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Acoustic Myography in Assessment of Isokinetic and Isometric Muscle Strength in a Healthy Danish Population

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ABSTRACT

Isokinetic and isometric force measurements involving a dynamometer setup are widely used in training and in muscle assessment. For further understanding of the method, we investigated the activation of key functional muscles during isokinetic and isometric movements. During force measurements in an isokinetic Biodex System 3 Pro™, acoustic myography (AMG) was applied. Ten healthy subjects (5 men/5 women) in each decade from 20 to 69 years of age participated in the study. Measurements were carried out during extension and flexion of the ankle, knee and elbow joints. Muscle fibre use was measured by efficiency (E-score) and fibre recruitment (temporal (T-score) and spatial (S-score) summation). AMG measurements showed good reproducibility, and the recruitment pattern of muscle fibres did not change with gender or age. Overall, a significantly higher E-score ($P < 0.05$) was found at the lower angular velocities than at the higher ones, indicating a lower level of muscle efficiency at higher velocities. Muscles used for knee movement exhibited higher scores than muscles associated with the ankle and elbow joints, most likely related to the greater degree of force production at this joint compared to the ankle and elbow. The ability to activate and inactivate muscle fibres during periods of isokinetic activity becomes increasingly more difficult as the velocity increases. When assessing training effects in sports or rehabilitation, AMG in parallel with isokinetic measurements adds important additional information by giving a measure of possible improvements in efficiency and fibre use.

1. INTRODUCTION

Isokinetic exercise is performed at a fixed angular velocity and with a constant resistance, using a specially designed rig [1-4]. What is measured using such a rig is work (Nm = J) under fixed and controlled conditions. As a technique, it is valuable when monitoring subjects during training, both in terms of sports and in terms of rehabilitation. Furthermore, this type of exercise as a training method also shows improvements in muscle function [4-8]. Recently, a study of healthy subjects, spanning an age range of 20 to 69 years, applied the non-invasive method acoustic myography (AMG) to assess changes in a diverse array of muscles, performing daily activities such as stair climbing, cycling, walking, upper arm flexion/extension etc. [9]. AMG detects activation of motor units during a particular movement for the particular muscles involved in that movement. The method differs from surface electromyography (sEMG), since it measures the pressure waves generated by activated muscles rather than their electrical depolarization, thus avoiding the influence of neuromuscular endplates, which confound the sEMG signal [10]. The data from the healthy AMG study [9] confirmed an earlier pilot study showing that muscle fibre recruitment (temporal and spatial summation) for a given type of force production is similar in men and women [11]. The study showed that the force produced by *m. palmaris longus*, although weaker for women than for men, was regulated identically for both genders and was closely correlated with the recorded AMG signal for this muscle [11]. The method applied in this study had the advantage over similar yet earlier methods, such as those used by Stokes and Blythe [12] and Madeleine [13], in that the sensors had a vastly improved frequency response, thereby providing a true image of muscle fibre use.

In the present study, our aim has been to relate isokinetic and isometric muscle strength in a healthy population to the AMG parameters: Efficiency (E), Spatial (S) and Temporal (T) summation, thereby giving a physiological understanding of the development of the well-defined isokinetic and isometric muscle strength measured in a isokinetic setup at a fixed angle; ultimately showing the advantages and limitations of this type of muscle assessment and type of training.

2. MATERIALS AND METHODS

The present study constitutes part of a larger study on a group of healthy adults [9].

2.1. Ethics

The methods applied were all non-invasive. The study followed the guidelines set by the Helsinki Declaration 2013 (https://www.wma.net/policies-post/wma-declaration-of-helsinki-ethical-principles-for-medical-research-involving-human-subjects/), and the subjects gave an informed written consent prior to participating in the study. The study was approved by the Capital Region of Denmark's Ethics Committee (H-15017787) and was registered at the Danish Data Protection Agency (Datatilsynet). Collected data was handled according to The Act on Processing of Personal Data 2018. Furthermore, the staff carrying out the measurements was fully trained in the use of the involved equipment and setups, and data handling was carried out blinded.

2.2. Subjects

50 healthy subjects participated. The number of subjects was decided based on a Power calculation on previously published data [11, 14], revealing a power of approximately 80% for this number of subjects in each age group. The age group was 20 - 69 years, with 5 men and 5 women in each age decade. Subjects were recruited via advertising and by subjects having heard about the project and approaching the institute themselves with a wish to participate. All measurements took place at the Parker Institute, Copenhagen University Hospital, Bispebjerg and Frederiksberg Hospital, Denmark.

Inclusion criteria were: 20 - 69 years of age, 18.5 < Body Mass Index (BMI) < 30, healthy according to a physical examination and a standard blood test used for assessment of patients with musculoskeletal diseases at Copenhagen University Hospital, Bispebjerg and Frederiksberg, prior to measurements, reporting of no chronic or no present illness, no intake of medicine except birth control pills, pain reported in the normal range, and showing the pain pattern for healthy subjects from answering the PainDETECT Questionnaire (PD-Q) ©2005Pfizer Pharma GmbH [15]. The participants refrained from consuming caffeine containing drinks 2 hours prior to measurements and alcoholic drinks 12 hours prior to measurements.

Exclusion criteria were: Not fulfilling inclusion criteria or having no understanding of the Danish language.

2.3. Isokinetic/Isometric Measurements

Isokinetic muscle strength and isometric muscle strength at a fixed angle was measured using a Biodex System 3 Pro™ dynamometer (Biodex Medical Systems Inc., Shirley, NY, USA). The dominant leg/arm was measured according to the protocol described in full by Danneskiold-Samsøe and colleagues [4].

The following measurements were carried out:

1) Ankle: Dorsi/plantar flexion. ROM between +30˚ and −10˚. The isokinetic measurements were performed at 15, 30 and 45˚ s^{-1}, and the isometric tests at 20˚ and 0˚ dorsi flexion. For AMG: *m. gastrocnemius* and *m. tibialis anterior* were measured.

