Received: 1 May 2020 | Revised: 30 September 2021 | Accepted: 3 October 2021

DOI: 10.1002/mus.27434

CLINICAL RESEARCH ARTICLE

MUSCLE&NERVE WILEY

A new method to determine stretch reflex latency

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Abstract

Introduction/Aims: Motion artifact signals (MASs) created by the relative movement of intramuscular wire electrodes are an indicator of the mechanical stimulus arrival time to the muscle belly. This study proposes a method that uses wire electrodes as an intramuscular mechanosensor to determine the stretch reflex (SR) latency without lag time.

Methods: Gastrocnemius SR was induced by tendon tap, heel tap, and forefoot tap. The MASs recorded by intramuscular wire electrodes were extracted from background electromyographic activity using the spike-triggered averaging technique. Simultaneous recordings were obtained from multiple sites to validate the MAS technique.

Results: Using intramuscular wire electrodes, the MASs were successfully determined and extracted for all stimulus sites. In the records from the rectus femoris, MASs were also successfully extracted; thus, the reflex latency could be calculated.

Discussion: Wire electrodes can be used as an intramuscular mechanosensor to determine the mechanical stimulus arrival time to the muscle belly.

KEYWORDS
conduction time, reflex latency, tonic vibration reflex, T-reflex, whole-body vibration

1 | INTRODUCTION

Muscle spindle-based reflexes, T-reflexes, and tonic vibration reflexes are widely used in clinical and experimental research. To elicit T-reflex and tonic vibration reflex responses, muscle spindles are activated by mechanical stimulation, such as tendon taps, tendon vibration, whole-body vibration, or stretch. Mechanical stimulation is usually applied to a remote tissue (eg, tendon, sole of foot) instead of skeletal muscle containing muscle spindles. Hitherto, the methods measuring the stretch reflex (SR) latency used a sensor (eg, accelerometer, load cell) placed either on a mechanical stimulator (eg, hammer, vibration platform) or a body site (eg, skin overlying tendon or muscle belly) and used the output of these sensors to estimate the onset time point of mechanical stimulation.[1-7] However, the muscle spindles are not stimulated exactly at the time of application of the mechanical stimulus. The lag time for the mechanical stimulus to reach the belly of the muscle is not predictable and may vary from person to person and from muscle to muscle, depending on the structural properties of the muscle. Different muscles can have different tendon lengths, and depending on the level of contraction, the tendons can become stiffer and hence conduct mechanical stimulus to the muscle faster; the exact reflex time may vary.[8] Therefore, it is not possible to accurately estimate the true latency for muscle spindle-based reflexes unless the lag time is determined.

Abbreviations: Acc, accelerometer; CTMS, conduction time of the mechanical stimulus; CVMS, conduction velocity of the mechanical stimulus; EMG, electromyography; MAS, motion artifact signal; SEMG, surface electromyography; SR, stretch reflex; TRL, true reflex latency; WBV, whole-body vibration.

The mechanical stimulation used for activating SR also induces the relative movement of intramuscular wire electrodes, creating motion artifact signals (MASs).[9-14] Therefore, the MASs created by the relative movement of intramuscular wire electrodes are an indicator of the mechanical stimulus arrival time to the belly of the muscle. The present study proposes a novel method in which the intramuscular recording electrodes can be used as a mechanosensor. Instead of using an accelerometer or load cell, it can be a more accurate way to determine the exact latency of the muscle spindle-based reflex. The determination of the exact time of arrival of the mechanical stimulus to the belly of the muscle is crucial, as it can be used to determine the latency of muscle spindle-based reflexes more accurately. Since the SR latency is routinely used to diagnose neurological disorders and determine speed in sports and exercise sciences,[15-17] the more accurate the measurement of its latency, the more precise its use will be.

2 | METHODS

The study protocol was registered with ClinicalTrials.gov (NCT04347083). This research protocol has two phases: first, the method of measuring latency using the MAS technique was determined in Experiment I. The validity of this method was assessed in Experiment II.

2.1 | Subjects

Healthy volunteers aged 20–45 y were included in this study. The exclusion criteria were skin lesions, hemorrhagic diathesis, and absence of the Achilles tendon reflex. All the participants provided written informed consent for the experimental procedures, which were in accordance with the Declaration of Helsinki and were approved by the local ethics committee.

