

Assessment of the mechanical characteristics of the lower extremity muscles with myotonometric measurements in healthy individuals

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Abstract

Introduction. This study aimed to establish normative values for stiffness, tone, and elasticity of lower extremity muscles and to determine age and sex differences.

Methods. Overall, 389 participants (199 females, 190 males) were included. The mechanical properties of the tibialis anterior (TA), rectus femoris (RF), vastus lateralis (VL), vastus medialis obliquus (VMO), gastrocnemius lateralis (GL), and gastrocnemius medialis (GM) muscles were assessed bilaterally with the MyotonPRO device. The data were compared with respect to sex and age.

Results. The mechanical properties were significantly higher bilaterally in males than females in all parameters ($p < 0.05$). Right TA stiffness and elasticity, GM tone and stiffness were higher than left in females ($p < 0.05$). Greater VMO elasticity and RF tone were found in left vs. right ($p < 0.05$). Right GL-VL stiffness and elasticity were greater than left in males ($p < 0.05$). VMO-RF tones were bigger in left compared with right ($p < 0.05$). Among age groups, right-left TA tone and stiffness were greater in group 1 (18–28 years old) ($p < 0.05$). Right-left VL-VMO tones, and left RF stiffness and tone were lower in group 2 (29–39 years old) ($p < 0.05$). Right GM-VMO stiffness, right-left TA-GL-GM-VMO and RF elasticity were greater in group 3 (40–50 years old) compared with others ($p < 0.05$).

Conclusions. The mechanical properties of leg muscles were bilaterally higher in males. Different characteristics were observed of different muscles for both sexes. GL-GM-VMO-RF elasticity decreased, and TA stiffness and VMO tone increased with advancing age in both legs.

Key words: muscle tone, viscoelastic properties, age factors

Introduction

Skeletal muscles have complex features and functions. With aging, significant changes occur in the function and physiology of the muscles [1]. Age-related structural and biochemical changes in the cellular and extracellular matrix include muscle fibre redistribution, altered protein concentration, changes in cross-bridges, and degeneration [1–3]. This age-related process called sarcopenia is associated with reduced muscle mass and strength. While females have less muscle mass and strength than males, they have higher endurance. This difference in functional capacity is actually a result of the loss of type II (fast twitching) muscle fibres, particularly in the peripheral areas. Aging has no impact on type I (slow twitching) muscle fibres.

Physiological changes in the muscle affect its mechanical properties such as tone, elasticity, stiffness, causing reduced elasticity and increased stiffness of the muscle [4, 5]. It is important to evaluate the changes in the mechanical properties of the muscles, which can limit physical performance and impair quality of life. It is difficult to objectively distinguish whether any changes in the muscle tone or its mechanical properties are due to aging or a result of neurodegenerative disorders [6].

Technological devices are used for the assessment of the mechanical properties of the musculoskeletal system. Electromyography (EMG) is one of the best known and commonly used methods to evaluate muscle functions objectively. Total muscle activity can be determined by using surface (skin) or needle (inserted into the muscle) electrodes. The isolated

muscle activity can only be recorded with an EMG device which involves an invasive and painful procedure [7, 8]. Other modalities such as elastography, shear wave ultrasound elastography, or free oscillation techniques are valid and reliable tools for quantifying the mechanical properties of muscles and tendons. However, access to these systems is not always possible or may be limited in clinics because of high purchasing and maintenance costs and the requirement of technical expertise [9]. Thus, there is a need for objective, cost-effective, reliable, valid, and easy-to-use methods to evaluate the mechanical properties of the musculoskeletal system [9, 10]. More recently, a new hand-held device known as MyotonPRO (Müomeetria Ltd., Tallinn, Estonia) was introduced. MyotonPRO offers a non-invasive, cost-effective, and quantitative method to measure the mechanical properties of the musculoskeletal system [11, 12]. This portable, feasible, and user-friendly device has shown good to excellent reliability in healthy individuals [11, 13], athletes [14], elderly [15], cancer and stroke patients [16–18], and patients with neurodegenerative disease [19]. The objective measurement of soft tissue stiffness, tone, and elasticity provided by MyotonPRO has high test-retest reliability and repeatability [9, 20–22]. MyotonPRO may be used as a tool for quick assessment of soft tissue properties, as well as a convenient diagnostic and monitoring device in small-volume clinics and research laboratories [23–26]. There are many studies that investigated age- and sex-related changes in the mechanical properties of the musculoskeletal system reporting variable results [27, 28].

The purpose of this study was to establish normative data for stiffness, tone, and elasticity of the lower extremity muscles

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and to determine age and sex differences in a healthy population. It was hypothesized that advanced age would increase muscle stiffness and tone and decrease muscle elasticity.

Subjects and methods

Participants

A total of 390 healthy individuals with an age range of 18–50 years (mean age: 28.64 ± 9.68 years), including 200 females (mean age: 29.92 ± 9.63 years) and 190 males (mean age: 27.29 ± 9.57 years) were involved in the study. A power analysis was performed to estimate the sample size on the basis of the confidence interval of the mean. The MedCalc software, version 18.11.3, was used for statistical analyses [29]. Healthy individuals with current systemic or metabolic diseases, a history of psychiatric illness or drug abuse, any conditions associated with muscle disorders or muscle atrophy, or a recent (within the previous 3 months) surgery involving the musculoskeletal system were excluded from the study.

One individual was excluded from the study because of a recent surgical operation, which resulted in a total sample of 389 subjects. The participants were divided into 3 different subcategories by sex (male and female), leg dominance (right and left leg dominant), and age.

The age range of the subgroups was as follows: group 1: 18–28 years ($n = 235$), group 2: 29–39 years ($n = 85$), and group 3: 40–50 years ($n = 69$). Among the 190 male participants, 131 (55.7%) were in group 1, 29 (34.1%) in group 2, and 30 (43.5%) in group 3. Among the 199 female participants, 104 (44.3%) were in group 1, 56 (65.9%) in group 2, and 39 (56.5%) in group 3.

The study was conducted in Hasan Kalyoncu University, Faculty of Health Sciences. The nature and purpose of the study was explained to the participants.