2) Knee: Range of Motion (ROM) between 100˚ and 0˚ of flexion. The isokinetic measurements were performed at 30, 60 and 90˚ s^{-1}, and the isometric tests were performed at 65˚ and 30˚ knee flexion. For AMG: *m. vastus lateralis* and *m. rectus femoris* were measured.

3) Elbow: Flexion/extension. ROM between 0˚ and 180˚. The isokinetic measurements were performed at 30, 60 and 90˚ s^{-1}, and the isometric tests at the 60˚ and 120˚ flexed position. For AMG: *m. biceps* and *m. triceps* were measured.

2.4. Acoustic Myography (AMG) Measurements

AMG measurements were carried out with a CURO unit, using 20 mm CURO sensors (CURO-Diagnostics ApS, Bagsværd, Denmark). The frequency recording range of the sensors was 0.5 - 20 ±0.5 kHz. The data were stored as WAV files on the CURO unit until further processing. During measurements, real-time recordings were followed on an iPad Air (Apple Inc, Cupertino, CA, USA) to ensure that recordings were successfully obtained.

The sensors were coated with ultrasound gel and placed on the skin over the body of the muscle concerned and held in place using a soft self-adhesive bandage (Danamull haft LF, Mediplast AB, Malmö, Sweden). The measured AMG parameters are Efficiency (E), Spatial (S) and Temporal (T) summation [9].

Recordings have shown that using the 15 dB setting on the AMG sensor cable it is possible to measure AMG signals from independently active muscles to a depth of 20 cm from the skin surface: stimulation of the hamstrings with 1 - 10 Hz, 200 µsec, 40 mA pulses using an InTENSity 12 stimulator (Roscoe Medical, Ohio, USA) and recording from the quadriceps muscle of a human subject using a 50 mm sensor connected to a CURO and iPad (Source: CURO-Diagnostics ApS, Bagsværd, DK).

2.5. Data Handling

The data recorded on the CURO unit was transferred to the the CURO Clinic data handling software (CURO-Diagnostics ApS, Bagsværd, Denmark) to calculate the AMG parameters; E- (Efficiency); S- (Spatial); and T- (Temporal) scores [10]. The raw data were processed using T-max = 250, S-max = 0.99 and Threshold = 0.07. The three scores were expressed on a scale from 0 - 10, where 10 expresses no activity, and 0 expresses maximal activity. For details of the calculation used see a recently published article involving AMG measurements on cerebral palsy subjects [14]. A combined mean ST score, an average of the S- and T-scores, was also used to assess muscle activation. The E-score expresses how long a muscle actively switched on out of the total measuring time, the S-score the recruitment of motor units, and the T-score the motor unit firing rate.

Muscle Strength: The average peak torque was chosen for each muscles and movements at each joint.

The data were obtained directly from the Biodex System 3 Pro™ dynamometer (Biodex Medical Systems Inc., Shirley, NY, USA).

2.6. Statistical Analysis

Linear Mixed Model was applied via R software (version 3.4.3, 2017) with add-on package lme4 (r-project.org) to perform a linear mixed relation between AMG parameters with the angular velocities/angle of movement. Angular velocities were used as fixed effect, and intercepts for subjects were used as random effect in the model.

P-values were obtained by likelihood ratio tests of the full model with the interaction of age with angular velocities/angle of movement against the model without the interaction of age with angular velocities/angle of movement. P values > 0.05 were considered non-significant (NS). Data of five age groups were pooled together as P-values were found to be non-significant (P value > 0.05). Multiple comparisons were carried out using the *multcomp* package of R, and adjusted P-values were obtained for each angular velocity. Values are presented as the mean ± the standard deviation (SD) of the men.

3. RESULTS

All participating subjects fulfilled the set criteria for being healthy, and the measured values for isokinetic and isometric muscle strength (see **Appendix 1**) were found to be in the range of previously published data for healthy subjects [4].

Although muscle strength varies with age and gender, it has been shown that the recruitment of muscle fibres in healthy individuals does not change with gender or age, despite of a decrease in motor units with age [9, 11]. Our present data are in line with this, and we have therefore in the following been able to pool the data from the whole study population.

3.1. AMG Data

Table 1 shows the AMG data recorded from the isokinetic and isometric measurements.

3.2. Ankle Isokinetic

The E, S, and T-score (mean ± SD) for dorsiflexion and plantarflexion of the ankle for *m. gastrocnemius* and *m. tibialis anterior* for the angular velocities of 15˚ s^{-1}, 30˚ s^{-1}, 45˚ s^{-1} is presented in **Table 1(a)**.

The E-score for *m. gastrocnemius* for dorsiflexion and plantarflexion was significantly higher (P < 0.05) at 15˚ s^{-1} of angular velocity when compared to 30˚ s^{-1} and 45˚ s^{-1}. There was no significant difference (P > 0.05) between the S and T-score of *m. gastrocnemius* at the three different angular velocities in dorsiflexion and plantarflexion.

The E-score for *m. tibialis anterior* for dorsiflexion and plantarflexion was significantly different (P < 0.05) between 15˚ s^{-1}, 30˚ s^{-1} and 45˚ s^{-1}. There was no significant difference (P > 0.05) between the S-score of *m. tibialis anterior* at the three different angular velocities in dorsiflexion, but in plantarflexion the S-score at 15˚ s^{-1} was significantly different (P < 0.05) from the score at 45˚ s^{-1}. There was no significant difference (P > 0.05) between the T-score of *m. tibialis anterior* at the three different angular velocities measured during dorsiflexion and plantarflexion.

3.3. Ankle Isometric

The E, S, and T-score (mean ± SD) for dorsiflexion and plantarflexion of the ankle for the two muscle groups recorded with the CURO unit at the angles of 0˚ and 15˚ is presented in **Table 1(a)**.

