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RESEARCH ARTICLE

# The slack test does not assess maximal shortening velocity of muscle fascicles in humans

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## ABSTRACT

The application of a series of extremely high accelerative motor-driven quick releases while muscles contract isometrically (i.e. slack test) has been proposed to assess unloaded velocity in human muscle. This study aimed to measure gastrocnemius medialis fascicle shortening velocity ($V_F$) and tendinous tissue shortening velocity during motor-driven quick releases performed at various activation levels to assess the applicability of the slack test in humans. Gastrocnemius medialis peak $V_F$ and joint velocity recorded from 25 participants using high frame rate ultrasound during quick releases (at activation levels from 0% to 60% of maximal voluntary isometric torque) and during fast contractions without external load (ballistic condition) were compared. Unloaded joint velocity calculated using the slack test method increased whereas $V_F$ decreased with muscle activation level ($P \leq 0.03$). Passive and low-level quick releases elicited higher $V_F$ values ($\geq 41.8 \pm 10.7$ cm s$^{-1}$) compared with the ballistic condition ($36.3 \pm 8.7$ cm s$^{-1}$), while quick releases applied at 60% of maximal voluntary isometric torque produced the lowest $V_F$. These findings suggest that initial fascicle length, complex fascicle–tendon interactions, unloading reflex and motor-driven movement pattern strongly influence and limit the shortening velocity achieved during the slack test. Furthermore, $V_F$ elicited by quick releases is likely to reflect substantial contributions of passive processes. Therefore, the slack test is not appropriate to assess maximal muscle shortening velocity *in vivo*.

**KEY WORDS: Unloaded velocity, Muscle–tendon interaction, High frame rate ultrasound, Muscle activation**

## INTRODUCTION

The relationship between shortening velocity and force-generating capacity is a fundamental property of muscle fibres (Hill, 1938). Force produced by the muscle and its associated shortening velocity can define crucial estimates of muscle performance, such as the maximal force and power (Bottinelli et al., 1996; Edman, 2010; Lieber and Ward, 2011). However, assessment of the maximal shortening velocity a muscle can reach is more challenging *in vivo* (Hill, 1970). Hill (1970) proposed calculating maximal shortening velocity by extrapolation of the force–velocity relationship, which accounts for the maximal shortening velocity of the whole muscle. As an alternative to the assessment of maximal shortening velocity, Edman (1979) introduced a technique that involves a series of quick releases applied to an isolated muscle fibre following a steady-state isometric contraction over various amplitudes. Referred to as the slack test, this technique consists of measuring the time needed for the muscle fibre to take up the slack. The slope of the relationship between the amplitude of the release movement and the time to take up the slack provides a measure of the maximum unloaded shortening velocity independently of the level of muscle activation.

In humans, the torque–velocity relationship is widely used to explore the characteristics of the neuromuscular system and to determine performance capacities in a sport context (Cormie et al., 2011; Reeves and Narici, 2003). By analogy with isolated muscle experiments, the torque–angular velocity relationship has been used to assess the maximal joint velocity during voluntary activation by extrapolation of the velocity reached at zero torque (Hauraix et al., 2015). However, this global estimate includes a composite contribution of several synergistic muscles and depends on various biomechanical features such as moment arm (Lee and Piazza, 2009), muscle gearing (Azizi et al., 2008; Randhawa et al., 2013) or elastic properties of tendinous tissues (Hauraix et al., 2013, 2015). In their recent study, Hauraix et al. (2015) demonstrated that the maximal velocity of the gastrocnemius medialis (GM) fascicles can be appraised from high frame rate ultrasound images collected during fast (ballistic) shortening contractions performed without external load. The implementation of this methodology opens the possibility of accessing this fundamental parameter *in situ*. However, the authors reported significant fascicle–tendon interactions, reflective of a strong influence of the tendon on motor performance during fast shortening contractions (Farcy et al., 2014; Hauraix et al., 2013, 2015). As for isolated muscle fibre experiments, it is possible that the slack test procedure would elicit higher joint velocities than those obtained during ballistic contractions (Hauraix et al., 2015).

Sasaki and Ishii (2005, 2010) adapted the slack test to assess maximal velocity achieved by human muscles during voluntary activation. While Edman (1979) showed that the unloaded shortening velocity of single muscle fibres is independent of activation level (twitch versus tetanus), Sasaki and Ishii (2005, 2010) reported that unloaded joint velocity elicited by the slack test tends to increase with activation level *in vivo* up to 487 deg s$^{-1}$ at an activation level of 60% of maximal voluntary contraction (MVC). The increase in unloaded joint velocity can be explained by the higher percentage of fast fibres recruited at higher muscle activation levels, suggesting that contractile properties could in turn be inferred from the slack test adapted for human experiments. However, unloaded joint velocity obtained with this method showed a substantial variability between individuals. Sasaki and Ishii (2005, 2010) suggested that this could

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originate from the time required for the human contractile component to effectively become slack (i.e. loaded period) and the involvement of compliant tendinous tissues. Indeed, the contribution of tendinous tissues increases with activation level *in vivo* (Beaumatin et al., 2017), which may also participate to increase joint velocity. In addition, neural control involved in such unnatural tasks performed at high velocity (i.e. presynaptic inhibition, unloading and stretch reflexes) may prevent muscle shortening velocity from reaching its full potential (Chow and Darling, 1999; Duclay et al., 2014; Johansson et al., 2014). Overall, these observations question the applicability of the slack test in humans. In this context, the exploration of the role of muscle–tendon interactions and neural control involved in the successive motor-driven quick releases is paramount to better understanding the reasons for the aforementioned limitations related to the slack test when applied *in situ*.

This study aimed to partition muscle–tendon contributions to joint velocity elicited by motor-driven quick releases in order to assess the applicability of the slack test method *in vivo* in humans. For this purpose, we collected high frame rate ultrasound images to measure GM muscle fascicle shortening velocity ($V_F$) during quick releases applied at various activation levels. These velocities were compared with those obtained during ballistic contraction. Electromyographic activity was consistently monitored to determine the magnitude of activation of the plantar flexor muscles throughout the slack test. We hypothesized that (i) high joint velocity achieved during motor-driven quick releases *in vivo* mainly results from the shortening of tendinous tissues, especially at a high level of activations and, in turn, (ii) the slack test procedure does not allow assessment of the true maximal shortening velocity of human muscle fascicle.