2.2 | Protocol

2.2.1 | Experiment I: Latency measurement with MAS technique

The participants were requested to lie down in the prone position. Surface electromyography (SEMG) electrodes and an intramuscular wire electrode were placed on the left gastrocnemius muscle belly. The Achilles tendon was tapped 20 times at 3- to 5-s intervals using a reflex hammer (Achilles tap), while the angle of the left ankle joint was 90°. Subsequently, the participants were asked to stand up and stand quietly on their left foot. To maintain static balance, the participants were asked to hold the wall. While the subject was standing on his/her left foot, 20 consecutive mechanical stimulations (taps) with 3- to 5-s intervals were applied to the left heel (heel tap) and subsequently to the left forefoot (forefoot tap) (Figure 1). The taps were applied by the same researcher (I.K.).

Data acquisition

The surface and intramuscular electromyography (EMG) recorded from the gastrocnemius lateralis and acceleration data were collected simultaneously using a data acquisition and analysis system (Power1401 MK 2 and CED Spike2® software, Cambridge Electronic Design Limited, Cambridge, England).

Disposable self-adhesive bipolar Ag/AgCl (Covidien Kendall, Dublin, Ireland) SEMG electrodes were placed on the left lateral gastrocnemius belly 4 cm apart (Figure 1). The skin overlaying the muscle was shaved, light abrasion was applied, and the skin was cleaned with alcohol to reduce the skin resistance. To record the intramuscular MASs induced by tap, a pair of silver wire (75 μm in core diameter) electrodes was used. Except for the cut ends, the wires were insulated with Teflon. This pair of wires was passed through the channel of the 25 G needle. The wire ends were then shaped like a fishhook. The distance between the ends of the two wires was approximately 3 mm. This pair of silver wires was inserted between two surface EMG electrodes to a depth of approximately 3 cm

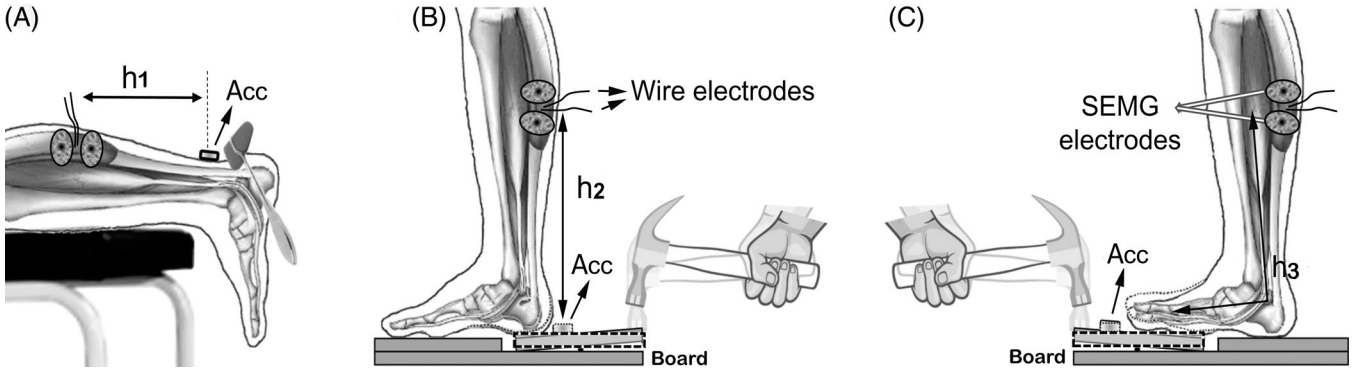

FIGURE 1 Illustration of the experimental setup. The EMG recordings were taken from the lateral gastrocnemius muscle. A, Using a reflex hammer, tendon taps were applied around the accelerometer fixed on the skin overlying the Achilles tendon (Achilles tendon tap). B, Using a hammer, tap was applied around the accelerometer fixed closely to the heel on the platform (heel tap). C, Using a hammer, tap was applied around the accelerometer fixed closely to the forefoot on the platform (forefoot tap). Acc, accelerometer; h1, the distance between the accelerometer fixed on the skin overlying the Achilles tendon and wire electrodes; h2, the distance between the heel and wire electrodes; h3, the distance between the first finger of foot and wire electrodes

into the lateral gastrocnemius. The needle was withdrawn, leaving a pair of fish-hooked wires inside the lateral gastrocnemius.[18] A lip-clip electrode was used as the ground electrode.[19] The EMG potentials were amplified 100 times and recorded at a sampling frequency of 20 KHz.