The E-score for *m. gastrocnemius* for dorsiflexion and plantarflexion was significantly higher (P < 0.05) at 0˚ as compared to 15˚. There was no significant difference (P > 0.05) in the S and T-score of *m. gastrocnemius* at the two angles in dorsiflexion and plantarflexion.

The E-score for *m. tibialis anterior* for dorsiflexion and plantarflexion was significantly different (P <

0.05) between 0˚ and 15˚. There was no significant difference ($P > 0.05$) in the S and T-score of *m. tibialis anterior* at the two angles in dorsiflexion and plantarflexion.

Table 1. E-, S-, T- and S-T-scores for isokinetic and isometric measurements of the ankle (a), knee (b) and elbow (c), respectively, of the subjects in this study.

(a)

Ankle						
Ankle dorsi flexion and plantar flexion (isokinetic)						
	Dorsi flexion (up)			Plantar flexion (down)		
	15˚ s^{-1}	30˚ s^{-1}	45˚ s^{-1}	15˚ s^{-1}	30˚ s^{-1}	45˚ s^{-1}
m.gastrocnemius						
E score	8.2 ± 1.9[b]	6.6 ± 2.6[a]	6.0 ± 2.8[a]	7.2 ± 2.3[b]	5.6 ± 2.6[a]	5.0 ± 2.4[a]
S score	9.4 ± 0.5	9.5 ± 0.4	9.4 ± 0.2	9.5 ± 0.4	9.3 ± 0.6	9.3 ± 0.3
T score	4.4 ± 2.8	4.7 ± 2.8	5.1 ± 3.0	2.8 ± 2.4	2.8 ± 2.5	2.4 ± 2.1
ST score	6.9 ± 1.5	7.1 ± 1.4	7.3 ± 1.5	6.1 ± 1.2	6.0 ± 1.2	5.9 ± 1.2
m.tibialis anterior						
E score	7.4 ± 1.9[c]	5.5 ± 2.2[b]	4.6 ± 2.1[a]	7.6 ± 1.9[c]	5.8 ± 2.4[b]	4.4 ± 2.2[a]
S score	9.5 ± 0.4	9.4 ± 0.2	9.3 ± 0.4	9.5 ± 0.1[b]	9.3 ± 0.3[ab]	9.2 ± 0.5[a]
T score	3.2 ± 2.8	3.3 ± 2.5	2.5 ± 2.1	4.1 ± 2.1	4.4 ± 2.3	3.6 ± 2.8
ST score	6.3 ± 1.4	6.4 ± 1.2	5.9 ± 1.2	6.8 ± 1.0	6.9 ± 1.1	6.4 ± 1.4
Ankle dorsi flexion and plantar flexion (isometric)						
	Dorsi flexion (up)			Plantar flexion (down)		
	0˚	15˚		0˚	15˚	
m.gastrocnemius						
E score	8.8 ± 1.3[b]	8.2 ± 1.8[a]		7.0 ± 2.3[a]	7.5 ± 2.4[b]	
S score	9.6 ± 0.2	9.6 ± 0.2		9.5 ± 0.2	9.5 ± 0.3	
T score	4.1 ± 2.5	3.8 ± 2.9		3.4 ± 2.0	3.7 ± 2.1	
ST score	6.9 ± 1.3	6.8 ± 1.5		6.5 ± 1.0	6.8 ± 1.0	
m.tibialis anterior						
E score	7.9 ± 1.6[b]	7.3 ± 1.6[a]		8.0 ± 1.8[a]	8.6 ± 1.2[b]	
S score	9.5 ± 0.1	9.6 ± 0.1		9.5 ± 0.2	9.4 ± 0.7	
T score	3.2 ± 1.7	2.9 ± 1.9		4.0 ± 2.2	4.3 ± 2.1	
ST score	6.5 ± 0.8	6.3 ± 0.9		6.8 ± 1.1	6.8 ± 1.2	

(b)

Knee						
	Knee extension and flexion (isokinetic)					
	Extension			Flexion		
	30° s^{-1}	60° s^{-1}	90° s^{-1}	30° s^{-1}	60° s^{-1}	90° s^{-1}
m. rectus femoris						
E score	6.1 ± 2.5[b]	5.5 ± 2.8[b]	4.6 ± 2.6[a]	7.0 ± 2.7[c]	5.4 ± 2.6[b]	4.6 ± 2.5[a]
S score	9.5 ± 0.3[b]	9.4 ± 0.3[ab]	9.2 ± 0.8[a]	9.5 ± 0.3[b]	9.3 ± 0.3[b]	9.1 ± 0.8[a]
T score	6.1 ± 1.7[b]	5.4 ± 1.5[a]	5.5 ± 1.5[ab]	7.0 ± 1.4	6.9 ± 1.3	6.7 ± 1.8
ST score	8.0 ± 0.6	7.9 ± 0.9	7.8 ± 1.0	8.2 ± 0.6	7.8 ± 0.8	7.9 ± 0.7
m. vastus lateralis						
E score	6.2 ± 2.8[b]	5.4 ± 2.5[b]	4.4 ± 2.7[a]	7.1 ± 2.8[c]	5.6 ± 2.6[b]	4.6 ± 3.0[a]
S score	9.4 ± 0.3[b]	9.4 ± 0.2[b]	9.2 ± 0.5[a]	9.4 ± 0.3[b]	9.1 ± 0.8[a]	9.0 ± 0.8[a]
T score	6.5 ± 1.3	6.3 ± 1.7	6.4 ± 1.9	6.9 ± 1.3	6.5 ± 1.7	6.8 ± 1.4
ST score	7.8 ± 0.8	7.4 ± 0.8	7.4 ± 0.9	8.3 ± 0.7	8.1 ± 0.6	7.9 ± 1.0
	Knee extension (isometric)					
		Extension				
		30°	60°			
m. rectus femoris						
E score		7.7 ± 2.1[b]	6.7 ± 2.9[a]			
S score		9.5 ± 0.3	9.5 ± 0.2			
T score		7.3 ± 1.0	7.1 ± 0.8			
ST score		8.2 ± 0.6	8.2 ± 0.4			
m. vastus lateralis						
E score		7.1 ± 2.7[b]	6.4 ± 3.1[a]			
S score		9.4 ± 0.9	9.4 ± 0.5			
T score		7.0 ± 1.0	6.9 ± 1.0			
ST score		8.4 ± 0.5	8.3 ± 0.4			