## MATERIALS AND METHODS

### Participants

Seventeen males and eight females (age: 26.0±2.7 years, height: 176±8 cm and body mass: 72.1±12.1 kg) with no history of ankle disorder or injury took part in this study. This sample size was chosen to ensure adequate power (0.80) for the main outcomes.

### Ethical approval

All volunteers were informed of the nature, aims and risks associated with the experimental procedure before they gave their written consent to participate. This study was approved by the ethics committee Ouest IV and conformed to the standards of the Declaration of Helsinki.

### Experimental design

All participants attended two familiarization sessions, and a test session several days later. After a 10 min standardized warm-up, participants performed randomized maximal ballistic shortening contractions and several sets of motor-driven quick releases at different levels of isometric torque and amplitude. Mechanical parameters (i.e. torque, displacement, velocity) were recorded with a dedicated mechatronic ergometer. The behaviour of the GM fascicle was recorded with an ultrafast ultrasound during each test. A total of nine males and seven females (age: 24±2.3 years, height: 171±6 cm, body mass: 65.1±8.1 kg) among the 25 initial participants attended a second test session dedicated to electromyographic (EMG) recordings.

### Equipment and procedure

#### MVC

Peak torque was assessed during plantar flexion MVC performed in the isometric condition at 15 deg of dorsiflexion (0 deg=foot perpendicular to the tibia). Of the three trials, the one with the highest isometric peak torque was considered for further analysis.

#### Passive torque

The passive plantar flexor torque–angle relationship was assessed at 1 deg $s^{-1}$.

#### Slack test

A series of quick releases were performed on a specifically designed mechatronic ergometer (Eraclès-Technology, Compiègne, France; Fig. 1). Participants were placed in the prone position and secured by a harness attached to the ergometer. The right ankle rotation axis was adjusted to the motor axis. The starting position of the ankle was consistently set at 15 deg in dorsiflexion. The footplate of the ergometer was connected to a geared motor driven by an electronic controller, encoding a displacement between two joint angles (including all possible positions throughout the amplitude). The slack test consisted of applying a set of extremely fast footplate rotations over different fixed amplitudes while the participant sustained a targeted isometric torque (motor-driven quick releases), with the help of visual feedback. Acceleration was set at 27,400 deg $s^{-2}$ followed by a symmetric deceleration of the footplate. The participant was instructed to maintain the level of exertion until asked to relax. The footplate motion was automatically trigerred 20 ms after the start of ultrasound video using an output signal from the ultrasound device. A motor-driven quick release was 'non-completed' if: (i) target torque was not properly reached and stable; (ii) torque was not completely suppressed during the quick release; and (iii) the participant failed to produce a significant positive torque after the completion of the footplate rotation. A pilot analysis demonstrated a low level of completion when quick releases were performed at 80% of targeted isometric torque (51%). Considering this, we adapted the initial protocol proposed by Sasaki and Ishii (2005) by performing quick releases at various initial levels of isometric contraction (5%, 10%, 20%, 40% and 60% of isometric torque) and amplitude of rapid angle change (25–55 deg with 5 deg intervals). Two trials of quick releases in the passive condition were also achieved at each amplitude. All trials were performed in a randomized order. The positioning of the probe and the proper clamp system prevented an adequate placement of EMG electrodes. Consequently, EMG signals were collected during an additional test session with the same initial levels of isometric contraction at a fixed amplitude which elicited the highest rate of quick release completion (45 deg).

#### Ballistic shortening contractions

Participants performed five trials on a specific ergometer composed of a rotational footplate and a bench (Bio2M, Compiègne, France) (Lambertz et al., 2008), as previously described (Hauraix et al., 2015). They were instructed to displace the footplate in the plantarflexion direction without external load 'as fast as possible' over the entire amplitude (from −15 to 40 deg). When the participants thought they could achieve a faster movement, an additional trial was performed, with no more than 7 trials to avoid fatigue.

### Data collection and processing

#### Mechanics

Joint angle, motor angular velocity and torque were recorded at 4000 Hz sampling frequency using a custom-made analog-to-digital converter. During ballistic contractions, joint velocity was calculated as the mean velocity from −15 to 40 deg of ankle angle. The time to torque re-development following the unloading phase was defined as the interval between the onset of motor angular

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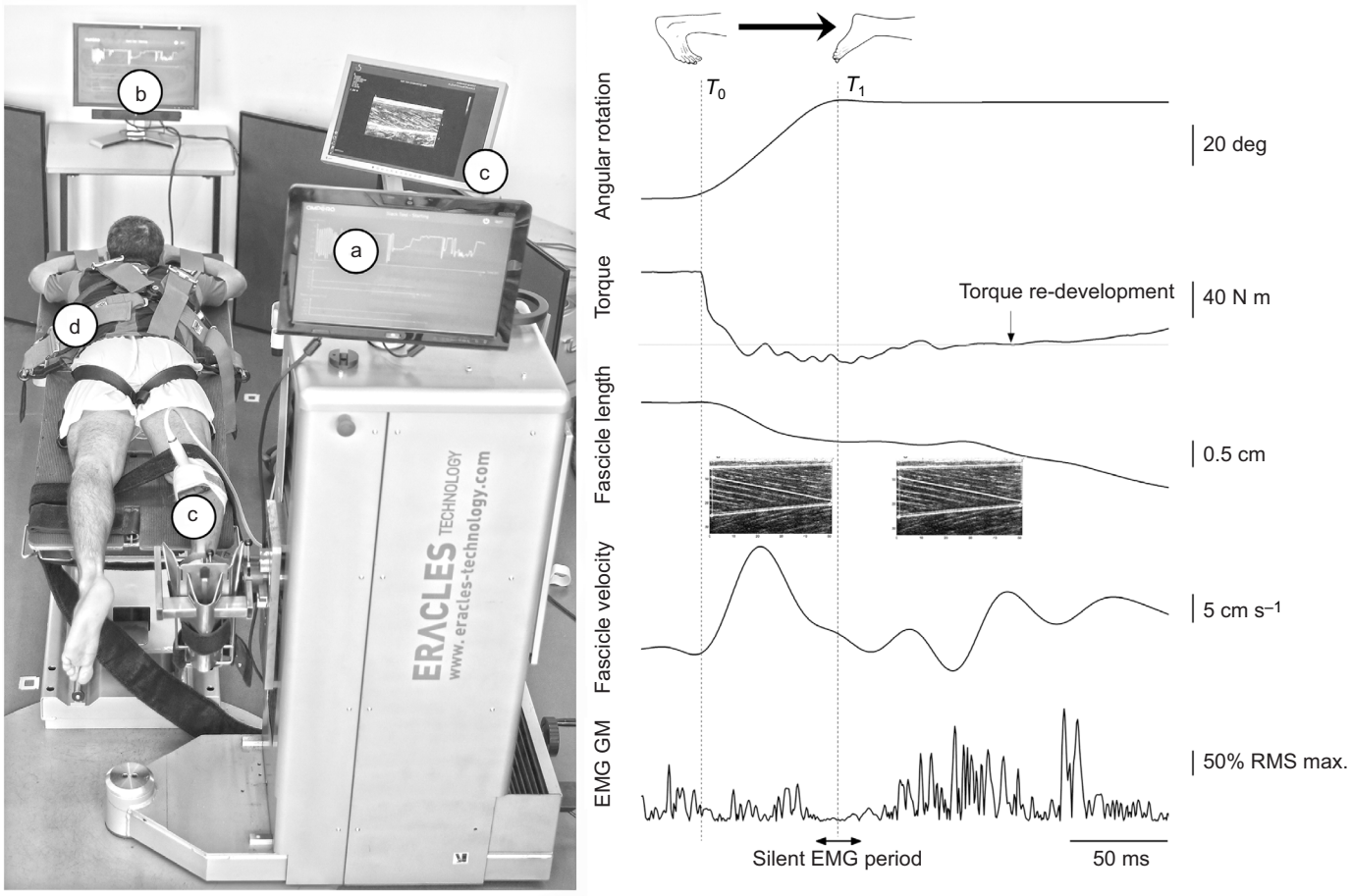