To determine the timing of the tap stimulus onset, a light (2.9 g) piezoelectric accelerometer (LIS344ALH; Ecopack, Mansfield, TX, USA) was firmly fixed using an adhesive tape on the skin overlying the Achilles tendon. The same accelerometer was subsequently firmly attached using adhesive tape on a platform placed under the foot (Figure 1). The acceleration signals were recorded at a sampling frequency of 10 KHz.

All the records were processed using infinite impulse response filters (Butterworth, 2nd order). The surface EMG recordings were filtered using a 20–500 Hz band-pass filter, while the accelerometer and intramuscular recordings were filtered using a 5–5000 Hz band-pass filter.

Extraction of motion artifact signals

The onset point of the tap-induced mechanical stimulus was visually marked on the accelerometer recording. Using this onset point as the trigger and the electrical signals recorded by intramuscular electrodes as the source, spike-triggered averaging was performed to extract MASs from the total background tension produced by the asynchronous firing of active synergist motor units (Figures 2 and 3).

Determination of the conduction time of the mechanical stimulus (lag time)

Using MASs recorded by intramuscular electrodes, the time required for the mechanical stimulus to reach the muscle belly was determined. The onset point of the motion artifact was visually determined using the spike-triggered averaging technique, where the acceleration signal was used as the trigger and the intramuscular signal as the source. The lag time between the onset point of mechanical stimulus on the acceleration signal and the onset point of intramuscular MAS was defined as "conduction time of the mechanical stimulus (CTMS)" (Figure 3). The CTMS varies depending on the distance between the point where the body site (Achilles tendon, heel, or forefoot) is mechanically stimulated, and the point where the pair of wire electrodes is inserted into the belly of the lateral gastrocnemius. The distance between the mechanically stimulated body site and the location of the wire electrodes was measured for each participant. For standardization, the CTMS was adjusted by the distance using the following formula:

$$\text{Adj CTMS} = \frac{D}{d}(\text{CTMS})$$

D: the mean distance between the point where the pair of wire electrodes is inserted into the muscle and the point where the body site is mechanically stimulated.

d: distance between the point where the pair of wire electrodes is inserted into the muscle and the point where the body site is mechanically stimulated.

We also calculated the conduction velocity of the mechanical stimulus (CVMS) using the following formula:

$$\text{CVMS} = \frac{d}{CTMS.}$$

Determination of reflex latency

The onset point of the tap-induced mechanical stimulus was marked on the accelerometer recording. Using this onset point as the trigger and the signals recorded by surface electrodes as the source, spike-