Method

The physical characteristics and demographic data of the participants were recorded before myotonometric mea-

surements. The dominant leg was determined as the one used for kicking a ball. Muscle tone, stiffness, and elasticity were measured bilaterally for the specific muscles in the lower extremity (Table 1) (Figure 1). The mean value of 3 consecutive measurements was recorded as the final value. A MyotonPRO device (Müomeetria Ltd., Tallinn, Estonia) was used for the assessment of muscle tone, elasticity, and stiffness. The device provides data about these 3 mechanical features of any muscle: (1) tone [f]; oscillation frequency (Hz) refers to the passive or resting tone (intrinsic tension) of a muscle without any voluntary contraction [12]; (2) stiffness [S]; stiffness (N/m) is a biomechanical feature of a muscle that characterizes its resistance to a contraction or an external force that disrupts its initial state [12]; (3) elasticity [D]; elasticity is measured as a logarithmic reduction of the natural oscillations in a tissue. Elasticity is a biomechanical feature that characterizes the ability of a tissue to restore its initial state after a contraction or an impact of an external force. It describes the diffusion of mechanical energy within the oscillation cycle [12].

When placed vertically on the muscle, a probe (3-mm diameter) creates constant pre-excitations (0.18 N) and generates short-term (15 ms), low-force (0.4 N) mechanical stimu-

Table 1. The measurement reference points of muscles

Muscles	Reference points
Rectus femoris (RF)	2/3 of the anterior superior iliac spine and patella [6]
Vastus lateralis (VL)	25 cm below the trochanter major
Vastus medialis obliquus (VMO)	18 cm above the medial edge of the midpoint of patella
Tibialis anterior (TA)	12 cm below the lateral condyle of femur
Gastrocnemius lateralis (GL)	30 cm above the posterior edge of the lateral malleolus
Gastrocnemius medialis (GM)	30 cm above the posterior edge of the medial malleolus

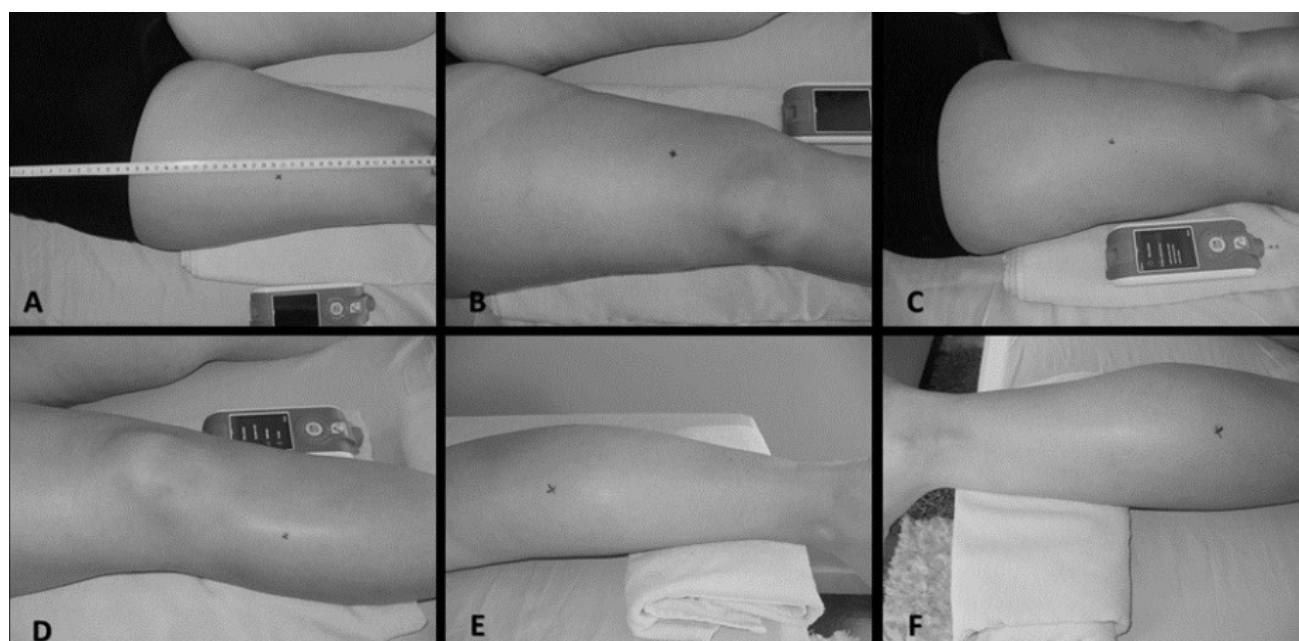


Figure 1. The reference points of muscles for myotonometric assessment: (A) rectus femoris, (B) vastus medialis obliquus, (C) vastus lateralis, (D) tibialis anterior, (E) gastrocnemius medialis, (F) gastrocnemius lateralis



Figure 2. Examples of myotonometric assessment: (a, b) rectus femoris (c) tibialis anterior (d) gastrocnemius lateralis

lations. These stimulations induce damped natural oscillations in the tissue and the device records these oscillations with an accelerometer. Muscle tone, elasticity, and stiffness are calculated separately by the device [30].

Reference points for myotonic measurement

The measurements were obtained from the assumed reference points of the muscles in supine and prone positions (Table 1, Figure 2). Study legs were randomly measured for each participant, i.e. no criteria were applied for starting measurements at the right or left leg. The arithmetic mean of 3 consecutive measurements was recorded as the final value.

The participants were asked to either drive or use public transportation to come to the laboratory for the measurements and avoid intense activities that could affect the muscles, such as cycling, running, or brisk walking.

Statistical analysis

Descriptive statistics were summarized as mean \pm standard deviation for numerical data. The Shapiro-Wilk test was used to check whether the data were normally distributed. For non-normally distributed data, the Mann-Whitney U test was applied to compare the differences between males and females (sex), and right and left legs (leg dominance). In addition, all individuals were subcategorized by age. When 3 or more groups were considered (e.g. in the case of analysis by age), the Kruskal-Wallis test served to compare non-normally distributed variables. If Kruskal-Wallis test yielded a significant p value, post-hoc pairwise comparisons (after Dunn correction) were used to determine the source of the difference. In addition, the Spearman rank correlation test established correlations between numerical variables. The magnitude of the correlations was interpreted as follows: below 0.499: poor, 0.500–0.699: moderate, 0.700–0.899: good, and 0.900–1.000: excellent [31]. The SPSS software (Windows version 24) was used for statistical analyses and a p value less than 0.05 was considered statistically significant.

Ethical approval

The research related to human use has complied with all the relevant national regulations and institutional policies, has followed the tenets of the Declaration of Helsinki, and has been approved by the Ethics Committee for Non-Interventional Research of the Faculty of Health Sciences, Hasan Kalyoncu University, on 14.06.2019 (No. 2019-61).

Informed consent

Informed consent has been obtained from all individuals included in this study.