**Fig. 1. Experimental design.** Motor-driven quick releases were performed on a mechatronic ergometer (a). Targeted isometric activation level [20% of maximal voluntary contraction (MVC) in this example] was defined by the investigator and displayed to the participant (b) placed in the prone position and secured by a harness attached to the ergometer (d). Ultrafast ultrasound images were recorded using a high frame rate ultrasound (c), which triggered the quick release 20 ms after the start of ultrasound image acquisition. The time corresponding to the onset of footplate rotation was defined as $T_0$, and the end of footplate rotation was defined as $T_1$. Torque re-development corresponded to a positive torque after the end of footplate rotation. The duration of the silent electromyographic (EMG) period (unloading reflex) at the end of the footplate rotation was defined as the time when EMG activity was below baseline +5 s.d. from the beginning of the footplate rotation. GM, gastrocnemius medialis; RMS, root mean square.

displacement and the first moment corresponding to a positive torque after the stop of the footplate (Fig. 1). Mechanical signals were analysed using custom-written scripts (Origin 9.1, OriginLab Corporation, Northampton, MA, USA). Position was low-pass filtered (80 Hz) using a 2nd order Butterworth filter; torque and velocity were low-pass filtered (150 Hz) using a 3rd order Butterworth filter. The method described by de Zee and Voigt (2001) and used by Sasaki and Ishii (2005) was applied to torque signals to remove the artefacts due to high acceleration and deceleration phases (Fig. S1). Briefly, passive torque measured during the stretching cycle was first subtracted from torque signal measured during quick releases. Then, Fourier transform was calculated from the acceleration measured during quick releases. This Fourier transform was divided by a transfer function $H$ transformed in the time domain and subtracted from the torque signal corrected for passive torque. $H$ was calculated by Fourier transform of the acceleration measured during quick releases in the passive condition divided by the Fourier transform of quick release torque obtained in the passive condition minus passive torque.

### Unloaded joint velocity

According to the original slack test experiment, the unloaded joint velocity is calculated as the slope of the relationship of amplitude and time between release and torque re-development (Edman, 1979). As in the study by Sasaki and Ishii (2005), we applied this procedure in a pilot study performed on 15 participants. Using this method, our results showed: (i) a low test–retest reproducibility of unloaded joint velocity elicited by the slack test (intraclass correlation coefficient, ICC=0.36; standard error of the measurement, s.e.m.=191.5 deg s$^{-1}$; coefficient of variation, CV=66.8%), while the actual velocity measurements performed for each range were reliable; (ii) a high variability and inconsistent unloaded joint velocity between individuals (from 21.7 to 1141.1 deg s$^{-1}$); (iii) the $y$-intercept value, which represents the strain of the series elastic component *in vitro*, was frequently negative as previously reported by Sasaki and Ishii (2005). The problem was probably due to the linear fitting, which can be strongly influenced by very small measurement errors. Therefore, we found that this processing method could not be applied to *in vivo* data. Thus, the maximal joint velocity elicited by the motor-driven quick release was calculated for each activation level from the mean velocity measured for each amplitude.

### Ultrasound

An ultrafast ultrasound scanner (Aixplorer, Supersonic Imagine, Aix en Provence, France) coupled with two, 55 mm linear

transducer arrays (4–15 MHz, SuperLinear 15-4, Vermon, Tours, France) was used to acquire images from GM muscle at a sampling frequency of 2000 Hz. The probe was placed on the skin surface at 30% of the distance between the popliteal fossa area and the centre of the lateral malleolus (Kawakami et al., 1998). The ultrasonic raw data were used to create B-mode images by applying a conventional beam using Matlab software (version 2015a, The MathWorks, Natick, MA, USA). The changes in GM fascicle length and pennation angle were obtained using the automatic tracking method proposed by Cronin et al. (2011) (Fig. 1) and low-pass filtered (50 Hz) using a 3rd order Butterworth filter. Tendinous tissue length was considered to be the difference between muscle–tendon unit length, calculated using the anthropometric model proposed by Grieve et al. (1978), and fascicle horizontal length, calculated as fascicle length multiplied by the cosine of pennation angle. $V_F$ and tendinous tissue shortening velocity ($V_{TT}$) were computed as the first time derivative of fascicle length and tendinous tissue length, respectively. These signals were low-pass filtered (30 Hz) using a 3rd order Butterworth filter. Peak $V_F$ and $V_{TT}$ were calculated for each motor-driven quick release and ballistic contraction. For each activation level, peak $V_F$ and $V_{TT}$ were calculated from the mean value measured at each amplitude.