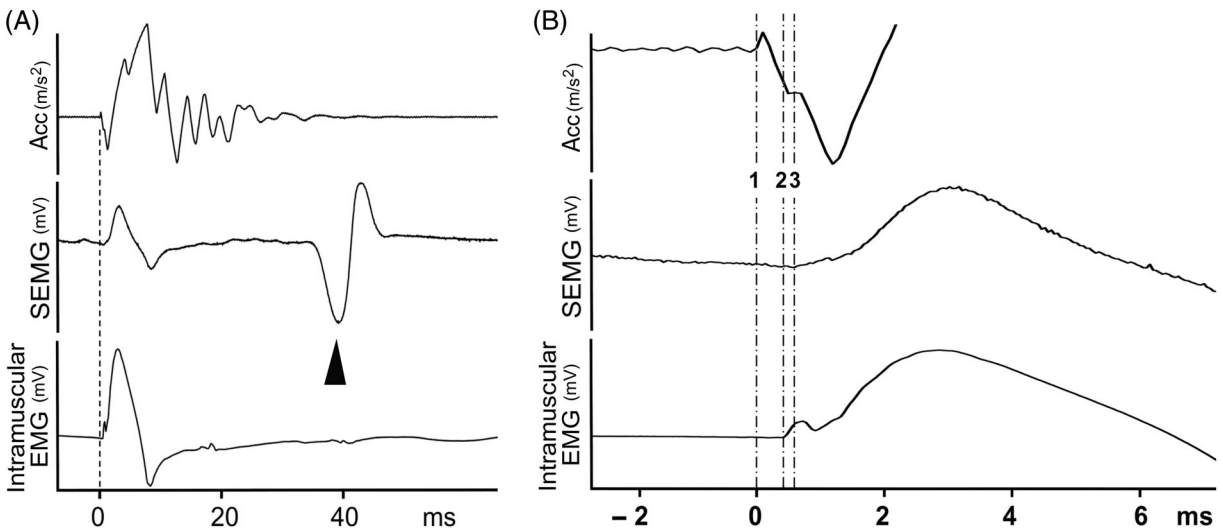

FIGURE 2 The representative signals of acceleration (Acc), surface EMG, and intramuscular EMG (raw data) for the forefoot tap Experiment I. All the signals were simultaneously recorded. A, the dotted line represents the onset point of the tap-induced mechanical stimulus determined by the accelerometer recordings. The arrowhead shows a reflex response. Since the onset point of motion artifact signal recorded by the surface and intramuscular electrodes cannot be visually distinguished from the onset point of the tap-induced mechanical stimulus in (A), all signals shown in (B) were magnified in the −2- to 6-ms range. B, In the intramuscular EMG recording, the onset point of the motion artifact, as shown by the second line is visible. However, in the surface EMG recording, the onset point of the motion artifact represented by line 3 is not evident. Line 1 represents the onset point of the tap induced-mechanical stimulus determined by the board accelerometer