(c)

Elbow						
	Elbow extension and flexion (isokinetic)					
	Extension			Flexion		
	30° s^{-1}	60° s^{-1}	90° s^{-1}	30° s^{-1}	60° s^{-1}	90° s^{-1}
m. biceps brachii						
E score	4.7 ± 3.0^{b}	2.8 ± 2.6^{ab}	2.6 ± 2.0^{a}	4.4 ± 2.8^{c}	3.4 ± 2.9^{b}	2.6 ± 1.9^{a}
S score	8.8 ± 1.0^{b}	8.3 ± 1.3^{b}	7.5 ± 1.8^{a}	9.0 ± 0.8^{b}	8.0 ± 1.6^{a}	7.7 ± 1.9^{a}
T score	4.6 ± 3.0	5.3 ± 3.3	5.7 ± 3.0	3.0 ± 2.7	2.8 ± 2.6	2.6 ± 2.0
ST score	6.7 ± 1.5	6.8 ± 1.5	6.6 ± 1.5	6.0 ± 1.5	5.4 ± 1.8	5.2 ± 1.9
m. triceps brachii						
E score	4.6 ± 3.0^{b}	3.6 ± 2.9^{a}	2.9 ± 2.8^{a}	5.0 ± 2.7^{b}	3.9 ± 2.9^{a}	3.8 ± 2.8^{a}
S score	9.1 ± 0.7^{c}	8.4 ± 1.6^{b}	7.7 ± 2.0^{a}	9.3 ± 0.4^{b}	8.8 ± 1.1^{a}	8.5 ± 1.4^{a}
T score	2.5 ± 2.1	2.5 ± 2.4	2.5 ± 2.4	4.7 ± 2.6	4.6 ± 3.3	4.2 ± 3.2
ST score	5.8 ± 1.0	5.5 ± 1.3	5.1 ± 1.6	7.0 ± 1.3	6.7 ± 1.6	6.3 ± 1.6
	Elbow extension and flexion (isometric)					
	Extension			Flexion		
	60°			60°		
m. biceps brachii						
E score	5.9 ± 2.7			6.1 ± 3.0		
S score	9.4 ± 0.2			9.3 ± 0.3		
T score	3.1 ± 1.9			4.1 ± 2.1		
ST score	6.2 ± 0.9			6.7 ± 1.0		
m. triceps brachii						
E score	5.2 ± 3.0			5.8 ± 3.0		
S score	9.3 ± 0.6			9.4 ± 0.3		
T score	3.2 ± 1.9			3.7 ± 2.1		
ST score	6.2 ± 0.8			6.5 ± 1.1		

Data presented are the mean ± SD of the mean. a, b values within a row lacking common superscripts differ significantly from one another ($P < 0.05$).

3.4. Knee Isokinetic

The E, S, and T-score (mean ± SD) for extension and flexion of the knee for the two muscle groups recorded with the CURO unit for the angular velocities of 30˚ s^{-1}, 60˚ s^{-1}, 90˚ s^{-1} is presented in Table 1(b).

The E-score for *m. rectus femoris* in extension was significantly higher ($P < 0.05$) at 30˚ s^{-1} and 60˚ s^{-1} of angular velocity as compared to 90˚ s^{-1} but the E-score for *m. rectus femoris* for flexion was significantly different ($P < 0.05$) between 30˚ s^{-1}, 60˚ s^{-1} and 90˚ s^{-1}.

The S-score for *m. rectus femoris* in extension was significantly higher ($P < 0.05$) at 30˚ s^{-1} as compared to 90˚ s^{-1} but remained unchanged ($P > 0.05$) at 60˚ s^{-1}. The S-score for *m. rectus femoris* in flexion was significantly higher ($P < 0.05$) at 30˚ s^{-1} and 60˚ s^{-1} of angular velocity as compared to 90˚ s^{-1}.

The T-score for *m. rectus femoris* in extension was significantly higher ($P < 0.05$) at 30˚ s^{-1} as compared to 60˚ s^{-1} and 90˚ s^{-1}. But for flexion the T-score of m. *rectus femoris* remained unchanged between the three angular velocities.

The E-score for *m. vastus lateralis* in extension was significantly higher ($P < 0.05$) at 30˚ s^{-1} and 60˚ s^{-1} compared to 90˚ s^{-1} but for flexion the E-score was statistically different ($P < 0.05$) between the angular velocities.

The S-score for *m. vastus lateralis* in extension was significantly higher ($P < 0.05$) at 30˚ s^{-1} and 60˚ s^{-1} compared to 90˚ s^{-1} and for flexion the S-score was significantly different ($P < 0.05$) between the angular velocities.

There was no significant difference ($P > 0.05$) in the T-score of *m. vastus lateralis* at the three different angular velocities either in extension or in flexion.

3.5. Knee Isometric

The E, S, and T-score (mean ± SD) for extension and flexion of the knee for the two muscle groups recorded with the CURO unit at the angles of 30˚ and 60˚ is presented in Table 1(b).

The E-score for *m. rectus femoris* for extension and flexion was significantly higher ($P < 0.05$) at 30˚ as compared to 60˚. There was no significant difference ($P > 0.05$) between the S and T-score of *m. rectus femoris at* 30˚ and 60˚ of extension or flexion.

The E-score for *m. vastus lateralis* for extension and flexion was significantly higher ($P < 0.05$) at 30˚ as compared to 60˚. There was no significant difference ($P > 0.05$) in the S and T-score of *m. vastus lateralis* at the two angles of extension or flexion.