### Electromyography

Surface EMG activity was recorded with a wireless remote unit (Zerowire, Aurion, Italy), placed on the GM, soleus (SOL) and tibialis anterior (TA). The skin was shaved, gently abraded and cleaned with a solution containing ether, acetone and alcohol to minimize inter-electrode impedance. The bipolar, silver/silver chloride, surface disc electrodes (Blue Sensor N-00-S/25, Medicotest, France) were placed with a centre distance of 2 cm, and longitudinally with respect to the underlying muscle fibre arrangement and located according to the recommendations of the SENIAM (Surface EMG for the Non-Invasive Assessment of Muscles) project (Hermens et al., 2000). EMG signals were pre-amplified (input impedance: 20 MΩ; common mode-rejection ratio: 90 dB; gain: 1000; bandwidth: 10–500 Hz), digitized and sampled at 2000 Hz. All EMG signals were first band-pass filtered (high pass: 30 Hz, 3rd order Butterworth filter, low pass: 450 Hz, 3rd order Butterworth). EMG signals obtained during ballistic and quick releases were analysed as the root mean square (RMS) with a 10 ms moving rectangular window to produce the RMS envelope. EMG signals obtained during MVC were analysed as the RMS with a 500 ms moving rectangular window to produce the RMS envelope. The maximal RMS EMG amplitude was selected as the reference to normalize EMG data. The baseline of the EMG amplitude was quantified over a period during which the muscles were inactive at the joint angle corresponding to torque re-development following the quick release phase (30 deg of plantarflexion), and at the start of the movement in ballistic contractions (15 deg of dorsiflexion). The duration of the silent period in the EMG activity at the end of the footplate rotation (unloading reflex) was considered as the time when EMG activity was under the baseline +5 s.d. from the beginning of the footplate rotation. The end of this period (i.e. the time corresponding to an EMG activity above 5 s.d.) corresponded to the onset of muscle activation after footplate rotation and prior to torque re-development. The post-silent period was defined as the time between the end of the silent period and torque re-development. EMG data were averaged before, during and after the quick release. Each value was calculated over the period of time corresponding to footplate displacement (78 ms).

### Statistical analysis

All statistical analyses were performed with Statistica (StatSoft, Tulsa, OK, USA). The normality of the data was tested using a Shapiro–Wilk's test and all data are expressed as means±s.d. Statistical significance was set at $P<0.05$.

The reliability of unloaded joint velocity between tests was determined using the CV, s.e.m. and ICC.

The potential effect of voluntary activation level (0%, 5%, 10%, 20%, 40% and 60% of isometric torque) and amplitude (35, 40, 45 and 50 deg of amplitude) on unloaded joint velocity, peak $V_F$ and peak $V_{TT}$ was determined by two-way ANOVA with repeated measures. One-way ANOVA with repeated measures were used to test the effect of activation level (0%, 5%, 10%, 20%, 40% and 60% of MVC torque) on initial GM fascicle length and tendinous tissue length. The possible influence of activation level on EMG amplitude throughout the quick release was determined by a two-way ANOVA [activation level×phase (before, during and after the quick release)] applied to EMG activity patterns (GM, SOL, TA). One-way ANOVA with repeated measures were used to test the effect of activation level on the silent EMG period in GM and SOL. One-way ANOVA (condition) were used to compare joint velocity, peak $V_F$, peak $V_{TT}$ and EMG RMS between motor-driven quick releases performed at each activation level (5%, 10%, 20%, 40% and 60% of MVC torque) at 45 deg of amplitude and ballistic condition. When the sphericity assumption was violated (Mauchley's test), a Geisser–Greenhouse correction was used. The mean imputation method was performed on missing values of unloaded joint velocity, $V_F$ and $V_{TT}$. *Post hoc* tests were performed by means of Newman–Keuls procedures for comparison between time points when appropriate.

## RESULTS

### Muscle slack completion

The number of motor-driven quick releases that reached the criteria to be considered as 'completed' (i.e. cancellation of torque, occurrence of torque re-development) per participant for each condition is displayed in Table 1. A total of 761 out of 945 trials were completed. The amplitudes that elicited a high rate of completion (>75%) were considered for statistical analysis, i.e. from 35 to 50 deg. Two participants did not succeed at one activation level and were excluded across all conditions.

### Unloaded joint velocity

Unloaded joint velocity showed a robust reproducibility between tests (ICC=0.74, s.e.m.=19.2 deg $s^{-1}$, CV=7.0%). We observed a main effect of activation level ($P=0.01$, Table 2) and amplitude ($P=0.003$), with no amplitude×activation level interaction ($P=0.2$) on unloaded joint velocity values. *Post hoc* tests revealed that unloaded joint velocity at 5% of MVC was significantly lower than that elicited at 40% and 60% of MVC ($P\leq0.03$).

### Muscle–tendon interactions

Initial tendinous tissue length increased ($P<0001$; Fig. 2B) while initial fascicle length significantly decreased with increasing activation level ($P<0001$; Fig. 2A).

In the early phase of the quick releases, fascicle shortening velocity increased, while joint torque synchronously decreased until the achievement of peak $V_F$ (Fig. 1). For all activation levels, $V_F$ peaked when external torque was cancelled (between 41.6±9.1% and 60.1±7.5% of release amplitude for 0% and 60% of MVC, respectively). The amplitude required to achieve peak $V_F$ (+18.5% of release amplitude from 0% to 60% of activation level; $P<0.001$)

**Table 1. Number of participants who completed the trial under the various motor-driven quick release conditions**

| Isometric torque (%MVC): Amplitude (deg) | 5 | 10 | 20 | 40 | 60 |
|---|---|---|---|---|---|
| 25 | 23 (92%) | 24 (96%) | 21 (84%) | 9 (36%) | 4 (16%) |
| 30 | 22 (88%) | 25 (100%) | 25 (100%) | 13 (52%) | 9 (36%) |
| 35 | 25 (100%) | 24 (96%) | 24 (96%) | 23 (92%) | 20 (80%) |
| 40 | 24 (96%) | 24 (96%) | 25 (100%) | 25 (100%) | 24 (96%) |
| 45 | 24 (96%) | 24 (96%) | 22 (88%) | 20 (80%) | 20 (80%) |
| 50 | 21 (84%) | 23 (90%) | 23 (92%) | 20 (80%) | 19 (76%) |
| 55 | 18 (72%) | 19 (76%) | 18 (72%) | 16 (64%) | 13 (52%) |

Values in parentheses represent the associated percentage of participants who completed the motor-driven quick release experiment under each condition relative to the whole sample of participants.

increased with activation level. After the first half of footplate rotation (above 50% of release amplitude), $V_F$ progressively decreased for all activation levels ($P$<0.001). After the end of the quick releases, we observed a short period with no fascicle shortening (from 99.5% to 134% and 105% to 120% of footplate rotation for 5% and 60% of MVC, respectively) followed by a low increase of fascicle shortening, with a peak achieved around torque re-development (Fig. 1). $V_{TT}$ briefly plateaued, and then achieved its peak value before the end of footplate rotation (Fig. 2).