Results

The descriptive statistics of age, height, body weight, and body mass index (BMI) with respect to sex are shown in Table 2. Females were older than males ($p = 0.002$); however, males were taller ($p = 0.001$) and heavier ($p = 0.001$) than females. Males and females had similar BMI values ($p = 0.408$).

Comparison of the mechanical properties of lower extremity muscles in all individuals

The tone and elasticity of tibialis anterior (TA) were greater in the right legs vs. the left legs ($p = 0.024$, $p = 0.043$, respectively). Gastrocnemius lateralis (GL) stiffness was greater in the left legs than in the right legs ($p = 0.003$). Gastrocnemius medialis (GM) tone was greater in the left legs than in the right legs ($p = 0.010$). Vastus lateralis (VL) elasticity was greater in the right legs vs. the left legs ($p = 0.003$). Vastus medialis obliquus (VMO) elasticity was greater in the left legs vs. the right legs ($p = 0.001$). Left legs showed greater rectus femoris (RF) tone than the right legs ($p = 0.010$). RF elasticity of the right legs was greater than of the left legs ($p = 0.047$). No statistically significant difference was observed for the remaining parameters ($p > 0.05$) (Table 3).

Table 2. Demographic characteristics of participants

Parameter	All individuals (<i>n</i> = 389) (mean ± <i>SD</i>)	Males (<i>n</i> = 190) (mean ± <i>SD</i>)	Females (<i>n</i> = 199) (mean ± <i>SD</i>)	<i>z</i>	<i>p</i>
Age (year)	28.64 ± 9.68	27.29 ± 9.57	29.92 ± 9.63	-3.064	0.002
Height (m)	1.69 ± 0.86	1.75 ± 0.07	1.64 ± 0.06	-13.614	0.001
Body weight (kg)	71.91 ± 14.42	76.11 ± 13.98	67.9 ± 13.72	-5.399	0.001
BMI (kg/m ²)	25.14 ± 4.58	24.87 ± 4.06	25.40 ± 5.02	-0.827	0.408

BMI – body mass index. Bold denotes statistically significant values.

Table 3. Comparison of the mechanical properties of lower extremity muscles in all individuals

Parameter	All participants (<i>n</i> = 389)			
	Right (mean ± <i>SD</i>)	Left (mean ± <i>SD</i>)	<i>z</i>	<i>p</i>
TA tone (Hz)	18.5 ± 3.38	18.27 ± 3.32	-2.256	0.024
TA stiffness (N/m)	365.27 ± 85.03	360.57 ± 80.5	-1.603	0.109
TA elasticity (log)	1.13 ± 0.32	1.1 ± 0.32	-2.023	0.043
GL tone (Hz)	15.34 ± 1.75	15.44 ± 1.92	-1.776	0.076
GL stiffness (N/m)	267.68 ± 37.84	274.35 ± 53.33	-3.023	0.003
GL elasticity (log)	1.18 ± 0.27	1.21 ± 0.26	-1.886	0.059
GM tone (Hz)	15.17 ± 1.9	15.02 ± 1.89	-2.572	0.010
GM stiffness (N/m)	256.29 ± 35.53	254.24 ± 37.07	-1.095	0.273
GM elasticity (log)	1.32 ± 0.29	1.3 ± 0.3	-1.418	0.156
VL tone (Hz)	15.1 ± 2.67	15.02 ± 2.66	-0.765	0.444
VL stiffness (N/m)	260.1 ± 61.31	258.17 ± 62.05	-0.824	0.410
VL elasticity (log)	1.25 ± 0.34	1.21 ± 0.33	-2.994	0.003
VMO tone (Hz)	12.87 ± 1.74	13.07 ± 2.37	-1.717	0.086
VMO stiffness (N/m)	202.51 ± 40.3	205.71 ± 45.22	-1.652	0.099
VMO elasticity (log)	1.16 ± 0.33	1.22 ± 0.37	-3.779	0.001
RF tone (Hz)	14.11 ± 1.78	14.27 ± 1.74	-2.559	0.010
RF stiffness (N/m)	233.33 ± 41.08	236.05 ± 45.71	-1.304	0.192
RF elasticity (log)	1.16 ± 0.31	1.13 ± 0.28	-1.983	0.047

TA – tibialis anterior, GL – gastrocnemius lateralis, GM – gastrocnemius medialis, VL – vastus lateralis, VMO – vastus medialis obliquus, RF – rectus femoris

Bold denotes statistically significant values.

Comparison of the mechanical properties of lower extremity muscles depending on leg dominance

The comparison of right and left lower extremity muscles according to leg dominance revealed no significant differences in the mechanical properties in the participants with left or right leg dominance (*p* > 0.05) (Table 4).

Comparison of the mechanical properties of lower extremity muscles depending on sex

The comparison of right and left lower extremity muscles according to sex revealed greater values in males for all of the mechanical properties (*p* < 0.05) (Table 5).

Comparison of the mechanical properties of lower extremity muscles in male and female groups separately

In males, left legs showed greater GL stiffness and elasticity than the right legs (*p* = 0.001, *p* = 0.015, respectively). VL

elasticity was greater in the right legs vs. the left legs (*p* = 0.002). VMO tone and elasticity were greater in the left legs than in the right legs (*p* = 0.002, *p* = 0.006, respectively). No significant differences were found for other parameters (*p* > 0.05).

In females, right leg TA stiffness and elasticity were greater vs. the left leg (*p* = 0.017, *p* = 0.020, respectively). GM tone and stiffness were greater in the right legs than in the left legs (*p* = 0.030, *p* = 0.039, respectively). VMO elasticity and RF tone of the left legs were greater vs. the right legs (*p* = 0.008, *p* = 0.006, respectively). Other parameters did not show a statistically significant difference (*p* > 0.05) (Table 6).