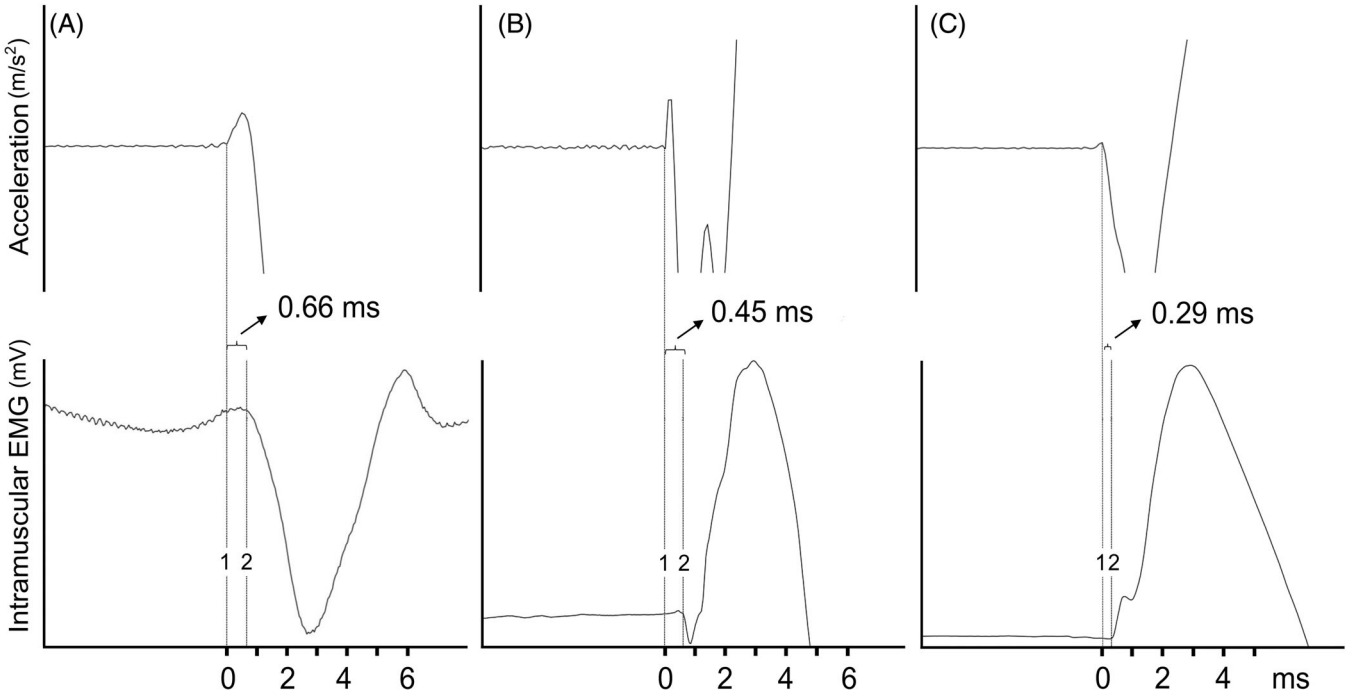

FIGURE 3 Representative acceleration and intramuscular EMG signals averaged using the spike-triggered averaging method for determining the onset point of motion artifact for the Achilles tendon tap (A), heel tap (B), forefoot tap (C). The first dotted line (1) represents the onset point of tap-induced mechanical stimulation in the acceleration signal, the second dotted line (2) represents the onset point of motion artifact

triggered averaging was used to determine the reflex latency (Figure 2).

The reflex latency was corrected using the formula below to calculate true reflex latency (TRL).

$$TRL = \mathrm{RL} - CTMS$$

RL: reflex latency obtained using spike-triggered averaging

The mean height of the participants was used to standardize the true reflex latency. The reflex latency was negatively correlated with body height.[20] Therefore, the reflex latency was adjusted for the height given in the formula below:

$$Adj\,TRL = (\mathrm{TRL})\,\frac{H}{h}$$

H: mean body height

H: body height of a participant

2.2.2 | Experiment II: Determination of the validity of the MAS method

Simultaneous recordings were obtained from multiple sites (gastrocnemius and rectus femoris) to characterize the movement artifact changes based on the location of the recording electrode. Two surface electrodes were placed on the belly of the left lateral gastrocnemius and left rectus femoris muscle, and a pair of intramuscular electrodes was inserted in the muscle belly between these surface electrodes. In addition to the board accelerometer, a piezoelectric accelerometer was placed on the skin between two surface electrodes for each of the rectus femoris and gastrocnemius muscles. Thus, the moment when the mechanical stimulus reached the belly of the muscle was determined using both the intramuscular electrodes and the skin accelerometer.

All records were recorded at a sampling frequency of 40 KHz and were processed using infinite impulse response filters (Butterworth, 2nd order). The accelerometers and intramuscular recordings were filtered using a 5–5000 Hz band-pass filter. The surface EMG recordings were filtered using a 5–500 Hz band-pass filter.

2.2.3 | Statistical analysis

The fit of the data to the normal distribution was tested using the Shapiro–Wilk test. The arithmetic means and standard deviations (SDs) were calculated for each variable. A paired t-test was used to compare the mean of the adjusted lag time for skin accelerometers and the lag time for intramuscular electrodes. The statistical significance was set at $P < .05$. The software package used for data management was PASW Statistics for Windows, Version 18.0 (SPSS Inc, Armonk, NY).

3 | RESULTS

A total of eight volunteers (one woman, seven men) participated in Experiment I, and five healthy volunteers (five men) participated in

TABLE 1 Summarized data of the acceleration, SEMG, and motion artifact

	Mean	SD	95% CI Lower	95% CI Upper
Distance for Achilles tap (h_1) (cm)	26.2	1.9	24.6	27.8
Distance for heel tap (h_2) (cm)	35.9	2.0	34.2	37.6
Distance for forefoot tap (h_3) (cm)	45.9	2.0	44.2	47.6
Adj CTMS for heel tap (ms)	0.20	0.06	0.15	0.25
Adj CTMS for forefoot tap (ms)	0.30	0.20	0.14	0.47
Adj CTMS for Achilles tap (ms)	0.64	0.35	0.34	0.93
CVMS for heel tap (m/s)	2014	717	1414	2613
CVMS for forefoot tap (m/s)	2062	1036	1196	2929
CVMS for Achilles tap (m/s)	598	481	195	1000
Adj TRL for heel tap (ms)	34.1	2.2	32.2	35.9
Adj TRL for forefoot tap (ms)	33.6	1.7	32.2	35.1
Adj TRL for Achilles tap (ms)	34.4	1.9	32.7	36.0

Abbreviations: Adj, adjusted; CI, confidence interval.