Peak $V_F$ measured during the quick releases showed a significant effect of activation level ($P$<0.001; Fig. 2C) without any effect of amplitude ($P$=0.7) and activation level×amplitude ($P$=0.3). Peak $V_F$ progressively decreased from 0% to 60% of activation level (from 42.6±9.9 to 31.3±10.2 cm s$^{-1}$; $P$<0.03; Table 2). Peak $V_{TT}$ showed a significant effect of activation level ($P$<0.001; Fig. 2D), amplitude ($P$<0.001) and activation level×amplitude interaction ($P$<0.001). Peak $V_{TT}$ gradually increased from 35 to 50 deg plantar flexion ($P$≤0.003) and from 0% to 60% of activation level (+6.3±4.2 deg cm s$^{-1}$; $P$<0.001; Table 2).

### Muscle activity

The duration of the silent EMG period of GM and SOL muscles significantly decreased with activation level ($P$=0.002; Fig. 3). *Post hoc* tests showed that the silent period measured at 60% activation level was significantly shorter than that at 20%, 10% and 5% of MVC ($P$=0.004). Muscle EMG activity patterns showed significant effects of activation level ($P$<0.001) and phase ($P$<0.04), with no significant effect of activation level×phase ($P$<0.07) for GM, SOL and TA muscles for the comparison between activation levels upon quick releases (Fig. 3). *Post hoc* tests showed that GM, SOL and TA EMG activity was significantly higher after the quick release compared with both initial isometric contraction (before the quick release) and during the quick release ($P$<0.001).

**Table 2. Maximal joint velocity, peak muscle fascicle shortening velocity ($V_F$) and peak tendinous tissue shortening velocity ($V_{TT}$) measured during motor-driven quick releases and ballistic contractions**

| | Unloaded joint velocity (deg s$^{-1}$)* | Peak $V_F$ (cm s$^{-1}$) | Peak $V_{TT}$ (cm s$^{-1}$) |
|---|---|---|---|
| Quick release | | | |
| 0% | | 42.6±9.9$^{e,f,g}$ | 44.2±11.0$^{f,g}$ |
| 5% | 282.1±33.1$^{d,e,f,g}$ | 42.3±10.7$^{e,f,g}$ | 43.7±11.8$^{f,g}$ |
| 10% | 289.8±30.9$^{d,e,f,g}$ | 41.8±10.7$^{e,f,g}$ | 42.6±10.9$^{f,g}$ |
| 20% | 305.3±30.0$^{b,c,e,f,g}$ | 41.4±9.7$^{e,f,g}$ | 44.1±9.3$^{f,g}$ |
| 40% | 323.9±33.9$^{b,c,e,g}$ | 37.4±9.8$^{a,b,c,d}$ | 47.6±8.9$^{g}$ |
| 60% | 327.2±33.8$^{b,c,e,g}$ | 31.3±10.2$^{a,b,c,d,g}$ | 50.5±9.0$^{a,b,c,d,g}$ |
| Ballistic | 506.8±70.1$^{a,b,c,d,e,f}$ | 36.3±8.7$^{a,b,c,d,f}$ | 30.2±10.8$^{a,b,c,d,e,f}$ |

Maximal joint velocity, $V_F$ and $V_{TT}$ data are pooled for all subjects at 35, 40, 45 and 50 deg of amplitude for each activation level. Two-way ANOVA (amplitude×activation level) were performed on values obtained at various activation levels (0%, 5%, 10%, 20%, 40% and 60% of MVC torque). One-way ANOVA (condition) were used to compare maximal joint velocity, peak $V_F$ and $V_{TT}$ between motor-driven quick releases at each activation level and ballistic contraction. Data are means±s.d.

*Values reported for the ballistic condition correspond to the mean velocity calculated from −15 to 30 deg of ankle angle.

$^{a,b,c,d,e,f,g}$Significant difference ($P$<0.05) from, respectively, 0%, 5%, 10%, 20%, 40% and 60% of MVC torque and ballistic contraction.

### Quick releases versus ballistic contractions

Maximal joint velocity measured in ballistic contractions was significantly higher than the unloaded joint velocity regardless of the activation level (Table 2; $P$<0.001).

Peak $V_F$ during quick releases was significantly higher than ballistic contraction from 5% to 20% of MVC ($P$<0.03), and lower at 60% of MVC ($P$<0.005). No significant difference was found between quick releases performed at 40% of MVC and ballistic contraction ($P$<0.2).

Muscle EMG activity patterns showed significant effects ($P$<0.001) for GM, SOL and TA muscles for the comparison between quick releases and ballistic contractions (Fig. 3).

## DISCUSSION

To the best of our knowledge, this study is the first to explore human fascicle–tendon interactions elicited by motor-driven quick releases *in vivo*, with the aim of checking the applicability of the slack test in human muscle. High frame rate ultrasound images revealed a rapid shortening of GM fascicles, with a peak velocity reached while the footplate underwent a high-accelerative rotation. While unloaded joint velocity increased with the initial activation level, our major finding was a concomitant decrease in peak $V_F$. In contrast, over the same period, $V_{TT}$ increased along with the increasing activation level, reflecting a larger contribution of tendinous tissues that probably explains the decrease in $V_F$. The highest peak $V_F$ was obtained in the passive condition and at low activation levels (from 0% to 20% of MVC), with values slightly higher than those for ballistic contractions. The analysis of muscle activation further suggests that neural control during the quick releases prevents full activation of the plantar flexor muscles and induces significant TA co-activation towards the end of the release. Together with the unavoidable and variable contribution of tendinous tissues to muscle–tendon unit shortening, these findings showed that the slack test in humans, as designed by Sasaki and Ishii (2005), is not transferable from the reference method used *in vitro* (Edman, 1979).