Comparison of the mechanical properties of lower extremity muscles depending on age

When data were compared between age groups (males and females combined to eliminate sex effect), the post-hoc analysis showed significantly greater right-left TA muscle tone and stiffness in group 1 than in group 2 (right side, *p* = 0.000, *p* = 0.000; left side, *p* = 0.007, *p* = 0.007, respectively). No significant differences were found between group 2 vs. group 3

Table 4. Comparison of the mechanical properties of lower extremity muscles depending on leg dominance

Parameter	Right (n = 366) (mean ± SD)	Left (n = 23) (mean ± SD)	z	p
Right TA tone (Hz)	18.47 ± 3.37	19.02 ± 3.68	-0.543	0.587
Right TA stiffness (N/m)	364.44 ± 84.82	378.48 ± 89.19	-0.569	0.570
Right TA elasticity (log)	1.13 ± 0.32	1.1 ± 0.3	-0.350	0.726
Left TA tone (Hz)	18.25 ± 3.32	18.63 ± 3.44	-0.561	0.575
Left TA stiffness (N/m)	359.93 ± 80.52	370.83 ± 81.18	-0.591	0.555
Left TA elasticity (log)	1.1 ± 0.32	1.1 ± 0.24	-0.322	0.747
Right GL tone (Hz)	15.34 ± 1.76	15.27 ± 1.59	-0.027	0.979
Right GL stiffness (N/m)	267.55 ± 38.26	269.7 ± 30.99	-0.241	0.810
Right GL elasticity (log)	1.18 ± 0.27	1.21 ± 0.25	-0.786	0.432
Left GL tone (Hz)	15.42 ± 1.91	15.76 ± 2.01	-0.969	0.332
Left GL stiffness (N/m)	272.56 ± 42.3	302.74 ± 39.88	-0.489	0.625
Left GL elasticity (log)	1.21 ± 0.26	1.24 ± 0.24	-1.040	0.298
Right GM tone (Hz)	15.16 ± 1.9	15.34 ± 1.84	-0.466	0.641
Right GM stiffness (N/m)	256.12 ± 35.4	259.04 ± 38.27	-0.107	0.915
Right GM elasticity (log)	1.32 ± 0.29	1.33 ± 0.22	-0.546	0.585
Left GM tone (Hz)	15 ± 1.89	15.5 ± 1.91	-1.124	0.261
Left GM stiffness (N/m)	253.52 ± 36.67	265.57 ± 42.14	-1.184	0.237
Left GM elasticity (log)	1.3 ± 0.3	1.3 ± 0.31	-0.113	0.910
Right VL tone (Hz)	15.12 ± 2.7	14.68 ± 1.98	-0.594	0.553
Right VL stiffness (N/m)	260.5 ± 62.08	253.7 ± 47.92	-0.440	0.660
Right VL elasticity (log)	1.25 ± 0.34	1.26 ± 0.38	-0.032	0.975
Left VL tone (Hz)	15 ± 2.67	15.3 ± 2.46	-0.516	0.606
Left VL stiffness (N/m)	257.42 ± 62.43	270.17 ± 55.56	-1.158	0.247
Left VL elasticity (log)	1.21 ± 0.33	1.26 ± 0.31	-0.944	0.345
Right VMO tone (Hz)	12.89 ± 1.75	12.57 ± 1.63	-0.750	0.453
Right VMO stiffness (N/m)	202.26 ± 40.26	206.52 ± 41.62	-0.187	0.851
Right VMO elasticity (log)	1.16 ± 0.33	1.25 ± 0.34	-1.069	0.285
Left VMO tone (Hz)	13.11 ± 2.41	12.48 ± 1.37	-1.034	0.301
Left VMO stiffness (N/m)	205.91 ± 45.89	202.48 ± 33.47	-0.078	0.938
Left VMO elasticity (log)	1.22 ± 0.37	1.27 ± 0.38	-0.397	0.692
Right RF tone (Hz)	14.12 ± 1.79	13.97 ± 1.53	-0.440	0.660
Right RF stiffness (N/m)	233.55 ± 41.26	229.74 ± 38.89	-0.748	0.455
Right RF elasticity (log)	1.15 ± 0.31	1.21 ± 0.35	-0.803	0.422
Left RF tone (Hz)	14.26 ± 1.76	14.42 ± 1.4	-0.549	0.583
Left RF stiffness (N/m)	234.6 ± 41.89	259.09 ± 84.57	-1.208	0.227
Left RF elasticity (log)	1.13 ± 0.28	1.21 ± 0.33	-1.222	0.222

TA – tibialis anterior, GL – gastrocnemius lateralis, GM – gastrocnemius medialis, VL – vastus lateralis, VMO – vastus medialis obliquus, RF – rectus femoris

and between group 1 vs. group 3 ($p > 0.05$). While right GM muscle stiffness was significantly greater in group 3 vs. group 2 ($p = 0.012$), no significant differences were found between group 1 vs. group 2 and between group 1 vs. group 3 ($p > 0.05$). Right-left VL and VMO muscle tones were significantly greater in group 1 vs. group 2 (right side, $p = 0.009$, $p = 0.004$; left side, $p = 0.003$, $p = 0.002$, respectively), with

no significant differences between group 1 vs. group 3 and between group 2 vs. group 3 ($p > 0.05$). Left RF muscle tones of groups 1 and 3 were significantly greater vs. group 2 ($p = 0.007$, $p = 0.009$, respectively) and no significant difference was found between group 1 and group 3 ($p > 0.05$). Although left RF muscle stiffness of group 3 was significantly greater vs. group 2 ($p = 0.005$), no significant differences were found

Table 5. Comparison of the mechanical properties of lower extremity muscles depending on sex

Parameter	Right				Left			
	Males (n = 190) (mean ± SD)	Females (n = 199) (mean ± SD)	z	p	Males (n = 190) (mean ± SD)	Females (n = 199) (mean ± SD)	z	p
TA tone (Hz)	20.95 ± 2.66	16.16 ± 2.1	-14.525	0.001	20.63 ± 2.65	16.02 ± 2.11	-14.468	0.001
TA stiffness (N/m)	405.65 ± 74.17	326.72 ± 76.54	-10.222	0.001	403.59 ± 63.66	319.49 ± 73.19	-11.114	0.001
TA elasticity (log)	1.2 ± 0.38	1.06 ± 0.23	-3.663	0.001	1.18 ± 0.38	1.03 ± 0.22	-3.736	0.001
GL tone (Hz)	16.18 ± 1.54	14.53 ± 1.55	-9.659	0.001	16.34 ± 1.78	14.58 ± 1.64	-9.184	0.001
GL stiffness (N/m)	279.38 ± 35.56	256.5 ± 36.62	-5.803	0.001	290.86 ± 60.82	258.58 ± 39.12	-6.739	0.001
GL elasticity (log)	1.24 ± 0.29	1.13 ± 0.24	-3.322	0.001	1.29 ± 0.29	1.13 ± 0.2	-5.781	0.001
GM tone (Hz)	16.22 ± 1.74	14.18 ± 1.46	-11.056	0.001	16.07 ± 1.59	14.03 ± 1.6	-11.198	0.001
GM stiffness (N/m)	269.05 ± 33.85	244.11 ± 32.78	-7.484	0.001	269.46 ± 34.2	239.7 ± 33.78	-8.577	0.001
GM elasticity (log)	1.41 ± 0.3	1.24 ± 0.24	-5.473	0.001	1.39 ± 0.32	1.22 ± 0.26	-5.376	0.001
VL tone (Hz)	16.94 ± 2.01	13.33 ± 1.91	-14.190	0.001	16.87 ± 2	13.25 ± 1.89	-14.160	0.001
VL stiffness (N/m)	298.66 ± 47.58	223.29 ± 49.15	-13.281	0.001	297.52 ± 46.69	220.61 ± 50.59	-13.643	0.001
VL elasticity (log)	1.43 ± 0.32	1.08 ± 0.27	-10.304	0.001	1.36 ± 0.3	1.06 ± 0.29	-9.702	0.001
VMO tone (Hz)	13.86 ± 1.58	11.93 ± 1.31	-11.720	0.001	14.37 ± 2.5	11.83 ± 1.36	-12.846	0.001
VMO stiffness (N/m)	225.89 ± 37.6	180.2 ± 28.42	-11.667	0.001	229.59 ± 44.4	182.9 ± 32.49	-10.695	0.001
VMO elasticity (log)	1.26 ± 0.29	1.07 ± 0.35	-7.099	0.001	1.34 ± 0.37	1.12 ± 0.34	-6.819	0.001
RF tone (Hz)	14.83 ± 1.38	13.42 ± 1.85	-8.374	0.001	14.91 ± 1.4	13.65 ± 1.81	-8.046	0.001
RF stiffness (N/m)	252.6 ± 32.62	214.93 ± 39.94	-9.577	0.001	255.62 ± 43	217.37 ± 40.14	-9.130	0.001
RF elasticity (log)	1.28 ± 0.29	1.04 ± 0.28	-7.889	0.001	1.25 ± 0.26	1.02 ± 0.26	-8.479	0.001

