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# The Influence of Different Elbow Angles on the Twitch Response of the Biceps Brachii Muscle Between Intermittent Electrical Stimulations

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## 1. Introduction

Prolonged or repeated contractions of skeletal muscles lead to impaired muscle action or to a decrease in force-generating ability. This phenomenon is due to fatigue. Fatigue may be caused by factors/processes within the muscle cells (peripheral fatigue) or by diminished activation from the central nervous system (central fatigue). When observing a muscle twitch, decreases in contraction amplitude, decreases in contraction speed, and prolonged relaxation phases are the main indicators of fatigue.

There are several mechanisms involved in muscle fatigue. The variation of responsible mechanisms has been termed the task dependency of muscle fatigue (Enoka, 2002; Mottram, 2005a; 2005b; Baudry, 2010).

The use of different protocols of electrical stimulation can characterize the variety of the task dependency of muscle fatigue. Two examples of the task dependency of fatigue are the phenomena known as low-frequency fatigue (LFF), first described by Edwards et al. (1977), and high-frequency fatigue (HFF), described by Bigland-Ritchie et al. (1979), Jones (1979) and Jones, et al. (1986).

Another possible variable in muscle response could be initial muscle tension/length. Muscle tension can influence the muscle activation pattern and the amplitude of muscle contraction. Two other phenomena also show relation between length of muscle and muscle contraction properties during electrical stimulation “the catchlike property” (Lee et al., 1999; Binder-Macleod and Ketlar, 2005) and “twitch potentiation” (Raissier, 2000; Place, 2005).

The level of fatigue depends on muscle length, in which the contractile response is measured. Using the human tibialis anterior muscle, Sacco et al. (1994) observed that, when

the muscle was fatigued at short muscle lengths, the decline in force was more pronounced than when the muscle was fatigued at optimal length. Although this result was confirmed by Gauthier (1993), other researchers (Fitch et al. 1985; McKenzie et al. 1987; Lee et al., 2007) have observed reduced fatigue in short muscle lengths.

In our preliminary study (unpublished data), we observed the influence of different elbow angles on muscle twitch parameters, measured with the tensiomyography (TMG) method on the biceps brachii (BB) muscle during short-acting electrical stimulation (ES). We observed a shorter contraction time in pre-stretched muscles (long muscles, elbow angle 5°) compared to relaxed muscles (short muscles, elbow angle 60°). The question that arose was whether this observation was an isolated phenomenon or whether similar changes would also occur in different circumstances.

Therefore, the present study explored the twitch-to-twitch effect of an intermittent stimulation protocol at variable muscle lengths (different elbow angles) on the muscle twitch response of the human BB muscle.

Our working hypothesis was that the changes in muscle response to intermittent electrical stimulation would be dissimilar at different elbow angles.

## 2. Materials and methods

### 2.1 Subjects

Nine healthy, sedentary subjects (6 men, 4 left-handed) ranging from 25 to 45 years old (mean of  $32.7 \pm 8$ ), with no history of muscle or joint problems, participated in this study. All subjects were informed of the purpose and procedures of the study and gave written, informed consent for their participation.

The local ethics committee approved the tensiomyographical measurements.

### 2.2 Measurements

The contractile properties of the BB muscles on the left and right side were measured with the TMG method, which is classified as a mechanomyographical (MMG) method based on the 1995 convention (CIBA Foundation Symposium, 1995). TMG is based on a displacement sensor detecting muscle belly enlargement in the radial direction. TMG was invented in 1997 (Valencic et al. 1997) and has since become more established (Dahmane et al. 2001; 2005; 2006; Zagar and Krizaj, 2005; Tous-Fajardo, 2010; García-Manso, 2011).

For the measurements, an inductive sensor, incorporating a spring with a coefficient of 0.17 N/mm, was used. It provided an initial pressure of approximately  $1.5 \times 10^{-2}$  N/mm<sup>2</sup> on the tip area of 1.13 mm<sup>2</sup>. The responses of the BB muscles on the right and left sides were compared.

