; 1-mm diameter and 8-mm inter-electrode distance; GR08MM1305, OT Bioelettronica) was attached to the skin with a bi-adhesive sheet (KITAD064, OT Bioelettronica) and conductive paste (Elefix Z-181BE, Nihonkohden, Tokyo, Japan). After cleaning the skin with alcohol or water, the grid was attached along the line

between the head of the greater trochanter and the superior and lateral edges of the patella, and the center of the grid was placed at the midpoint of the line. A reference electrode (WS2; OT Bioelettronica) was attached to the tibial tuberosity. Monopolar EMG signals were filtered with a bandpass filter from 10 to 500 Hz, amplified by a factor of 150, and sampled at 2,000 Hz. The recorded EMG signals were transferred to analysis software (MATLAB R2019a, MathWorks GK, MA, USA), and the signals were differentiated between adjacent electrodes in the longitudinal direction. Thus, 59-channel differentiated signals were decomposed and individual motor units were identified by the Convolution Kernel Compensation (CKC) technique using Decomposition of Motor Unit Surface EMG (DEMUSE) software (ver. 6.1; The University of Maribor, Slovenia) (Holobar et al., 2009). A single experimental investigator (TH) manually inspected all the decomposition results. Any physiological irregular motor unit discharge (<4 or >50 Hz) and motor unit showing a coefficient of variation of the firing rate over 30% were discarded (Hirono, Takeda, Nishikawa, Okudaira, et al., 2024; Hirono, Takeda, Nishikawa, & Watanabe, 2024).

Because detected motor units from one individual have various motor unit size at random and firing rates of motor units depend on their recruitment threshold (i.e. motor unit size), firing rates of individual motor units are not used simply in the present study. Instead, normalized index by individual motor unit recruitment thresholds and firing rates are used. The

recruitment thresholds of the motor units were determined from the force level when the motor units first discharged. The median firing rate of individual motor units during 40-50% of MVC was calculated from the interspike intervals, and the regression line between recruitment thresholds and firing rate was described. The slope (MU slope) and y-intercept (MU intercept) were calculated using the regression line (Fig. 1). To ensure the motor unit firing property within an individual participant, participants from whom five or more motor units were detected were analyzed and used for subsequent analysis (Hirono, Takeda, Nishikawa, Okudaira, et al., 2024; Hirono, Takeda, Nishikawa, & Watanabe, 2024; Okudaira et al., 2023).

Muscle thickness and echo intensity

The participants were seated relaxed in a chair with their right knees flexed at 90 degrees without their feet in contact with the ground. B-mode ultrasound images were obtained from the right lateral thigh site using an ultrasound device (MicrUS EXT-1H, TELEMED, Vilnius, Lithuania) with a multifrequency linear array probe (L12-5L40S-3). The equipment settings were a frequency of 7.5 MHz, dynamic range of 66 dB, gain of 72%, and depth of 70 mm. Longitudinal and transverse images were acquired at the midpoint between the head of the greater trochanter and superior and lateral edges of the patella. The images were acquired at minimal pressure when the bone echo was brightest. The muscle thickness of the vastus lateralis and intermedius was measured as the distance between the superficial vastus lateralis

and the bone edge in longitudinal images. Muscle echo intensity of the vastus lateralis was evaluated in the transverse image and computed by the average gray scale in the region of interest, which was determined manually as wide as possible while excluding the surrounding fascia using software (Image J ver. 1.53k; National Institutes of Health, Bethesda, MD, USA). Grayscale values ranged from 0 (black) to 255 (white). A high value indicates a greater amount of fat and fibrous tissue within the muscle (Pillen et al., 2009). Muscle thickness and echo intensity were evaluated as the mean values between two longitudinal and transversal images, respectively.