3.6. Elbow Isokinetic

Mean with Standard deviation of E, S, T-score for extension and flexion of elbow for two muscle groups recorded with CURO unit for AMG for the angular velocities of 30˚ s^{-1}, 60˚ s^{-1}, 90˚ s^{-1} is presented in Table 1(c).

The E-score for *m. biceps brachii* in extension was significantly higher ($P < 0.05$) at 30˚ s^{-1} of angular velocity as compared to 90˚ s^{-1} but remained unchanged ($P > 0.05$) in 60˚ s^{-1}. The E-score for *m. biceps brachii* in flexion was significantly different ($P < 0.05$) between 30˚ s^{-1}, 60˚ s^{-1} and 90˚ s^{-1}. The S-score for *m. biceps brachii* in extension was significantly different ($P < 0.05$) at 30˚ s^{-1} and 60˚ s^{-1} of angular velocity as compared to 90˚ s^{-1}. However, for flexion the S-score for *m. biceps brachii* was significantly different ($P < 0.05$) at 30˚ s^{-1} of angular velocity as compared to 60˚ s^{-1} and 90˚ s^{-1}. No statistically significant difference ($P > 0.05$) was observed between 60˚ s^{-1} and 90˚ s^{-1}. There was no significant difference ($P > 0.05$) in the T-score of *m. biceps brachii* at the three different angular velocities of extension or flexion.

The E-score for *m. triceps brachii* in extension and flexion was significantly higher ($P < 0.05$) at 30˚ s^{-1} of angular velocity as compared to 60˚ s^{-1} and 90˚ s^{-1}. The S-score for *m. triceps brachii* during extension was significantly different ($P < 0.05$) between 30˚ s^{-1} , 60˚ s^{-1} and 90˚ s^{-1} but in flexion the S-score was higher for 30˚ s^{-1} as compared to 60˚ s^{-1} and 90˚ s^{-1}. There was no significant difference ($P > 0.05$) in the

T-score of *m. triceps brachii* at the three different angular velocities of extension or flexion.

3.7. Elbow Isometric

The E, S, and T-score (mean ± SD) for extension and flexion of the elbow for the two muscle groups recorded with the CURO unit at the angle of 60˚ is presented in **Table 1(c)**.

3.8. ST-Scores

Since a given muscle strength may be maintained either by activating the same fibres at a given frequency or by activating more fibres at the same time, the ST-score will in many situations give a better picture of muscle activation [11]. The ST-scores have been calculated for all movements and angles and are given in **Table 1**.

4. DISCUSSION

In order to better understand what isokinetic muscle strength represents, one can look upon it as being a defined piece of work, carried out by muscles associated with a defined movement, and in so doing, defining a true to life movement in a controlled way [1-5]. An understanding of the underlying muscle fibres used is therefore of great value when following an athlete or a patient over time, thereby giving a better understanding of what the method can achieve and what may be adjusted in any given training programme.

In this study, which has focused on adult healthy subjects, the measured peak torques were found to be in accordance with what has been measured in an earlier study of healthy adults [4].

In terms of the AMG scores, measured during isokinetic movement, there appears to be an overall finding of a higher E-score at the lower angular velocities than at the higher ones. Since the E-score represents the period of active muscle contraction during any given measuring time frame, such a finding may be interpreted as indicating a lower level of muscle efficiency at higher velocities. This finding can be observed even when the S-score follows the same trend in some instances, or even when it remains unchanged in others. Likewise, this is true even though the T-score remains unchanged. One interpretation of these data must surely be that relatively more fibres appear to be needed at the same time to generate the work required at the higher velocities, but this does not necessitate a higher firing frequency (temporal summation). That is to say; in order to work against a given load, a set number of fibres are necessary, but such work can be achieved without having to change the firing rate of the active fibres. Merely a longer duration of muscle activity is required. To ascertain that this finding was not influenced in some way separately by the two means by which muscle fibres generate and maintain force (spatial or temporal summation) [11], the average of these two parameters (T and S) was calculated, the ST-score, see **Table 1**. A different way of applying temporal and spatial summation was seen at the knee joint compared to the other joints (see below).

Returning to the observed decrease in the E-score with increasing velocity, it would appear to mean that switching muscles fibres on and off during periods of isokinetic activity becomes increasingly more difficult as the velocity increases. This finding could therefore prove to be a useful measure when assessing the benefits of patient rehabilitation training, since it has been shown that training over time gives rise to an improvement in muscle efficiency (higher E-score) [16].

When looking at the AMG scores at isometric force production at different angles, or at extension or flexion, only small differences are found, as would be expected, since the force production takes place with a fixed position of the joint.

If we focus on the different muscle groups, it appears that those muscles used for knee movement exhibit higher scores, even at the same angular velocity, than the other measured muscles. This is perhaps not so surprising as we are dealing with much larger muscles at and around the knee than for the other joints and, consequently, creating a greater degree of force production. Not only do the thigh muscles generally have an overall greater cross-sectional area than the other muscles measured in this study, they also have fibres with a greater cross-sectional area, giving them more force per fibre. A consequence of this is that fewer fibres are needed to generate a given level of force. This seems to be supported to some degree by our data, as the S-score for the knee muscles is rather high (low amplitude signal), although it is not that much lower for the muscles attached to the elbow and ankle. What is clearly different though is the T-score, which is between 2 - 4 for both the ankle and elbow, and up around 5 - 7 for the knee. The resulting higher ST score for the knee muscles is therefore mostly due to the higher T-score in the muscles attached to this joint, and a higher T-score means a lower firing frequency. Whilst this difference is most likely associated with the larger muscle fibres, one cannot completely ignore the possibility that it might also be due in part to a difference in innervation. Such a pattern of fibre use in large and powerful muscles may help prevent or at the very least reduce the risk of damage to tendons and their attachments, both in terms of the muscle-tendon junction (MTJ) and the osteo/tendinosus junction (OTJ) interfaces. If this assumption is indeed right, then the regulation of fibre stimulation in the knee may well be controlled by stretch receptors and proprioceptors in and around joints such as the knee [17, 18]. Indeed, such a notion is supported by the consistency of the acoustic myography scores at the various speeds, and by both the isokinetic and isometric force production.