The measured subject sat in a measuring chair. The measured arm was fastened to the frame with two bands to achieve isometric conditions during the measurement. In all of our experiments, isometric conditions were applied within physiological limits. Our referential definition for isometric contraction was “the total length of the muscle tendon complex remains constant.” The sensor location for each muscle was determined

anatomically, according to Delagi et al. (1975). Maximal muscle amplitude/response was used as an additional criterion for the optimal sensor position. For the BB, the sensor location was at the midpoint of the line between the lateral head of the clavicle and the head of the radius.

Muscle contraction was elicited by single-twitch electrical stimuli. Two self-adhesive electrodes were placed symmetrically around the TMG sensor. The anode was placed distally and the cathode proximally, 20-50 mm from the measuring point. Bipolar ES consisted of a single DC pulse of 1 ms in duration. A typical TMG record with parameters and definitions is shown in Figure 1. The measured parameters are shown in Figure 2. These parameters were the maximal amplitude of the signal ( $D_m$ ), the delay time from the stimulation to 10% of the maximal contraction ( $t_d$ ), the time of contraction from 10% to 90% of the maximal contraction ( $t_c$ ), the time of sustained contraction from 50% contraction to 50% of the relaxation ( $t_s$ ) and the relaxation time from 10% relaxation to 50% relaxation ( $t_r$ ).

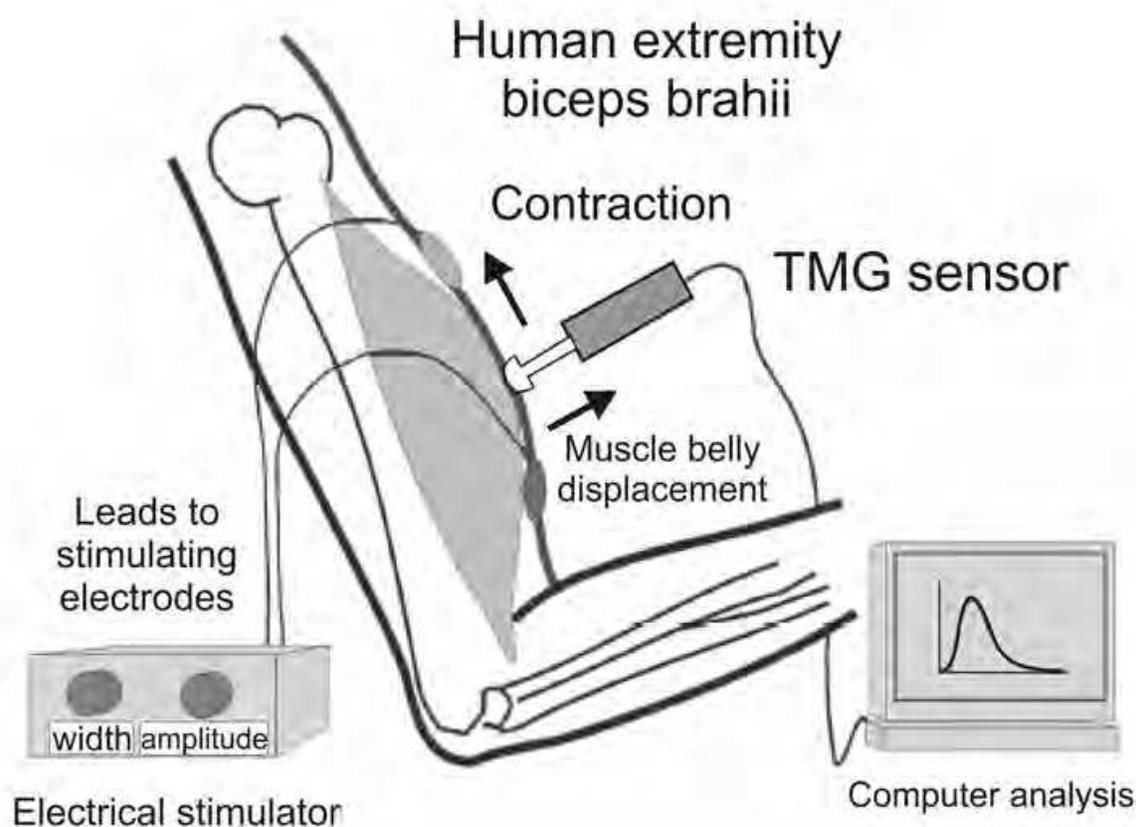


Fig. 1. Experimental setup used to evoke and measure the biceps brachii (BB) isometric twitch contraction responses. The TMG sensor measures muscle radial displacement during twitch contractions induced by short electrical stimuli. The stimulating electrodes are placed directly onto the skin.