TA – tibialis anterior, GL – gastrocnemius lateralis, GM – gastrocnemius medialis, VL – vastus lateralis, VMO – vastus medialis obliquus, RF – rectus femoris

Bold denotes statistically significant values.

between group 1 vs. group 2 and between group 1 vs. group 3 ($p > 0.05$).

Group 3 showed significantly greater muscle elasticity of the right-left TA, GL, GM (group 1, right side, $p = 0.001$, $p = 0.001$, $p = 0.001$; left side, $p = 0.001$, $p = 0.001$, $p = 0.001$; group 2, right side, $p = 0.000$, $p = 0.002$, $p = 0.007$; left side, $p = 0.001$, $p = 0.007$, $p = 0.011$, respectively), right VMO (group 1, $p = 0.000$; group 2, $p = 0.003$), and left RF (group 1, $p = 0.001$; group 2, $p = 0.029$) in comparison with other age groups. No significant difference was found between group 1 and group 2 ($p > 0.05$). Right RF muscle elasticity of group 3 was significantly higher vs. group 2 ($p = 0.001$) but there were no significant differences between group 1 vs. group 2 or between group 1 vs. group 3 ($p > 0.05$). Left VMO muscle elasticity of groups 2 and 3 were significantly greater than in group 1 ($p = 0.039$, $p = 0.000$, respectively), with no significant difference between group 2 and group 3 ($p > 0.05$). The comparison of mechanical properties among age groups is shown in Table 7.

A poor positive correlation was found between right-left GL elasticity and age ($r = 0.229$, $p = 0.001$; $r = 0.194$, $p = 0.001$, respectively). A poor positive correlation was observed between right-left GM elasticity and age ($r = 0.157$, $p = 0.002$; $r = 0.133$, $p = 0.001$, respectively). A poor negative correlation was detected between right VL tone and age ($r = -0.123$, $p = 0.015$). While there was a poor negative correlation between right VMO tone and age ($r = -0.154$, $p = 0.002$), VMO elasticity showed a poor positive correlation with age ($r = 0.169$, $p = 0.001$). A poor positive correlation was established between right-left VMO elasticity and age ($r = 0.169$, $p = 0.001$;

$r = 0.208$, $p = 0.001$, respectively). A poor positive correlation was noticed between right-left RF elasticity and age ($r = 0.160$, $p = 0.002$; $r = 0.140$, $p = 0.001$, respectively). Correlations between leg muscle mechanical properties and age are presented in Table 8.

Discussion

The present study was conducted to establish normative data for stiffness, tone, and elasticity of the lower extremity muscles and to investigate age and sex differences. This study is the first to report mechanical properties of 6 different muscles in both legs. A total of 389 healthy individuals, including 199 females and 190 males, in the age range of 18–50 years were involved in the study. While females were older than males, BMI values were similar between the sexes.

Mechanical properties can be used to define muscle mechanical firmness when skeletal muscles are in a relaxed position and steady-state condition. The RF muscle has been most commonly investigated in previous research. In our study, the mean values of the examined mechanical parameters of the RF muscles were lower in healthy participants than those reported for older adults in previous papers [6, 15]. A direct comparison of data for other muscles is not possible owing to differences in the experimental design, study population, and age groups.

Our results showed greater muscle tones for right TA, left GM and RF than their counterparts in all participants. The muscle tone can be classified as neural and non-neu-

Table 6. Comparison of the mechanical properties of lower extremity muscles in males and females