When considering limitations, this study has only looked at healthy subjects and has not made any comparison with injured or diseased subjects, which raises the limitation of comparison between disease-oriented *versus* healthy control-oriented findings. Furthermore, since this is the first study to examine acoustic myography measurements with isokinetic force production there are no published data with which to compare our findings giving rise to issues concerning external validity. Finally, there are those who might also consider the limitation of clinical importance *versus* statistical significance when assessing the findings of this study. Future studies may now wish to address these and other issues associated with the AMG signal of isokinetic measurements and follow for example the effects of rehabilitation.

5. CONCLUSION

In healthy subjects, the ability to activate and inactivate muscle fibres during periods of isokinetic activity becomes increasingly less efficient with increasing angular velocity, independent of age or gender. With this reference AMG data set for isokinetic and isometric measurements over three important joints, AMG in parallel with isokinetic measurements becomes a useful and easily applicable method in the clinic when assessing individuals with musculoskeletal problems or when training athletes. With the application of AMG as a new addition to measurements of isokinetic muscle strength, this training/rehabilitation method will be able to show possible increase in efficiency, as well as changes in spatial and temporal summation over time, together with changes in peak torque. The combined AMG and isokinetic data set will give a full picture of improvements in activation patterns, as well as in muscle strength development, thereby giving a more complete physiological description of effect of the applied isokinetic training, and a possibility of addressing what has to be improved to get optimal effect of the training/rehabilitation.

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CREDIT AUTHORSHIP CONTRIBUTION STATEMENT

Else Marie Bartels and Waqas Ahmed share joint first authors on this manuscript. Else Marie Bartels:

Design of study, Data curation, Writing—original draft. Jack Kvistgaard Olsen: Design, Data curation. Eva Littrup Andersen: Design, Data curation. Bente Danneskiold-Samsøe: Design, Henning Bliddal: Design, Data curation. Lars Erik Kristensen: Design, Data curation. Cecilie Rødgaard Bartholdy: Design, Data curation. Waqas Ahmed: Data curation, Writing. Adrian P. Harrison: Data curation, Writing—original draft.

All authors read and commented on the final manuscript.

DATA AVAILABILITY STATEMENT

All data is reported in the paper.

CONFLICTS OF INTEREST

AH is in the process of establishing a company to produce and market the Acoustic MyoGraphy equipment (CURO-Diagnostics ApS), and at no time was he involved in analysing the data. BDS's spouse is a member of the Executive Board of CURO-Diagnostics ApS and has only a very limited number of shares in the company. BDS has no direct economic or personal interests in CURO-Diagnostics ApS, nor was she involved in the data analysis.

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APPENDIX 1

Isokinetic and isometric peak torque values.

(a)

Ankle						
Ankle dorsi flexion and plantar flexion in female subjects (isokinetic)						
	Dorsi flexion (Nm)			Plantar flexion (Nm)		
Analysis set	15° s^{-1}	30° s^{-1}	45° s^{-1}	15° s^{-1}	30° s^{-1}	45° s^{-1}
20 - 29						
Median	28.7	27.5	27.4	38.1	35.6	34.1
[Min; Max]	[17.8; 32.5]	[18.4; 30.9]	[3.1; 49.9]	[16.1; 62.6]	[4; 51]	[21.5; 52.3]
30 - 39						
Median	28.0	25.0	19.3	34.0	18.2	7.7
[Min; Max]	[23; 62.9]	[9.1; 40.8]	[8.6; 28.6]	[8.7; 50.1]	[4.3; 28.2]	[0.5; 27.1]
40 - 49						
Median	24.2	17.6	17.2	28.9	11.9	15.7
[Min; Max]	[19.6; 37.6]	[14.8; 29.5]	[11.1; 30.5]	[22; 83.9]	[4.7; 35.3]	[2.4; 36.7]
50 - 59						
Median	23.1	16.8	13.0	21.9	8.8	2.4
[Min; Max]	[19.9; 78.4]	[15; 64.1]	[6.7; 50.8]	[5.5; 60.1]	[2.4; 64.1]	[1.1; 61]
60 - 69						
Median	26.2	19.5	18.4	18.8	36.6	15.0
[Min; Max]	[12; 33.9]	[6.6; 51.5]	[17.5; 25.7]	[7.7; 43.6]	[1.3; 39.4]	[0.2; 38.2]
Ankle dorsi flexion and plantar flexion in male subjects (isokinetic)						
	Dorsi flexion (Nm)			Plantar flexion (Nm)		
Analysis set	15° s^{-1}	30° s^{-1}	45° s^{-1}	15° s^{-1}	30° s^{-1}	45° s^{-1}
20 - 29						
Median	41.2	35.5	31.2	55.5	43.4	26.1
[Min; Max]	[30.8; 48]	[17.2; 38.6]	[12.7; 33.9]	[15.6; 124.4]	[2.5; 107.2]	[2.6; 95.9]
30 - 39						
Median	37.3	30.5	31.5	91.4	70.6	32.0
[Min; Max]	[27.8; 41.6]	[14.9; 39]	[9.2; 34.7]	[7.7; 144.2]	[1.9; 127.1]	[2.4; 108.1]
40 - 49						