### Unloaded joint velocity

The mechatronic ergometer used in the present study elicited average peak motor angular velocity between 757 and 846 deg s$^{-1}$,

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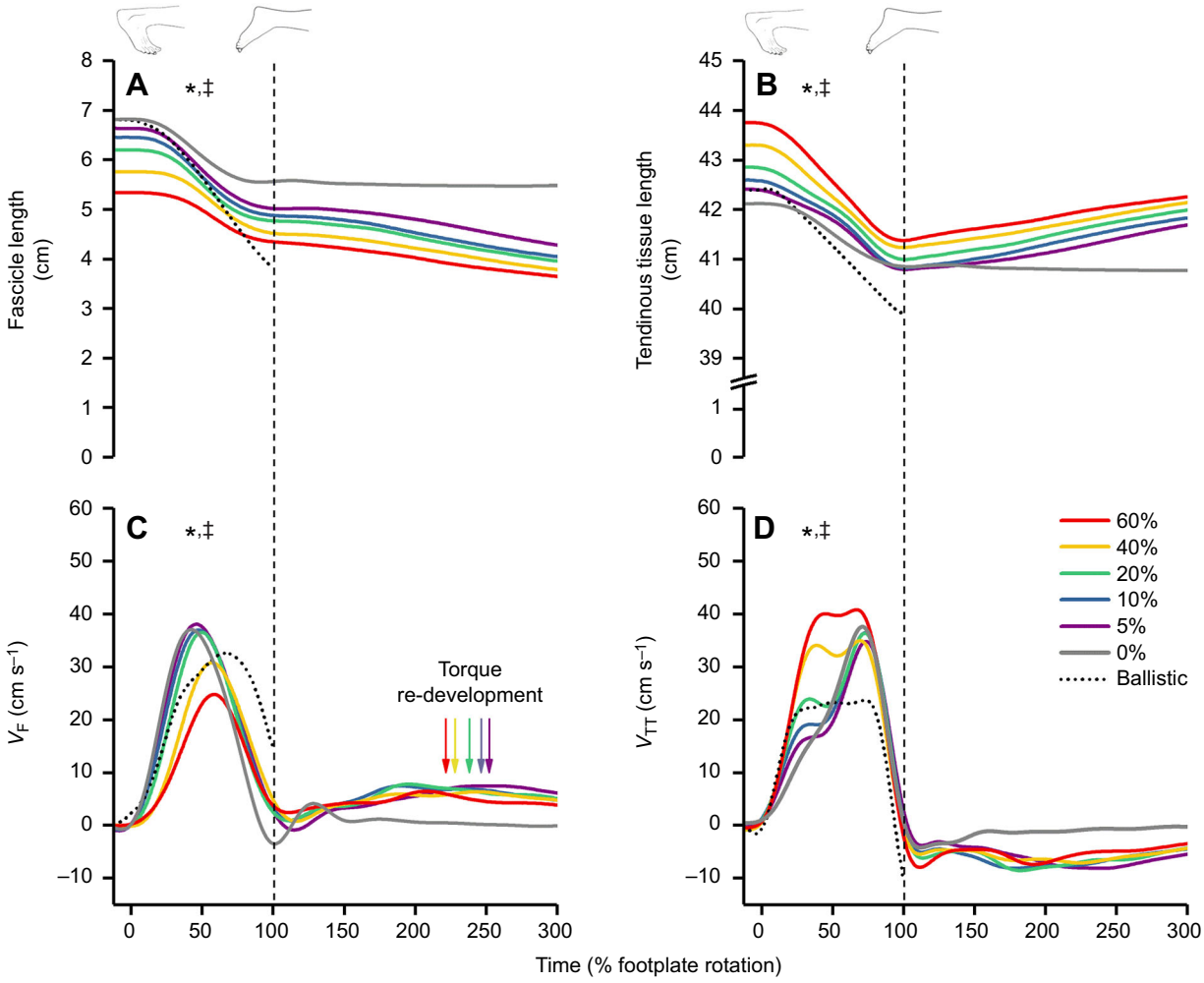



**Fig. 2. Fascicle and tendinous tissue behaviour according to activation level.** Fascicle length (A), tendinous tissue length (B), fascicle shortening velocity ($V_F$; C) and tendinous tissue shortening velocity ($V_{TT}$; D) patterns throughout motor-driven quick releases applied at various activation levels (from 0% to 60% of MVC) and during ballistic contractions. Time is expressed relative to the duration of footplate rotation (*x*-axis in %). Vertical dashed line corresponds to the end of footplate rotation. All amplitudes are pooled at each activation level. Coloured arrows in C correspond to the time of torque re-development for each activation level. For the sake of clarity, mean values are presented without s.d. *Significant difference between motor-driven quick releases and maximal ballistic shortening contractions for peak $V_F$ ($P$<0.03) and peak $V_{TT}$ ($P$<0.003). ‡Significant effect of activation level in the quick release on peak $V_F$ ($P$<0.001) and peak $V_{TT}$ ($P$<0.001). $n$=23.

consistently above the standard unloaded joint velocity threshold (i.e. >573.0 deg s$^{-1}$) (Hof, 1998). Nevertheless, 19% of the motor-driven quick releases were not adequately completed and consequently excluded from the analysis. This was mainly due to a lack of torque cancellation during the unloading phase or to the individuals' inability to exert positive torque at extreme plantarflexion positions, which prevented the detection of torque re-development. In line with previous findings (Sasaki and Ishii, 2005), unloaded joint velocity increased with initial activation level, reaching 327.2±33.8 deg s$^{-1}$ at 60% of maximal isometric torque (Table 2). This lower value compared with that obtained by Sasaki and Ishii (2005) (492.7 deg s$^{-1}$) could at least partly originate from the calculation method used to compute unloaded joint velocity (Claflin and Faulkner, 1985; Edman, 1979; Sasaki and Ishii, 2005). Overall, these findings also demonstrate the complexity of torque correction during movements involving such acceleration movement.

### Muscle–tendon interactions

Given that $V_F$ and muscle mechanics during maximal shortening contractions may not differ between the gastrocnemius lateralis and GM, the latter was used as a surrogate for plantar flexors. Because of the different fibre-type compositions between the GM and SOL (i.e. 49% versus 12% of type II fibres, respectively; Johnson et al., 1973), and the potential influence of the SOL on GM fascicle behaviour owing to shear and stress between aponeuroses, we attempted to simultaneously examine the changes in fascicle length in both muscles. Despite extensive pre-experimentation and set-up optimizations, it was not possible to track length changes in SOL fascicle reliably over the entire amplitude. This might be due to the complex architecture of the SOL, which involves 3D rotations particularly towards the end of plantarflexion movement (Azizi et al., 2008).