Parameter	Males (n = 190)				Females (n = 199)			
	Right (mean ± SD)	Left (mean ± SD)	z	p	Right (mean ± SD)	Left (mean ± SD)	z	p
TA tone (Hz)	20.95 ± 2.66	20.63 ± 2.65	-1.656	0.098	16.16 ± 2.1	16.02 ± 2.11	-1.543	0.123
TA stiffness (N/m)	405.65 ± 74.17	403.59 ± 63.66	-0.109	0.913	326.72 ± 76.54	319.49 ± 73.19	-2.395	0.017
TA elasticity (log)	1.2 ± 0.38	1.18 ± 0.38	-0.754	0.451	1.06 ± 0.23	1.03 ± 0.22	-2.333	0.020
GL tone (Hz)	16.18 ± 1.54	16.34 ± 1.78	-1.556	0.12	14.53 ± 1.55	14.58 ± 1.64	-0.941	0.347
GL stiffness (N/m)	279.38 ± 35.56	290.86 ± 60.82	-3.24	0.001	256.5 ± 36.62	258.58 ± 39.12	-1.001	0.317
GL elasticity (log)	1.24 ± 0.29	1.29 ± 0.29	-2.424	0.015	1.13 ± 0.24	1.13 ± 0.2	-0.208	0.835
GM tone (Hz)	16.22 ± 1.74	16.07 ± 1.59	-1.459	0.144	14.18 ± 1.46	14.03 ± 1.6	-2.172	0.030
GM stiffness (N/m)	269.05 ± 33.85	269.46 ± 34.2	-0.498	0.619	244.11 ± 32.78	239.7 ± 33.78	-2.065	0.039
GM elasticity (log)	1.41 ± 0.3	1.39 ± 0.32	-0.466	0.641	1.24 ± 0.24	1.22 ± 0.26	-1.76	0.078
VL tone (Hz)	16.94 ± 2.01	16.87 ± 2	-0.375	0.707	13.33 ± 1.91	13.25 ± 1.89	-0.817	0.414
VL stiffness (N/m)	298.66 ± 47.58	297.52 ± 46.69	-0.206	0.837	223.29 ± 49.15	220.61 ± 50.59	-1.058	0.290
VL elasticity (log)	1.43 ± 0.32	1.36 ± 0.3	-3.17	0.002	1.08 ± 0.27	1.06 ± 0.29	-1.076	0.282
VMO tone (Hz)	13.86 ± 1.58	14.37 ± 2.5	-3.244	0.001	11.93 ± 1.31	11.83 ± 1.36	-1.506	0.132
VMO stiffness (N/m)	225.89 ± 37.6	229.59 ± 44.4	-1.407	0.159	180.2 ± 28.42	182.9 ± 32.49	-0.851	0.395
VMO elasticity (log)	1.26 ± 0.29	1.34 ± 0.37	-2.756	0.006	1.07 ± 0.35	1.12 ± 0.34	-2.647	0.008
RF tone (Hz)	14.83 ± 1.38	14.91 ± 1.4	-0.892	0.372	13.42 ± 1.85	13.65 ± 1.81	-2.765	0.006
RF stiffness (N/m)	252.6 ± 32.62	255.62 ± 43	-0.547	0.584	214.93 ± 39.94	217.37 ± 40.14	-1.276	0.202
RF elasticity (log)	1.28 ± 0.29	1.25 ± 0.26	-1.268	0.205	1.04 ± 0.28	1.02 ± 0.26	-1.551	0.121

TA – tibialis anterior, GL – gastrocnemius lateralis, GM – gastrocnemius medialis, VL – vastus lateralis, VMO – vastus medialis obliquus, RF – rectus femoris

Bold denotes statistically significant values.

ral at rest. Non-neural aspects involve no neural activation and the muscle tone comprises passive stiffness and viscoelastic properties. Muscle tone is not irreversibly dependent on age but can be modulated by direct muscle contraction, physical exercise, or vigorous sports activities. Athletes have a larger cross-sectional area of muscle than sedentary people [32]. Greater muscle tone as observed in the TA, GM, and RF muscles in all participants could be associated with a smaller cross-sectional area or whether these muscles were relaxed in resting position or not [33]. However, we did not directly measure the cross-sectional area of the muscles or used EMG to predict muscular activation in resting position. Elasticity is an indicator of the ability of muscles to return to their original state after contraction or when directly compressed. In the present study, right TA, VL, RF, and left VMO were found to have lower muscle elasticity (the logarithmic decrement was higher). The myotonometric parameters depend on the tissue beneath the measuring point. If measured on the paraspinal muscle medially or laterally, the latter point shows substantially lower values owing to fascia and muscle tissue organization [34]. Significant differences in regional muscles have also been observed [35]. The myofascial fat tissue could be an important factor for our subjects who were sedentary middle-aged adults and the subcutaneous fat tissue was not prominent. Individual differences were also determined in different studies previously [22, 35]. In the present study, intramuscular components were not evaluated at the physiological level. It might be of value to compare the mechanical properties of the muscles in this aspect in future studies.

Clinicians often assess the contralateral side to determine abnormalities of the injured side. This approach is useful in conditions that cause muscle atrophy or imbalance. Accurate assessment and management of musculoskeletal problems are central to rehabilitation and monitoring treatment efficacy. Muscles of the dominant leg (which is the right leg for the majority) are usually found to be stronger than those of the other leg in the general population. In this study, the right leg was dominant in 91% of the participants and the left leg in 9%. It should not be assumed that the mechanical properties of a muscle will affect the muscle strength in the same way as leg dominance will affect the muscle strength. Comparing the right and left extremities of all individuals revealed that the magnitude of difference between both extremities was less than approximately 5%. According to Gapaeva and Vain [36], up to 5% of the difference between the extremities can be accepted as normal; however, further follow-up is recommended if the difference is 5–10%, and for the differences above 10%, the test should be repeated. Our results showed no significant difference in all mechanical parameters between the dominant and non-dominant legs. Three studies investigated differences in leg dominance using the Myoton device and revealed no significant difference between the dominant and non-dominant side [15, 22, 37]. The result of these studies support our findings.

Comparing the mechanical properties of the muscles by sex showed that bilateral muscle tone, stiffness, and elasticity were greater in males than in females. Stiffness, tone (non-neural form), and elasticity are dependent on the muscle structure (length and cross-sectional area) and the intrinsic

Table 7. Comparison of the mechanical properties of lower extremity muscles depending on age