Continued

Median	38.0	30.0	21.9	38.3	27.3	3.7
[Min; Max]	[31.8; 45]	[8.6; 41]	[8; 24.8]	[4.9; 96.1]	[3.7; 84.6]	[0.2; 79.5]
50 - 59						
Median	27.8	24.7	16.5	40.1	26.6	15.0
[Min; Max]	[17.7; 35.3]	[19.8; 30.3]	[10.7; 23.9]	[11.9; 74.9]	[10; 97.3]	0.4; 27.4]
60 - 69						
Median	26.9	24.2	22.1	42.1	39.1	31.0
[Min; Max]	[24.3; 29.3]	[23.1; 27.5]	[18.7; 22.2]	[2.6; 50.8]	[1.7; 72.2]	[2.6; 54.7]
Ankle dorsi flexion and plantar flexion in female subjects (isometric)						
	Dorsi flexion (N)			Plantar flexion (N)		
Analysis set	0°	15°		0°	15°	
20 - 29						
Median	21.4	21.8		70.4	48.6	
[Min; Max]	[15.5; 46]	[18.1; 43]		[16.5; 87.4]	[32.8; 64.2]	
30 - 39						
Median	23.6	24.9		70.4	44.5	
[Min; Max]	[17.5; 33.9]	[17.5; 31.5]		[24.7; 91]	[31.5; 54.1]	
40 - 49						
Median	23.6	24.4		82.8	54.6	
[Min; Max]	[21.4; 37.5]	[22.1; 29.1]		[38.6; 97.8]	[14.5; 73.9]	
50 - 59						
Median	26.0	23.4		67.3	36.1	
[Min; Max]	[22.3; 30.4]	[19.4; 28.8]		[42.3; 78.8]	[34.2; 59.2]	
60 - 69						
Median	26.2	21.4		41.8	33.1	
[Min; Max]	[7.7; 31]	[14.1; 36]		[25.3; 93.5]	[27.9; 58.9]	
Ankle dorsi flexion and plantar flexion in male subjects (isometric)						
	Dorsi flexion (N)			Plantar flexion (N)		
Analysis set	0°	15°		0°	15°	
20 - 29						
Median	42.8	35.9		98.4	66.1	
[Min; Max]	[31.1; 19]	[26.5; 52.1]		[38.5; 178.2]	[37.7; 111.4]	

Continued

30 - 39						
Median	29.7	35.0		158.4	106.8	
[Min; Max]	[21.9; 47.3]	[20.2; 40.4]		[90.3; 234.3]	[41.3; 162.7]	
40 - 49						
Median	45.7	42.5		89.5	61.3	
[Min; Max]	[28.8; 59.9]	[23.7; 49]		[11.2; 158.6]	[33.5; 89.5]	
50 - 59						
Median	34.0	33.4		92.5	75.1	
[Min; Max]	[27.6; 125.7]	[29.3; 46]		[23.7; 161.8]	[27.6; 125.7]	
60 - 69						
Median	26.8	26.6		67.4	54.7	
[Min; Max]	[26.2; 33.2]	[24.4; 33.3]		[4.9; 94.7]	[26.4; 64.3]	

(b)

Knee						
Knee extension and flexion in female subjects (isokinetic)						
	Extension (Nm)			Flexion (Nm)		
Analysis set	30° s^{-1}	60° s^{-1}	90° s^{-1}	30° s^{-1}	60° s^{-1}	90° s^{-1}
20 - 29						
Median	102.5	90.1	72.7	82.3	68.9	65.6
[Min; Max]	[8.6; 116]	[5; 106.4]	[13.8; 97]	[25.2; 103.6]	[13.9; 94.8]	[24.1; 99]
30 - 39						
Median	58.2	21.9	11.2	59.3	34.9	28.4
[Min; Max]	[15.1; 122]	[10.5; 104.7]	[4.1; 84.6]	[33.4; 79.1]	[27; 76.8]	[20.2; 65]
40 - 49						
Median	61.4	69.2	50.0	67.9	64.6	57.0
[Min; Max]	[35.9; 131.6]	[21.8; 111.6]	[9.4; 75.9]	[33.8; 1.101.8]	[32.1; 83]	[18.8; 84.6]
50 - 59						
Median	30.3	41.0	36.9	39.5	39.5	40.3
[Min; Max]	[15.5; 88.5]	[5.6; 84.5]	[8.8; 64.8]	[25.8; 73.7]	[18.2; 74.6]	[18.8; 61.8]
60 - 69						
Median	65.9	56.5	30.4	60.2	51.3	41.3

Continued

[Min; Max]	[55.4; 75]	[8.7; 70.1]	[6; 61.8]	[29.8; 63.4]	[20.4; 67.7]	[20.4; 66.1]
Knee extension and flexion in male subjects (isokinetic)						
	Extension (Nm)			Flexion (Nm)		
Analysis set	30˚ s^{-1}	60˚ s^{-1}	90˚ s^{-1}	30˚ s^{-1}	60˚ s^{-1}	90˚ s^{-1}
20 - 29						
Median	116.1	26.9	8.9	88.4	55.1	39.0
[Min; Max]	[55.4; 259]	[9.3; 206.4]	[4.6; 159.3]	[44.4; 169.5]	[34.7; 151]	[27.4; 137.1]
30 - 39						
Median	77.8	27.1	26.6	126.8	58.9	48.7
[Min; Max]	[15.8; 266.2]	[2.1; 200.8]	[2.1; 189.2]	[42.5; 191.1]	[34.7; 122.3]	[26.3; 119.2]
40 - 49						
Median	119.4	20.5	16.3	101.1	41.5	43.0
[Min; Max]	[17.2; 191]	[6.9; 178]	[3.9; 161]	[35.5;]	[21.2; 137]	[18.5; 114.6]
50 - 59						
Median	102.1	92.2	69.5	96.5	106.5	86.8
[Min; Max]	[56.6; 157.8]	[42.5; 134.7]	[20; 112.1]	[61; 157.7]	[55; 146.9]	[38; 133.9]
60 - 69						
Median	66.6	108.5	91.6	56.3	76.3	75.7
[Min; Max]	[31.8; 146.6]	[14.6; 130]	[15.2; 103.3]	[32.4; 105]	[27.6; 102.8]	[28.2; 94.7]
Knee extension in female subjects (isometric)						
	Extension (N)			Extension (N)		
Analysis set	30˚			60˚		
20 - 29						
Median	50.6			122.3		
[Min; Max]	[28.9; 56.6]			[96; 157.4]		
30 - 39						
Median	53.1			106.5		
[Min; Max]	[25.5; 74.7]			[96; 145.3]		
40 - 49						
Median	53.4			122.3		
[Min; Max]	[23.2; 63.7]			[84.4; 130.2]		