During the quick releases, $V_F$ reached its peak when joint torque was cancelled and motor angular velocity reached a very high value (Figs 1 and 2). Interestingly, GM muscle fascicles did not contribute to the increase in motor angular velocity observed as quick release amplitude increased (+38 deg s$^{-1}$ from 35 to 55 deg). Therefore, it is unlikely that faster quick releases would have increased $V_F$. The present experimental design allowed us to assess the maximal achievable $V_F$ during the release for trials which cancelled joint

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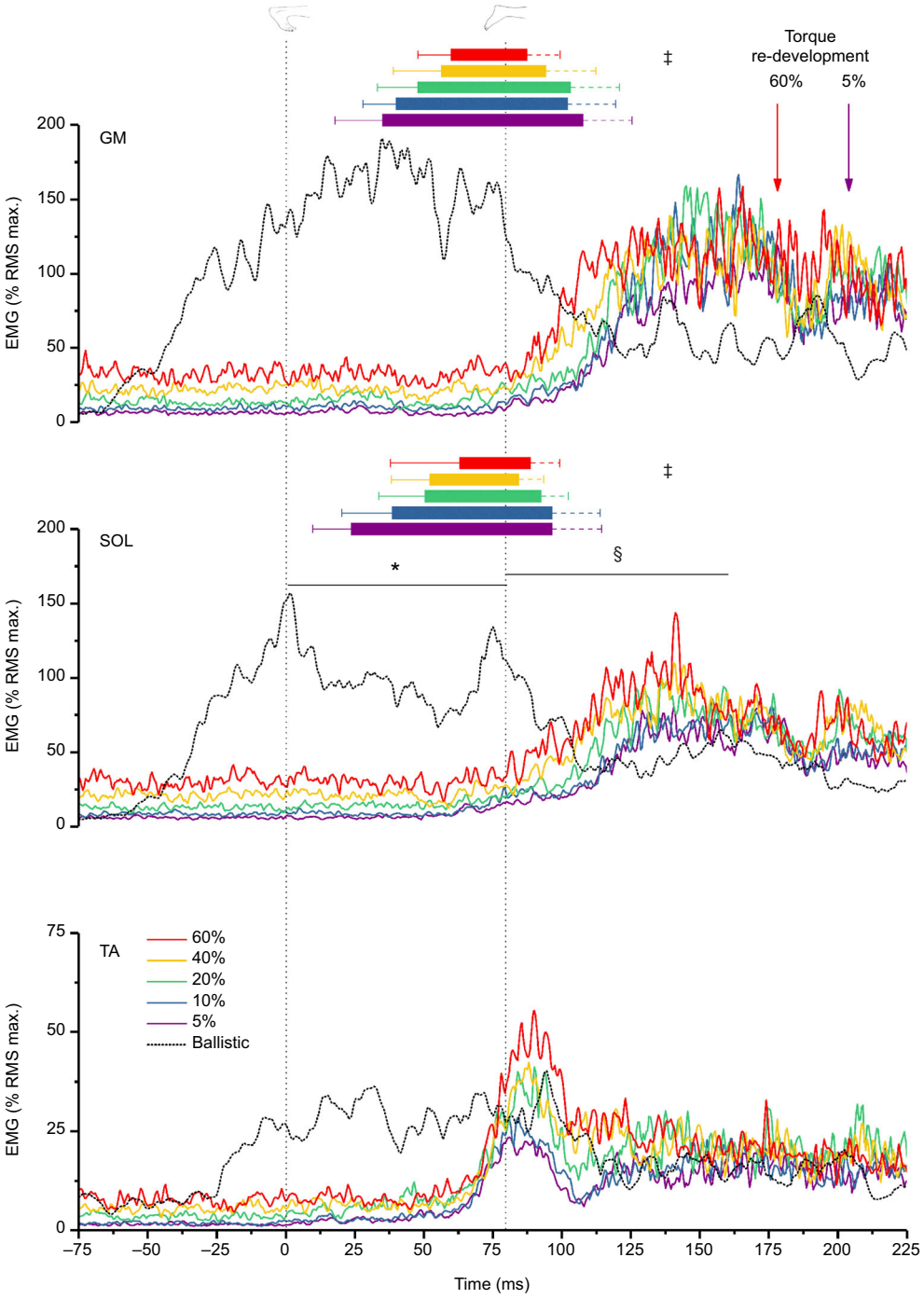



**Fig. 3. EMG activity patterns.** EMG activity patterns of gastrocnemius medialis (GM), soleus (SOL) and tibialis anterior (TA) muscles during motor-driven quick releases at different activation levels and ballistic contractions. EMG data were averaged before, during and after the quick release. Each value was calculated over the period of time corresponding to footplate displacement (78 ms). Average EMG patterns may hide significant individual silent EMG periods (unloading reflex) for GM and SOL following the motor-driven quick release. Therefore, the mean±s.d. duration of the silent period is displayed as horizontal bars for GM and SOL muscles. The time corresponding to the start and end point of the quick release and torque re-development at 5% and 60% of activation level is respectively displayed as vertical dotted lines and arrows. *Significant difference between slack test at all activation levels and maximal ballistic shortening contractions for EMG amplitude ($P$<0.001). §Significant effect of time on EMG amplitude during motor-driven quick releases ($P$<0.001). ‡Significant effect of activation level on duration of the silent EMG period after the completion of the motor-driven quick release ($P$<0.002). $n$=15.

torque. In these conditions, the obtained values decreased as the activation level increased, with lower values elicited at 60% of MVC compared with $V_F$ obtained in the ballistic condition (Table 2). The slack test therefore cannot determine the maximal shortening velocity of human muscle achieved upon voluntary activation.