Chair Stepping Test

According to previous reports (Araki et al., 2016; Kimura et al., 2012), the chair stepping test was performed. The participants sat on a 40-cm-high chair. First, they positioned their feet within 30-cm interval lines. They were instructed to repeat opening their legs in order to position their feet outside the two lines and close their legs to return their feet to the original position for a total period of 20 s as rapidly as possible. They performed symmetry leg movements rapidly over the lines with 30-cm interval. If they step without crossing the line (i.e. steps on the same sheet repeatedly) or they step asymmetry (i.e. only one leg moving over the line but the other not), the step was not counted, thus the interval gets long duration. The number of steps was counted using a step-counting sheet (T.K.K.5301; Takei Scientific

Instruments Co., Ltd., Niigata, Japan). The sheet has two separated sensor mats with widths of 30 cm; one counts the closing step, and the other counts one foot when opening (Fig. 2). The sheet was set on right side to count the timing of right foot striking. The step duration between steps were recorded, and the mean and coefficient of variation of the steps were calculated for a total period of 20 s. The mean steps [sec/step] and coefficient of variation [%] of the steps were interpreted as the speed and variability of the steps in the present study, respectively.

Statistical Analysis

All statistical analyses were performed using SPSS (Statistical Package for Social Sciences ver. 28.0, IBM Japan Inc., Tokyo, Japan). Partial correlation coefficients with sex as a covariate were calculated to identify the associations between step variables (mean steps or coefficient of variation of steps) and neuromuscular variables (maximal knee extension strength, muscle thickness, echo intensity, MU intercept, or MU slope). The Gaussian distribution of all variables was checked using Shapiro–Wilk test. T tests or Mann-Whitney U tests were performed to compare step variables and neuromuscular variables between sex. Significance was set as $p < 0.05$.

RESULTS

Twelve participants were excluded from further analysis because they failed to meet the minimum number of detected motor units, i.e., more than five motor units were needed for the analyses. Thus, the data from 46 participants were used for further analysis. Among these participants, the average number of detected motor units was 12.8 ± 7.7 .

Figure 3 shows the results of the correlation analysis. Echo intensity was significantly correlated with mean steps ($r = 0.388$, $p = 0.008$), but not knee extension strength ($p = 0.242$), muscle thickness ($p = 0.064$), MU intercept ($p = 0.292$), or MU slope ($p = 0.102$). With step variability, the MU slope showed the sole correlation ($r = 0.324$, $p = 0.030$), but not knee extension strength ($p = 0.180$), muscle thickness ($p = 0.782$), echo intensity ($p = 0.477$), or MU intercept ($p = 0.402$).

The comparison between sex were shown in Table 1. Step variables were not different between sex. There are no significant different between sex in MU slope or MU intercept, but significant differences were observed in knee extension strength, muscle thickness and echo intensity.

DISCUSSION

This study investigated the association between morphological and neural characteristics and chair-stepping skills in community-dwelling older adults. The mean step, meaning the speed of chair stepping, was significantly correlated with echo intensity, while step variability, meaning the coordination of lower limbs, was significantly correlated with the MU slope. This is the first study to identify an association between chair stepping variables and neuromuscular properties.

Our results showed a significant correlation between neural characteristics such as the MU slope and step variability (Fig. 3). The ability to control complex movements and execute accurate actions during physical function tests, such as the stepping test, requires the coordinated activation of the central nervous system to control muscle contractions. Indeed, patients with cerebellar ataxia perform inaccurate movements (Mark & Steve, 1993), while patients with Parkinson's disease show neurological degeneration related to motor dysfunction (Park et al., 2012). In addition, the inability to perform rapid alternate movements, such as repeated supination and pronation of the elbow, is often assessed in clinical settings to screen for adiadochokinesis. In patients with Parkinson's disease who have adiadochokinesis, motor unit firing behaviors are different from those in healthy individuals (Nishikawa et al., 2021),