Continued

50 - 59		
Median	38.8	109.1
[Min; Max]	[18.8; 54.6]	[86.9; 118.5]
60 - 69		
Median	50.2	104.4
[Min; Max]	[38.6; 59.3]	[85.5; 129.4]
Knee extension in male subjects (isometric)		
	Extension (N)	Extension (N)
Analysis set	30˚	60˚
20 - 29		
Median	161.0	178.2
[Min; Max]	[66.8; 190.1]	[97.1; 320.1]
30 - 39		
Median	141.4	190.5
[Min; Max]	[64.3; 262.8]	[114; 339.2]
40 - 49		
Median	136.2	253.4
[Min; Max]	[5.9; 146.7]	[19.5; 289]
50 - 59		
Median	70.6	156.7
[Min; Max]	[39.1; 101.6]	[112.6; 180.2]
60 - 69		
Median	68.4	132.2
[Min; Max]	[35.1; 79.4]	[79; 164.8]

(c)

Elbow						
Elbow extension and flexion in female subjects (isokinetic)						
	Extension (Nm)			Flexion (Nm)		
Analysis set	30˚ s^{-1}	60˚ s^{-1}	90˚ s^{-1}	30˚ s^{-1}	60˚ s^{-1}	90˚ s^{-1}
20 - 29						
Median	23.1	18.0	25.2	40.6	39.3	38.1
[Min; Max]	[4; 34.8]	[19.2; 33.5]	[3.1; 32.5]	[23.6; 53.7]	[16.1; 41.6]	[14.8; 41.1]
30 - 39						
Median	21.5	11.0	3.4	35.9	28.5	22.5
[Min; Max]	[4; 29]	[0.1; 54.9]	[0; 24.1]	[23.2; 40.1]	[14.2; 42.2]	[16.7; 38.4]

Continued

40 - 49						
Median	27.9	22.4	18.7	44.2	33.9	29.4
[Min; Max]	[12.5; 39]	[4.1; 33.2]	[7.7; 37.1]	[29.7; 59.3]	[17.4; 53.8]	[21.5; 52.1]
50 - 59						
Median	21.3	7.9	2.8	34.2	19.1	21.6
[Min; Max]	[6.7; 30.5]	[1.1; 14.1]	[0.3; 19.6]	[28.5; 37.7]	[18.2; 25]	[9.5; 27.6]
60 - 69						
Median	12.3	5.3	5.2	37.3	24.5	22.5
[Min; Max]	[4.9; 24.1]	[3.1; 26.1]	[1.7; 28.9]	[32; 81.5]	[15.1; 37.9]	[16.5; 36.5]
Elbow extension and flexion in male subjects (isokinetic)						
	Extension (Nm)			Flexion (Nm)		
Analysis set	30˚ s^{-1}	60˚ s^{-1}	90˚ s^{-1}	30˚ s^{-1}	60˚ s^{-1}	90˚ s^{-1}
20 - 29						
Median	41.1	20.9	7.0	72.4	45.6	24.5
[Min; Max]	[6.3; 63.7]	[4.6; 82.9]	[1; 59.8]	[56.8; 101.2]	[35.3; 101.6]	[20.2; 85.6]
30 - 39						
Median	34.7	8.3	0.8	75.8	49.7	18.7
[Min; Max]	[7.6; 66.2]	[30.3; 109.5]	[0; 58.5]	[30.3; 109.5]	[16.2; 111.7]	[16.8; 95.3]
40 - 49						
Median	9.2	5.3	6.2	38.4	26.7	18.3
[Min; Max]	[3; 82.5]	[2.3; 71.1]	[0; 55.5]	[25.5; 90.6]	[16; 107.6]	[18; 80.8]
50 - 59						
Median	30.1	30.5	26.3	52.5	48.4	49.3
[Min; Max]	[17.2; 44.3]	[16; 35.6]	[21; 33.5]	[37.2; 59.4]	[39.9; 82.7]	[41.1; 69.5]
60 - 69						
Median	38.1	28.5	22.2	56.9	52.9	45.1
[Min; Max]	[9.7; 50.4]	[5.4; 46.1]	[3.9; 45.3]	[30; 94.9]	[17.6; 97.3]	[17.1; 80.1]
Elbow extension and flexion in female subjects (isometric)						
		Extension (N)			Flexion (N)	
Analysis set		60˚			60˚	
20 - 29						
Median		36.1			44.5	
[Min; Max]		[24.3; 45.5]			[29.8; 49.9]	

Continued

30 - 39		
Median	40.5	47.0
[Min; Max]	[32.1; 43.1]	[38.5; 54.9]
40 - 49		
Median	44.9	51.1
[Min; Max]	[34.4; 65.3]	[27.8; 60.3]
50 - 59		
Median	35.4	46.3
[Min; Max]	[34.2; 42.8]	[26.9050;]
60 - 69		
Median	41.6	43.8
[Min; Max]	[34.5; 46]	[29.2; 55.6]
Elbow extension and flexion in male subjects (isometric)		
	Extension (N)	Flexion (N)
Analysis set	60˚	60˚
20 - 29		
Median	77.2	79.3
[Min; Max]	[28.7; 90.2]	[63.5; 123.3]
30 - 39		
Median	82.9	95.5
[Min; Max]	[66.8; 92.1]	[87; 111.9]
40 - 49		
Median	67.6	88.4
[Min; Max]	[40; 96.3]	[42; 112.2]
50 - 59		
Median	72.9	81.5
[Min; Max]	[60.7; 82.4]	[63.9; 90.4]
60 - 69		
Median	53.8	56.3
[Min; Max]	[37; 63.8]	[27; 80.5]