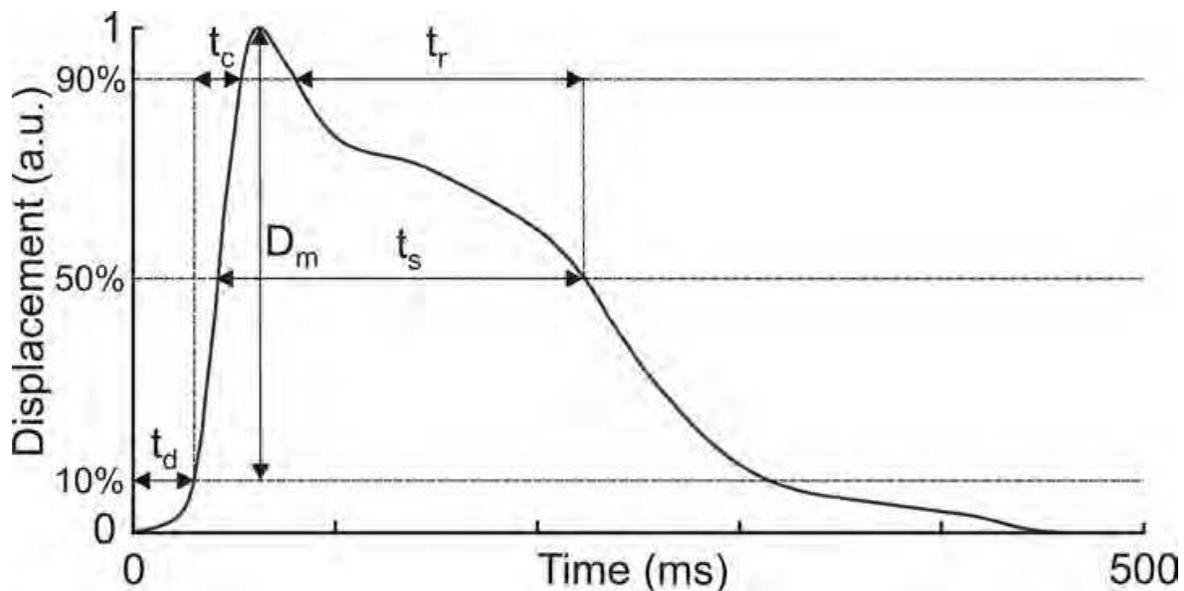


Fig. 2. (a) The parameters that were measured with the TMG signal:  $D_m$  – maximum amplitude (displacement),  $t_d$  – initial delay time,  $t_c$  – contraction time,  $t_s$  – sustained contraction time and  $t_r$  – half relaxation time.

### 2.3 Measurement protocols

Throughout the stimulation protocol, muscle activity was monitored with the TMG sensor. The stimulus intensity was set at 66% of a supramaximal twitch that was determined for a single 1-ms electrical impulse, before proceeding with the ES protocol for each muscle.

The electrical intermittent stimulation protocol (ISP) (Figure 3) consisted of 30 100-ms stimulation bouts (100 Hz, 0.1-ms impulse width) with 400-ms pauses between bouts, followed by a 1000-ms pause and an in-between twitch bout (IBT; also 100 Hz, 0.1-ms impulse width; Figures 3 and 4b), followed by a 900-ms pause. The protocol was repeated six times so that the whole stimulation lasted 90 s, which amounted to a total of 180 100-ms stimulation bouts. The entire protocol was flanked by two basic twitch stimuli (TS, single 1-ms impulse), one 3 s before and the other 3 s after end of the protocol.

The data were later read into Matlab (MathWorks, Natick, Massachusetts, USA) and analyzed with that software.

Measurements were performed on the biceps brachii muscles of both arms. On the left arm, the elbow angle was fixed at  $5^\circ$  (intermittent protocol of electrical stimulation at  $5^\circ$  [ISP5]), while the right arm was fixed at  $60^\circ$  (intermittent protocol of electrical stimulation at  $60^\circ$  [ISP60]). The reason for using the BB muscles of both arms was that the recovery from and/or the influence of a certain type of electrical stimulation can be quite prolonged. In severe cases, it may take as long as a few days to achieve full recovery (Jones et al. 1996).