suggesting that such behaviors, which are influenced by degeneration of the central nervous system, could be associated with physical dysfunction. The present study evaluated motor unit firing properties as physiological factors of the neural control system, and it is the first reported study to investigate the relationship between the ability to perform rapid alternating movements in the lower limb and motor unit firing properties. According to the size principle (Erim et al., 1996), motor units with low recruitment thresholds inherently fire in a high rate in general, which can cause a negative correlation between recruitment threshold and firing rate (Hirono, Okudaira, et al., 2024). For example, the participant in Figure 1B and D shows an irregular motor unit firing property, which shows a less negative MU slope, and could be suggested to influence motor performance such as the stepping skill. The MU slope alters with aging because the association between the individual motor unit recruitment threshold and firing rate changes in older adults (Erim et al., 1999; Watanabe et al., 2016). A less negative MU slope (near zero of the slope value) is mainly caused by a decline in the firing rate of motor units with a low recruitment threshold (like Fig.1 D), as various previous studies have reported lower firing rates in older adults, especially in motor units with low recruitment thresholds (Dalton et al., 2010; Rubinstein & Kamen, 2005; Watanabe et al., 2016). A decline in the motor unit firing rate can be caused by elongation of the motor neuron afterhyperpolarization (AHP) duration in the muscle of older individuals (Piotrkiewicz et al., 2007), because elongation of the AHP duration is related to a low firing rate (Christie & Kamen, 2010). On

exerting submaximal voluntary contraction, motor units with a low recruitment threshold generally fire at a high frequency (Erim et al., 1996); therefore, firing patterns of motor units with low recruitment thresholds can be selectively affected by the degeneration of the contraction-relaxation cycle due to the elongation of the AHP duration. According to these perspectives, we consider that the MU slope may be more sensitive to age-related neuromuscular degeneration than the MU intercept, which indicates the firing rates of whole motor units. It is reasonable that the stepping variability was significantly correlated with the MU slope but not with the MU intercept. We suggest that participants with age-related alterations in motor unit firing patterns cannot accurately and rapidly open and close their legs to position their feet. Therefore, the present correlation analysis revealed a significant correlation between the characteristics of the motor unit firing pattern and the capacity to step precisely. The motor unit firing properties would be important for accurate stepping.

The mean step number was only correlated with the echo intensity based on the present results (Fig. 3). Echo intensity is related to physical functions, such as the ability to rapidly perform sitting and standing (Lopez et al., 2017). Our results support previous findings, including the speed of chair-stepping. Moreover, echo intensity is reportedly associated with muscular properties of fast contractions (Hirata et al., 2022; Mota et al., 2018). In contrast, the maximal strength and muscle thickness were not correlated (Fig. 3). We can assume that activities of

daily life do not require maximum strength; thus, maximum strength and muscle thickness were not significantly correlated.

The assessment of motor unit firing properties is a non-invasive measurement that can identify neurophysiological characteristics, but it requires a specific device and mathematical algorithm. Neurophysiological assessments are not common in clinical settings, but this chair stepping test can be an alternative assessment of motor unit firing properties, although the correlation coefficient was not high ($r = 0.324$). The assessment of echo intensity, which also has the merit of being a noninvasive method, requires an ultrasound device and experimental examiner. Although further studies should investigate these relationships, assessment of the chair stepping test revealed the possibility of it being a good screening tool.

There are some limitations in the present study. It is difficult to record high-density EMG signals from hip muscles because there are extensive subcutaneous tissues. Thus, high-density EMG was obtained from the knee extensor muscle in the present study; however, chair stepping requires hip movements and not knee extension. High-density surface EMG obtained from knee extensor and chair stepping tests were used to characterize neural controls of motor unit properties and coordination, rather than to biomechanically assess the relationship between knee extensors and chair stepping skill. Furthermore, since knee extension strength