Our results show that peak $V_F$ was maximal under the passive condition and decreased when the activation level increased ($-10.2$ cm s$^{-1}$ on average; Table 2). This is in contrast with *in vitro* (Phillips and Petrofsky, 1980) and *in vivo* experiments (Chow and Darling, 1999; Farcy et al., 2014), which found an increase in peak muscle shortening velocity with the increase in activation level. This discrepancy could be partly explained by the interaction between tendinous tissues and contractile elements during the motor-driven quick releases. Similar to the free quick release (Beaumatin et al., 2017; Farcy et al., 2014), muscle fascicles shorten without joint movement during the initial isometric contraction, while tendinous tissues stretch to store elastic energy. At the onset of footplate rotation, this energy is released through tendon shortening thanks to a ‘catapult-like’ process (Astley and Roberts, 2012). For greater initial activation level, the energy stored and released by elastic structures is increased. This would imply that a plantar flexor torque persists while the tendon shortens, and the muscle fascicles are never truly unloaded during motor-driven quick release.

The fact that $V_F$ was higher in low activation and passive conditions suggests that fascicle shortening may be driven by 'passive' processes. In these conditions, the recoil of the tendon is slower and therefore more of the shortening occurred in the muscle fibres in comparison to the longer activation levels. Indeed, because of the higher operating fascicle lengths measured in these conditions, the shortening velocity of the fascicles resulted from an increase in elastic energy release due to an increased passive tension in their non-contractile structures (Beaumatin et al., 2017; Claflin et al., 2011; Edman, 1979; Galler and Hilber, 1994), confirming that the muscle was not truly slack.

### Muscle activation

We found a significant increase in EMG activity in GM and SOL muscles with increasing activation level (Fig. 3). This rise in voluntary activation has been attributed to an increase in motor unit recruitment and discharge rate (Van Cutsem and Duchateau, 2005). Although a contraction close to MVC was required to recruit the entire pool of motor units in the SOL (Oya et al., 2009), pilot experiments performed above 60% of MVC resulted in more than 80% of non-completed motor-driven quick releases. Therefore, the shortening velocity of human plantar flexors was considered as being near maximal at this activation level (Sasaki and Ishii, 2005).

The time course of agonist and antagonist EMG patterns showed a transient decrease (i.e. post-silent period) in GM and SOL activity when muscle fascicles were expected to take up the slack until torque re-development. Chow and Darling (1999) suggest this transient reduction in agonist EMG activity is related to the unloading reflex at the end of footplate rotation. The duration of this post-silent period decreased with increasing activation level. This could help to explain why torque re-development occurred earlier, reflective of higher unloaded joint velocity at high activation levels. At the same time, the EMG activity of the TA (antagonist muscle) increased prior to the agonist burst. This antagonist activity may delay torque re-development (Chow and Darling, 1999; Marsden et al., 1976). In addition, the average motor unit discharge rate is substantially reduced when a ballistic contraction is preceded by an isometric contraction compared with an isolated ballistic contraction (Van Cutsem and Duchateau, 2005). A significant decline in the maximal rate of force development is associated with this lower motor unit discharge rate (Duchateau and Baudry, 2014), with assumed consequences for movement velocity, as suggested by our results (Fig. 3). The relative contribution of the SOL and GM to joint torque can also change according to movement velocity and loading (Duchateau et al., 1986). One could thus expect potential adjustments in load sharing between synergist plantar flexors under quick releases and maximal ballistic shortening contractions. Together, these neural processes might contribute to preventing the muscle from staying active during force re-development.

### Limitations to the use of the slack test method in humans

One of the main methodological issues with the slack test applied *in vivo* relates to the fact that fascicle–tendon interactions undeniably differ in passive and active conditions. These differences include changes in initial length of the muscle and tendon, and alterations in tissue stiffness upon contraction (Fig. 2A,C). The hypothesis that it is possible to take the influence of elastic properties into account using the passive torque–angle curve to adjust the torque signal recorded during the release is in turn questionable. Consequently, the common correction of passive tension (de Zee and Voigt, 2001) may not be appropriate to correct joint torque and to determine the onset of torque re-development accurately. This could contribute to the considerable variability in unloaded joint velocity inferred from the slack test method applied *in vivo*. Moreover, suppressed torque during the quick release did not ensure that the muscle was truly unloaded. Theoretically, the slack test method requires that the muscle goes slack almost instantaneously and that the length change is sufficient (Edman, 1979). Thus, once the tendon was slack, the muscle would then have to continue shortening and presumably achieve a maximum shortening velocity. Taking advantage of the high frame rate ultrasound technique, the results showed that the tendon is still shortening during footplate rotation and hence the muscle is unlikely to be slack upon motor-driven quick releases (Fig. 2). Overall, these limitations suggest further investigations to measure local muscle force *in vivo* in humans for the very high velocity level.

### Conclusion

In conclusion, although the present study recreated the required conditions to apply the slack test *in vivo*, this method could not assess maximal $V_F$ in plantar flexor muscles as performed for muscle fibres *in vitro*. Such an outcome may have three main causes: (i) the angular velocity imposed by the motor may not systematically unload the tendinous tissues; (ii) the activation-induced changes in muscle–tendon interactions upon loading impact the respective contributions of contractile and elastic structures to $V_F$; (iii) the occurrence of the unloading reflex substantially alters the neuromuscular system's ability to activate the whole pool of available motor units during motor-driven quick releases. Although quick releases performed under low loading conditions in humans are more prone to eliciting greater $V_F$ compared with maximal ballistic contractions, this is probably a passive process. Therefore, the slack test does not assess the true maximal shortening velocity of muscle fascicle in humans and does not appear appropriate for *in vivo* measurements.

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#### Competing interests

The authors declare no competing or financial interests.

#### Author contributions

Conceptualization: R.H., S.D., A.N., G.R., A.C., J.D., G.G.; Methodology: R.H., S.D., A.N., G.R., A.C., H.H., J.D., G.G.; Software: R.H., A.C., H.H., G.G.; Validation: R.H., A.C., H.H., G.G.; Formal analysis: R.H., S.D., A.N., G.R., A.C., H.H., J.D., G.G.; Investigation: R.H., S.D., A.N., A.C., G.G.; Resources: R.H., S.D., A.N., A.C., H.H., G.G.; Data curation: R.H., S.D., A.N., A.C., H.H., J.D., G.G.; Writing - original draft: R.H., S.D., A.N., G.R., J.D., G.G.; Writing - review & editing: R.H., S.D., A.N., G.R., A.C., H.H., J.D., G.G.; Visualization: R.H., S.D., A.N., G.R., J.D., G.G.; Supervision: R.H., S.D., A.N., G.R., J.D., G.G.; Project administration: R.H., G.G.; Funding acquisition: R.H., J.D., G.G.

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