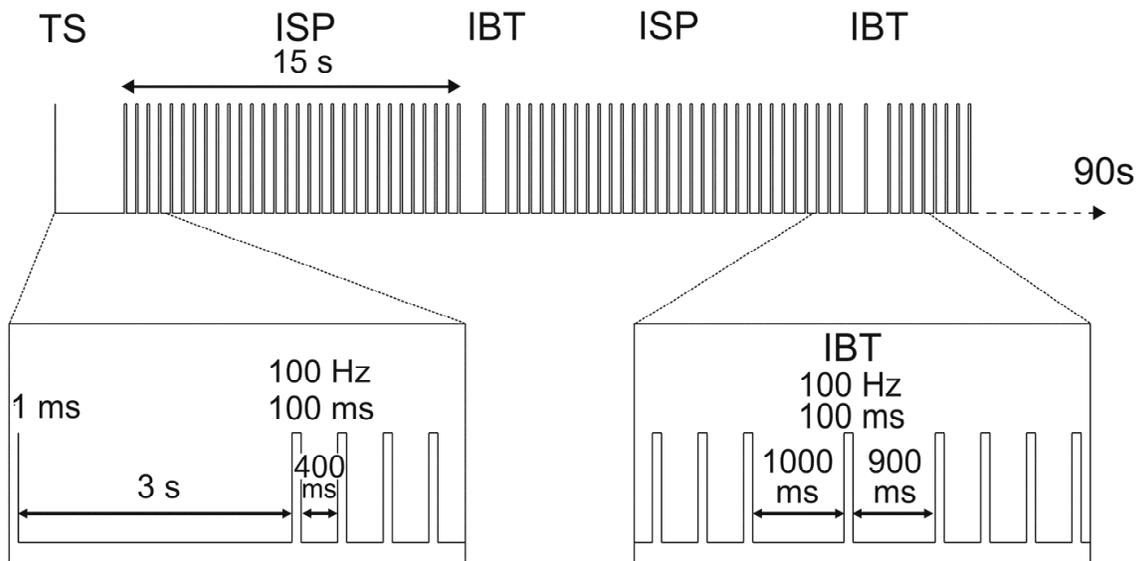


Fig. 3. Fatigue-inducing stimulation protocol: ISP = intermittent electrical stimulation protocol; IBT = in between twitch; TS = basic twitch stimulus.

The ISP was repeated six times so that the whole stimulation lasted 90 s, which amounted to a total of 180 100-ms stimulation bouts. IBT was repeated five times. The entire protocol was flanked by two basic twitch stimuli (a single 1-ms impulse), one 3 s before and the other 3 s after the end of the protocol.