Parameter	Age groups			χ^2	p
	Group 1 (n = 235) (mean ± SD)	Group 2 (n = 85) (mean ± SD)	Group 3 (n = 69) (mean ± SD)		
Right TA tone (Hz)	18.91 ± 3.35 ^a	17.46 ± 3.32	18.43 ± 3.33	12.224	0.002
Right TA stiffness (N/m)	376.11 ± 85.25 ^a	342.88 ± 89.24	355.94 ± 72.68	12.789	0.002
Right TA elasticity (log)	1.11 ± 0.31	1.07 ± 0.3	1.25 ± 0.35 ^{b,c}	15.941	0.001
Left TA tone (Hz)	18.53 ± 3.3 ^a	17.4 ± 3.29	18.45 ± 3.29	7.398	0.025
Left TA stiffness (N/m)	368.75 ± 79.25 ^a	341.96 ± 89.17	355.62 ± 69.63	7.572	0.023
Left TA elasticity (log)	1.1 ± 0.32	1.03 ± 0.25	1.22 ± 0.35 ^{b,c}	14.755	0.001
Right GL tone (Hz)	15.43 ± 1.69	15.11 ± 1.95	15.29 ± 1.68	2.541	0.281
Right GL stiffness (N/m)	266.57 ± 38.37	265.2 ± 38.86	274.51 ± 34.31	3.960	0.138
Right GL elasticity (log)	1.15 ± 0.26	1.17 ± 0.27	1.31 ± 0.26 ^{b,c}	20.309	0.001
Left GL tone (Hz)	15.46 ± 1.88	15.4 ± 2.08	15.45 ± 1.87	0.229	0.892
Left GL stiffness (N/m)	272.37 ± 43.6	269.08 ± 43.03	287.58 ± 84.69	3.602	0.165
Left GL elasticity (log)	1.18 ± 0.27	1.2 ± 0.22	1.32 ± 0.25 ^{b,c}	19.284	0.001
Right GM tone (Hz)	15.24 ± 1.88	14.97 ± 1.86	15.21 ± 2	1.424	0.491
Right GM stiffness (N/m)	253.94 ± 36.02	257.06 ± 35.93	263.35 ± 32.75 ^c	6.299	0.043
Right GM elasticity (log)	1.29 ± 0.29	1.31 ± 0.22	1.45 ± 0.32 ^{b,c}	14.080	0.001
Left GM tone (Hz)	15.02 ± 1.91	15 ± 2	15.07 ± 1.7	0.331	0.847
Left GM stiffness (N/m)	252.2 ± 39.05	255.95 ± 36.99	259.04 ± 29.34	3.408	0.182
Left GM elasticity (log)	1.28 ± 0.3	1.28 ± 0.27	1.42 ± 0.33 ^{b,c}	10.617	0.005
Right VL tone (Hz)	15.34 ± 2.6 ^a	14.53 ± 2.72	14.98 ± 2.74	7.093	0.029
Right VL stiffness (N/m)	260.73 ± 59.18	255.53 ± 62.93	263.59 ± 66.79	1.020	0.601
Right VL elasticity (log)	1.23 ± 0.34	1.22 ± 0.29	1.33 ± 0.41	2.926	0.231
Left VL tone (Hz)	15.21 ± 2.54 ^a	14.37 ± 2.66	15.15 ± 2.93	8.415	0.015
Left VL stiffness (N/m)	260.64 ± 59.96	247.55 ± 61.92	262.86 ± 68.39	5.922	0.052
Left VL elasticity (log)	1.2 ± 0.33	1.17 ± 0.28	1.3 ± 0.37	4.464	0.107
Right VMO tone (Hz)	13.07 ± 1.72 ^a	12.5 ± 1.7	12.68 ± 1.77	10.064	0.007
Right VMO stiffness (N/m)	200.28 ± 42.83	199.49 ± 35.81	213.84 ± 34.81 ^{b,c}	8.705	0.013
Right VMO elasticity (log)	1.11 ± 0.29	1.16 ± 0.33	1.35 ± 0.41 ^{b,c}	17.333	0.001
Left VMO tone (Hz)	13.28 ± 2.45 ^a	12.59 ± 2.41	12.96 ± 1.95	9.355	0.009
Left VMO stiffness (N/m)	203.02 ± 45.3	205.32 ± 48.34	215.35 ± 40.04	5.601	0.061
Left VMO elasticity (log)	1.17 ± 0.34 ^a	1.28 ± 0.44	1.35 ± 0.37 ^b	14.741	0.001
Right RF tone (Hz)	14.19 ± 1.63	13.8 ± 1.95	14.23 ± 2.01	4.266	0.118
Right RF stiffness (N/m)	230.87 ± 41.17	232.93 ± 43.94	242.2 ± 36.24	2.770	0.250
Right RF elasticity (log)	1.11 ± 0.28	1.17 ± 0.28	1.3 ± 0.37 ^c	14.855	0.001
Left RF tone (Hz)	14.38 ± 1.65 ^a	13.82 ± 1.78	14.42 ± 1.94 ^c	8.954	0.011
Left RF stiffness (N/m)	236.47 ± 49.13	226.62 ± 39.63	246.25 ± 38.2 ^c	7.768	0.021
Left RF elasticity (log)	1.1 ± 0.27	1.13 ± 0.26	1.24 ± 0.32 ^{b,c}	10.311	0.006

TA – tibialis anterior, GL – gastrocnemius lateralis, GM – gastrocnemius medialis, VL – vastus lateralis, VMO – vastus medialis obliquus, RF – rectus femoris

Significant difference after Kruskal-Wallis test results and post-hoc pairwise (Dunn correction) comparisons output of Kruskal-Wallis test. Bold denotes statistically significant values.

^a p < 0.05, group 1 vs. group 2

^b p < 0.05, group 1 vs. group 3

^c p < 0.05, group 2 vs. group 3

Table 8. Correlations between age and leg muscle mechanical properties

	Right			Left		
	TA tone (Hz)	TA stiffness (N/m)	TA elasticity (log)	TA tone (Hz)	TA stiffness (N/m)	TA elasticity (log)
<i>r</i>	-0.103*	-0.125*	0.088	-0.074	-0.115*	0.053
	GL tone (Hz)	GL stiffness (N/m)	GL elasticity (log)	GL tone (Hz)	GL stiffness (N/m)	GL elasticity (log)
<i>r</i>	-0.081	0.060	0.229**	-0.015	0.044	0.194**
	GM tone (Hz)	GM stiffness (N/m)	GM elasticity (log)	GM tone (Hz)	GM stiffness (N/m)	GM elasticity (log)
<i>r</i>	-0.036	0.084	0.157**	-0.031	0.061	0.133**
	VL tone (Hz)	VL stiffness (N/m)	VL elasticity (log)	VL tone (Hz)	VL stiffness (N/m)	VL elasticity (log)
<i>r</i>	-0.123*	-0.033	0.037	-0.090	-0.077	0.039
	VMO tone (Hz)	VMO stiffness (N/m)	VMO elasticity (log)	VMO tone (Hz)	VMO stiffness (N/m)	VMO elasticity (log)
<i>r</i>	-0.154**	0.064	0.169**	-0.110*	0.079	0.208**
	RF tone (Hz)	RF stiffness (N/m)	RF elasticity (log)	RF tone (Hz)	RF stiffness (N/m)	RF elasticity (log)
<i>r</i>	-0.034	0.070	0.160**	-0.051	0.023	0.140**

r – Spearman rank correlation coefficient, TA – tibialis anterior, GL – gastrocnemius lateralis, GM – gastrocnemius medialis, VL – vastus lateralis, VMO – vastus medialis obliquus, RF – rectus femoris

* significant difference at the level of 0.05, ** significant difference at the level of 0.01

composition of the muscle [38, 39]. The reasons for higher mechanical properties might be greater strength and muscle mass, lower fat content, different muscle fibre characteristics and cross-sectional area in males [40–45]. This could be related to the fact that females have a higher total body fat and lower muscle mass than males [46, 47], but we did not investigate anthropometric characteristics; however, males and females had comparable BMI. Differences in the muscle mass, increased myofascial content, and altered body fat distribution cannot be explained solely by the BMI [48]. Previous studies, using different measurement devices, indicated that BMI was an additional factor that did not affect the biomechanical muscle parameters [27, 49]. On the other hand, one study suggested that the variance in the stiffness (7.5%) and elasticity (4%) of the upper trapezius muscle as assessed by MyotonPRO could be attributed to BMI. In this regard, the use of a basic BMI calculated from height and body weight may not be an appropriate method for myotonometric assessments. Agyapong-Badu et al. [6] implied that there was an inverse relationship between muscle tone and subcutaneous adipose tissue in sedentary participants; a thicker subcutaneous fat layer can alter muscle response, decrease oscillations and frequencies, and therefore reduce muscle tone. Thus, other body composition measures, such as subcutaneous fat thickness, lean body mass, and fat mass index, could be investigated in future studies to explain changes in muscle mechanical properties.