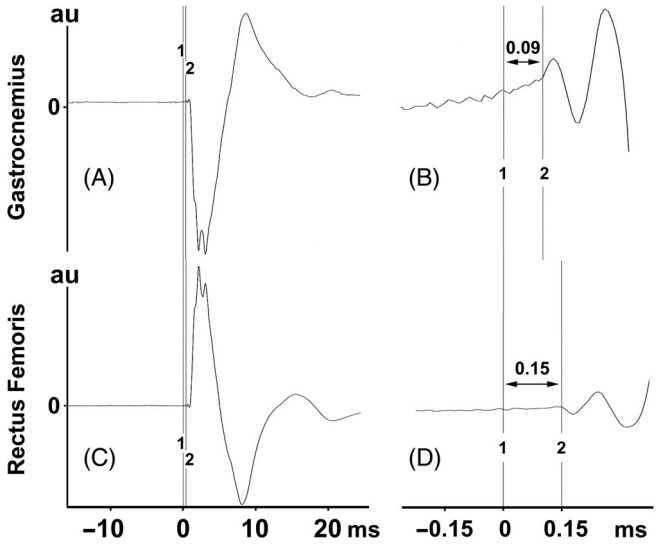

FIGURE 4 Representative MASs simultaneously recorded from the rectus femoris and gastrocnemius in Experiment II. Line 1 represents the onset point of the tap induced-mechanical stimulus determined by the board accelerometer. Line 2 represents the onset point of motion artifact signal recorded by intramuscular electrodes. Since the onset point of motion artifact recorded by intramuscular electrodes cannot be visually distinguished from the onset point of the tap induced-mechanical stimulus in (A) and (C), all the signals shown in (B) and (D) were magnified. The lag time for gastrocnemius and rectus femoris were 0.09 and 0.15 ms, respectively, in that participant

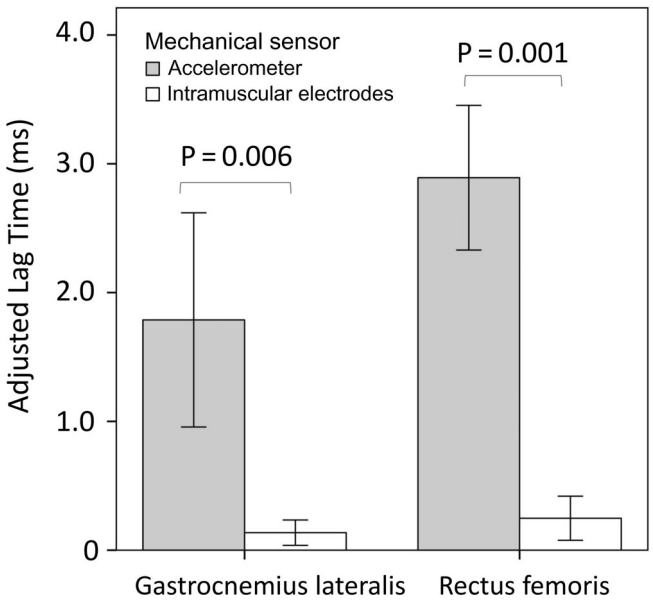

FIGURE 5 The adjusted lag time (conduction time of the mechanical stimulus). The lag time for the skin accelerometers is significantly longer than the lag time for the intramuscular electrodes

Experiment II. The mean ± SD of the age and body height were 29.4 ± 4.4 y and 177.1 ± 6.0 cm in the volunteers who participated in the Experiment I and 38.0 ± 4.5 y and 171.1 ± 7.3 cm in the volunteers who participated in the Experiment II.

Using the proposed method, intramuscular MASs were successfully extracted from the background EMG activity to determine the conduction time of the mechanical stimulus (lag time). In Experiment I, the mean conduction time of the mechanical stimulus for the Achilles tendon tap, heel tap, and forefoot tap was shorter than 0.7 ms. The CVMS of the Achilles tendon tap was approximately one-third of that of the heel or forefoot tap (Table 1). The surface EMG recordings showed that a reflex response occurred after the mechanical stimulation (Figure 2). The adjusted true latencies of these reflex responses for Achilles tendon tap, heel tap, and forefoot tap were 34.4, 34.1, and 33.6 ms, respectively.