has been shown to reflect overall lower-limb strength (Bohannon, 2009), our results would reflect individual lower-limb characteristics. Additionally, the variables of neuromuscular properties could be potentially sensitive to sex (Hirono, Okudaira, et al., 2024; Lulic-Kuryllo & Inglis, 2022). To consider the influence of sex on the present result, partial correlations with sex as a covariate were performed. Indeed, if we separate male and female individuals and Pearson's or Spearman's correlation coefficients were calculated, a significant correlation between echo intensity and mean steps is observed in males ($r = 0.547$, $p = 0.013$), but that is not observed in females ($r = 0.336$, $p = 0.093$). There are no significant correlations between step variability and MU slope in either males ($\rho = 0.335$, $p = 0.148$) or females ($r = 0.289$, $p = 0.153$). However, when separating the participants by sex, the sample size in each group would become small. Since there are no significant differences in step variables or motor unit properties (Table 1), the present result including both sex is considered reasonable.

In conclusion, the present study investigated the association between speed and variability during chair stepping and neuromuscular characteristics were evaluated using high-density EMG and ultrasound images in community-dwelling older adults. The stepping speed was associated with the echo intensity, and the variability in stepping was associated with the MU slope. These results suggest that the skill of performing rapid lower-limb movements would be determined by neuromuscular quality, such as echo intensity and motor unit firing patterns,

327 in older adults. Although the correlation coefficients were not high and it should be noted that
328 the indicators reflect only part, not all, of neuromuscular properties, the chair stepping test can
329 be employed as a screening tool as an alternative to specific assessments.

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Declaration of interest statement

The authors report there are no competing interests to declare.

Data availability statement

Data will be provided by the corresponding author upon request.

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FIGURE LEGENDS

Figure 1

Representative motor unit firing data from two participants. Gray lines represent exerted force and red lines represent individual motor unit firing rates. A and B show the firing rates of individual motor units during ramp-up contraction to 50% of maximum voluntary contraction (MVC). The recruitment threshold was defined as the force level at which the motor unit fired first. The firing rate was calculated as the median value at 40-50% of MVC. C and D show the regression lines described by the recruitment threshold and firing rate. From the regression lines, the slope (MU slope) and y-intercept (MU intercept) were calculated and used for further analysis.

Figure 2

The chair stepping test was performed as described in A. Two representative participants' data are shown in B and C. The step counting sheet measured the foot step times when both feet were placed on one sheet (dark gray mat in A, dark gray circles in B and C) and when one foot was moved to another sheet (light dark mat in A, light gray circles in B and C). The mean step time and coefficient of variation in step time (step variability) were calculated during 20-sec stepping.

Figure 3

Results of partial correlation coefficients with sex as a covariate. The vertical axis in the upper row indicates the mean step time [steps/sec] and that in the lower row indicates the coefficient of variation in the step time [%]. From the left column, maximum knee extension strength [Nm/kg], muscle thickness [mm], echo intensity [a.u.], MU intercept [pps], and MU slope [pps/%MVC] are indicated. Filled circles: men, unfilled circles: women. MVC: maximum voluntary contraction. MU: motor unit.

Table 1. Comparison of results for each measurement outcome between sex.

	Male (n=20)			Female (n=26)			p value
Mean steps, step/sec	0.31	±	0.05	0.30	±	0.07	0.430 ^a
Step variability, %	27.9	±	17.1	28.6	±	19.2	0.894 ^a
Knee extension strength, Nm/kg	2.12	±	0.32	1.83	±	0.37	0.008 ^a
Muscle thickness, mm	44.5	±	4.8	37.9	±	7.1	< 0.001 ^a
Echo intensity, a.u.	50.2	±	6.4	58.7	±	8.5	< 0.001 ^a
MU intercept, pulse per sec (pps)	14.47	±	4.40	14.45	±	3.76	0.773 ^b
MU slope, pps/%MVC	-0.10	±	0.09	-0.07	±	0.10	0.706 ^b

a; tested using t-test. b; tested using Mann-Whitney U tests.