Males and females showed diverse viscoelastic characteristics in different muscles and legs. Intragroup differences in males and females might have resulted from differences in activities of daily living, occupations [50], gait patterns [51], neuromuscular strategies [52], and physical activity levels [30] of the participants. The activities of daily living and job tasks are associated with repeated movements and static loading for maintaining posture. These activities include both static and dynamic movements that can cause a variety of musculoskeletal alignments or neuromuscular strategies. Also, the working position depends partly on the requirements of the surrounding factors and partly on other requirements, such as using force, personal fitness level, command of specific movement skills, as well as the ability to relax between

work operations. Two studies with Myoton-3 found that muscle stiffness, tone, and elasticity might change depending on occupational variation in healthy individuals [50, 53].

Comparing mechanical properties between the age groups revealed that bilateral muscle tone and stiffness of TA, muscle tone of VMO and VL, and left RF tone were greater in the younger groups. We believe that some factors, such as physical activity, activities of daily living, or soft tissue infiltration, might explain these results. Myofascial tissue can change dynamically in response to external and internal loads [54]. Kim et al. [55] reported that greater muscle tone indicated increased exercise load or pain. Hence, muscle tone can be modulated by physical activity. We suggest that the bilaterally greater level of muscle tone in the younger group might be related to their level of physical activity.

The elasticity of the bilateral TA, GM, GL, VMO, and RF muscles decreased with advancing age. This finding is in accordance with the results of a previous study, in which mechanical properties of the upper trapezius and sternocleidomastoid muscles were examined in relation to the sitting or lying position, and it was reported that aging decreased only the flexibility of upper trapezius [49]. Aging has more prominent effects (increased stiffness and decreased elasticity) on the mechanical properties of the lower extremity muscles than those of the upper extremity [56]. Ultrasound imaging has indicated that the non-contractile components of the VL muscle, which is a part of the quadriceps, are more numerous than those of the supraspinatus muscle [57]. Along with aging, the increase in fibrous components and the decrease in size and number of muscle fibres lead to a decrease in the contractile elements [58]. In a study by Kent-Braun et al. [59], older adults were shown to have smaller contractile areas compared with young adults. In our study, it is quite likely that the prominent difference in elasticity can be a result of the increase in non-contractile components. In a study comparing athletes and sedentary individuals, the fact that the former presented lower elasticity than the latter was attributed to the effect of physical activity on muscle flexibility during rest [30]. We suggest that lower levels of elasticity in the elder group than in other groups might be explained by their decreased level of physical activity.

Our results showed that age had a positive poor correlation with TA stiffness and VMO tone bilaterally, and a negative poor correlation with GL, GM, VMO, and RF elasticity bilaterally. Previous studies demonstrated higher stiffness and lower elasticity in lower and upper extremity muscles and face muscles in older individuals vs. younger people [6, 30, 49, 60]. Therefore, the muscles that were investigated in these studies were differently affected by age, which is consistent with our results. Gervasi et al. [30] tested two muscles from the dominant side (rectus femoris and biceps brachii) and found that age had a slight effect on the tone of females' biceps brachii but moderate effect on the stiffness and elasticity of both muscles. A poor correlation was demonstrated for stiffness and elasticity of both muscles in their male group, but not for the tone [30]. Agyapong-Badu et al. [6] observed that the muscle tone and stiffness were positively and elasticity was negatively correlated with age in the case of biceps brachii, with the same results for rectus femoris except for the age correlation with the tone. Kocur et al. [49] demonstrated that the stiffness and elasticity of the neck muscles were positively correlated with age, which accounted for their findings in the sternocleidomastoid muscle (stiffness: 28.4%, elasticity: 53%) and the upper trapezius muscle (stiffness: 22%, elasticity: 13%).

The major finding of our study was that age constituted the major correlate of the elasticity of specific lower extremity muscles, TA stiffness, and VMO tone. The higher stiffness and tone and lower elasticity in humans have previously been demonstrated to increase the risk for soft tissue and bone injuries [61–63]. From a practical viewpoint, increased TA stiffness and VMO tone can potentially lead to increased risk of falls because these muscles are important for gait and daily activities. Also, decreased muscle elasticity and higher stiffness may adversely affect the quality of muscle contractions and postural control [64, 65], impair blood circulation, and cause premature fatigue [34, 66]. Further studies are warranted to compare myotonic measurements with laboratory-based objective methods in order to postulate an accurate interpretation of findings regarding muscle physiology.

Limitations

Our study provides information that may be important for assessing the mechanical properties of the musculoskeletal system. A number of limitations have hindered our ability to draw definitive conclusions. First, we used only a classic BMI calculated from height and weight, and did not evaluate the adipose tissue of the participants. The subjects may have had a different body composition, which can affect the interpretation of our results. The second limitation is that the present study did not include older age groups (50–65 or over 65 years). If these groups had been involved, we could have better investigated the progressive effects of aging and additional data could have been presented for the reduced muscle elasticity and increased muscle stiffness and tone. The findings of the present study may provide an opportunity to establish normative values for the mechanical properties of different muscles in various populations and to form the basis for further research.

Conclusions

The measured mechanical properties of the lower extremity muscles were bilaterally higher in males than females in all parameters. These mechanical properties showed different characteristics in different muscles for both sexes. Leg dominance did not affect the mechanical properties of the muscles,

which were found comparable between the two sides. The elasticity of GL, GM, VMO, RF decreased, and TA stiffness and VMO tone increased with advancing age in both legs. We believe that increased muscle stiffness and reduced muscle elasticity with advancing age may adversely affect mobility, cause premature early fatigue, or impair blood circulation. The study presents data on altered mechanical properties of leg muscles in relation to sex, leg dominance, and age. These findings can provide important information for future studies involving different populations or disorders.

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Conflict of interest

The authors state no conflict of interest.

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