The MASs were successfully extracted from intramuscular EMG recordings of the rectus femoris and gastrocnemius lateralis simultaneously taken during tapping (Figure 4). The mean distance between the board and the rectus femoris muscle belly was 71.0 ± 5.6 cm. The mean

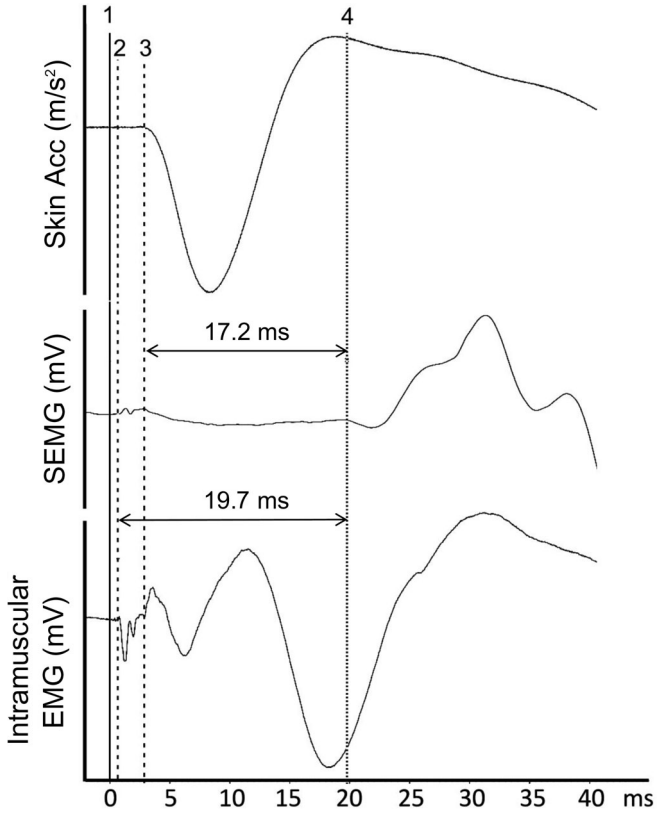

FIGURE 6 Determination of the actual reflex latency of the rectus femoris muscle for one participant. Line 1 represents the onset point of the tap induced-mechanical stimulus determined by the board accelerometer. Line 2 represents the moment when the mechanical stimulus detected by the intramuscular electrodes reaches the muscle (onset time of the mechanical stimulus). Line 3 represents the moment when the mechanical stimulus is detected by the skin accelerometer. The lag time for motion artifact signal recorded by the skin accelerometer was 2.52 ms. Line 4 represents the onset time of the rectus femoris reflex response in surface EMG. The lag time for the motion artifact signal recorded by intramuscular electrodes was 0.37 ms. the actual reflex latency was 19.7 ms

distance between the board and the lateral gastrocnemius muscle belly was 34.0 ± 1.9 cm in the Experiment II. This study showed that mechanical stimulation reaches the skin accelerometers placed on the gastrocnemius lateralis and rectus femoris muscle with a delay of 1.79 ± 0.67 and 2.89 ± 0.45 ms, respectively. This study also showed that the mechanical stimulation reaches the electrodes placed in the gastrocnemius lateralis and rectus femoris muscle with a delay of 0.14 ± 0.08 and 0.25 ± 0.14 ms, respectively. The adjusted CTMS determined using the skin accelerometer was significantly longer than the adjusted CTMS determined using intramuscular electrodes for both the rectus femoris and lateral gastrocnemius (Figure 5).

The velocity of mechanical stimulation reaching the intramuscular electrodes of the rectus femoris (CVMS) was 3606 ± 1661 m/s, and the velocity of reaching the intramuscular electrodes of the gastrocnemius was 3442 ± 2159 m/s. The adjusted true latency of rectus femoris and gastrocnemius reflex responses were 21.6 ± 4.5 and 36.3 ± 2.9 ms, respectively (Figure 6).

The participants reported feeling minimal pain during intramuscular electrode application. No other side effects or complications related to this technique were detected.

4 | DISCUSSION

The main findings of the present study are that using the proposed method, intramuscular MASs can be successfully extracted from the background electromyography activity; additionally, the wire electrodes can be used as an intramuscular mechanosensor to determine the reflex latency without the lag time for all stimulus sites. Simultaneous recordings obtained from multiple sites (the gastrocnemius and rectus femoris) validated the MAS technique. In the rectus femoris muscle, which is located farther from the mechanical stimulation point, MASs were also successfully extracted, and thus, the reflex latency could be calculated.

As the mechanical stimulus used to activate SR arrives in the belly of the muscle, it displaces the intramuscular electrodes and hence changes their relative positions against each other. Small changes in their relative position generate a weak potential in the recorded intramuscular activity.[9-14] According to Faraday's law, an increase in the rate of change of the position of the electrodes relative to each other and muscle fiber increases the power of the artifact signal.[21] To increase the rate of change of the position of the electrodes relative to each other, the tips of the silver wires were offset from each other by 3 mm to ensure a detectable change in the intramuscular potential recording. Therefore, by using the proposed method, intramuscular MASs can be successfully extracted.

Hitting (tapping) the platform or the tendon created mechanical oscillations that were verified with accelerometer records, defined as vibrations. The vibration propagates as a longitudinal wave in a medium (metals, biological tissue, etc.). While the longitudinal wave propagates through the medium, compressions and rarefactions occur between the particles of the medium.[22] The primary muscle spindles respond to vibrations of very low amplitudes (in the micron range).[23] Although vibration may not cause the lengthening or stretching of muscles, it activates the muscle spindles, as they are extremely sensitive to vibration.[24] The longitudinal vibration waves are transmitted to the muscles via the leg bones during whole body vibration.[1] Similarly, vibration traveling via the bone following the forefoot and heel tap can stimulate the muscle spindle in the gastrocnemius muscle. The reflex response induced by the heel tap and the forefoot tap was identified as the SR response because of mechanical stimulation (tap). Their latencies were similar to the latency of the T-reflex induced by the Achilles tap. These findings showed that the gastrocnemius muscle spindles and SR were activated by tap stimulation at all three stimulation sites (Achilles tendon, heel, and forefoot).

The longitudinal vibration waves (compressions and rarefactions) within a muscle tissue that activate the muscle spindles also lead to the motion artifacts by moving the electrodes in the micron range.[11]

Although the heel and forefoot were more distant from the intramuscular electrodes than the Achilles tendon, their CTMS was shorter than that of the Achilles tendon. The CVMS calculation showed that the mechanical stimulation applied to the sole of the foot (heel or forefoot) was transmitted to the gastrocnemius muscle belly much faster than the mechanical stimulus applied to the Achilles tendon. These findings support the view that mechanical stimulation applied to the sole of the foot is transmitted through the bones to the muscle.[1,25,26] Experiments I and II indicate that the mechanical stimulation conduction time and velocity may differ from person to person and from one region to another in the body. The source of this variability may be that the body tissues were not uniform. This variability indicates the importance and necessity of measurement with intramuscular mechanosensors (for example, with intramuscular electrodes as in this study) to detect the precise moment of stimulation of the muscle spindle.

This study showed that the skin accelerometers placed in the muscle belly detect the mechanical stimulus with a delay of a few milliseconds. The main reason for this delay may be that the rate of mechanical stimulus propagating along the bone to the skin may be reduced by the soft tissues between them. Consequently, the findings of Experiment II indicate that the estimation of the moment when the mechanical stimulus reaches the muscle spindle with the skin accelerometers placed on the muscle belly may be inaccurate.

One limitation of the method described in this study is that it is invasive; however, there are no non-invasive methods to achieve the same goal. Notably, this procedure is minimally painful, as this method uses thin (75 μm) intramuscular fine-wire electrodes.

In conclusion, the present study showed that thin silver wire electrodes could be used as intramuscular mechanosensors to determine the mechanical stimulus arrival time to the muscle belly. The recording of both the onset time of the mechanical stimulus on the muscle belly and the onset time of the reflex EMG response in the belly of the muscle make a perfect reflex circuit that can be used reliably in clinical and experimental studies. Therefore, the more precisely the latency is determined, the more accurately it can be applied to basic and clinical research.

ACKNOWLEDGEMENT

The authors also gratefully acknowledge the volunteers who participated in these experiments.

CONFLICT OF INTEREST

None of the authors has any conflict of interest to disclose.

ETHICS STATEMENT

We confirm that we have read the journal's position on the issues involved in ethical publication and affirm that this report is consistent with those guidelines.

DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available from the corresponding author upon reasonable request.

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How to cite this article: Topkara B, Aydin T, Corum M, et al. A new method to determine stretch reflex latency. *Muscle & Nerve*. 2021;64(6):726-733. doi:10.1002/mus.27434