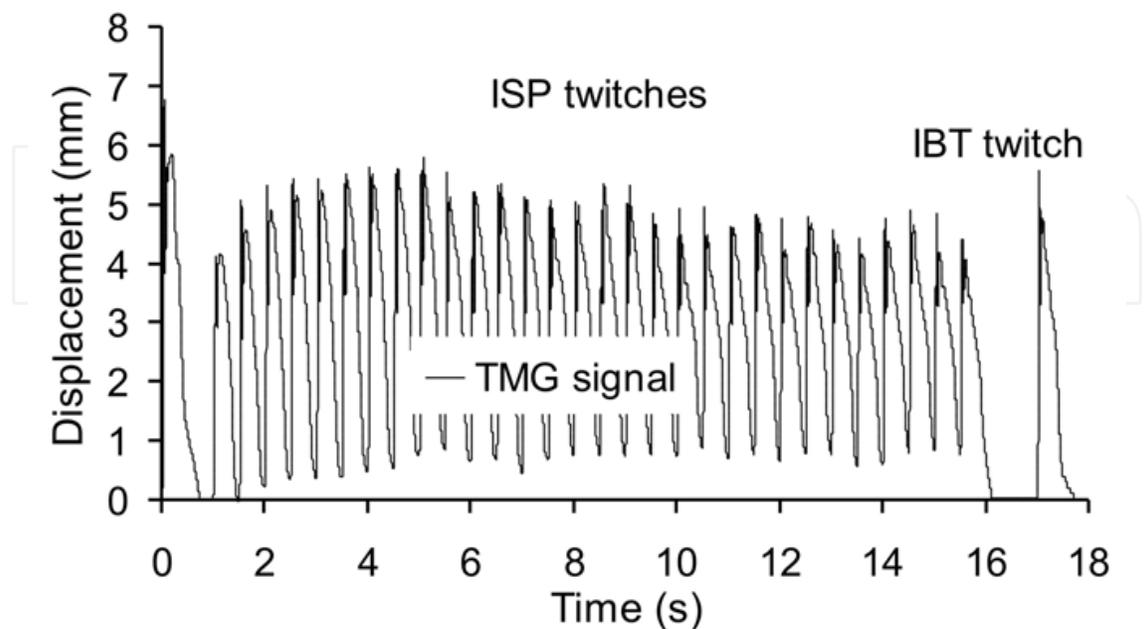


Fig. 4. (a) Muscle twitches between the first 18 s of ISP60, assessed on human BB.

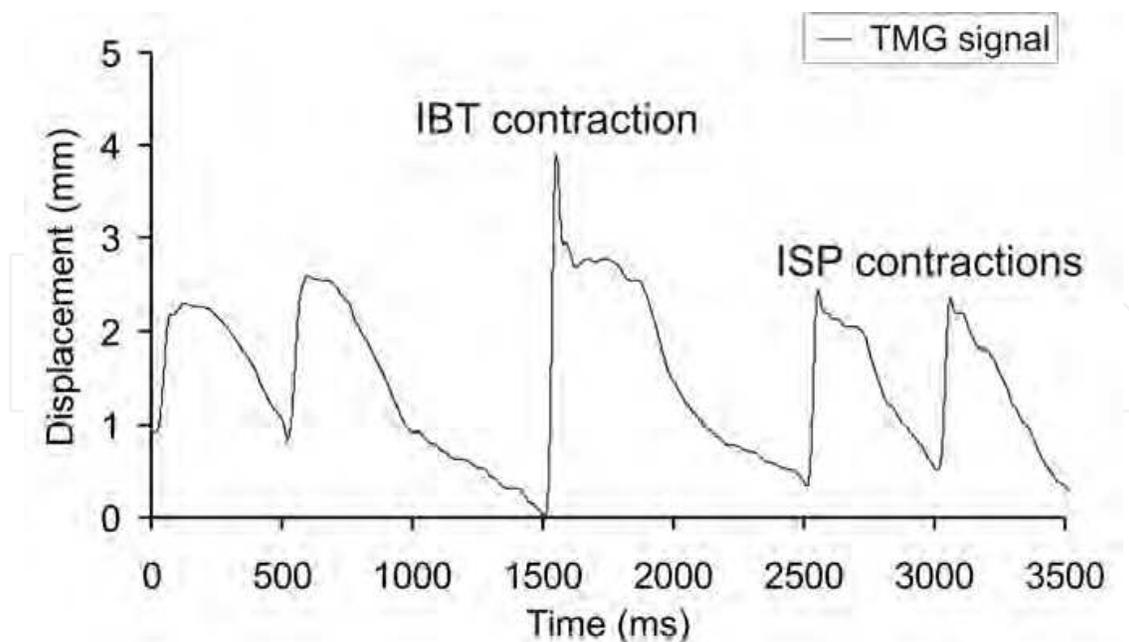


Fig. 4. (b) A detail with two types of twitches: IBT and ISP twitches.

## 2.4 Statistical Analysis

Paired-samples t-tests were used to compare differences in the changes in contraction parameters  $t_d$ ,  $t_c$ ,  $t_s$ ,  $t_r$ , and  $D_m$  as well as the differences between the two protocols (ISP5 and ISP60). Paired t-tests have greater power than unpaired tests when the paired units are similar with respect to "noise factors" that are independent of membership. Paired t-test was used to reduce the effects of confounding factors in our study. Significance for all tests was set at  $P < 0.05$ .

## 3. Results

A typical muscle response to ISP measured with the TMG sensor is shown in Figure 4a (first 18 s of 90-s intermittent stimulation with an extended or flexed elbow). An evident decrease in the contraction amplitude was observed during both ISPs at 15 to 20 s (Figure 5). There was a statistically significant change in responses during both protocols.

With the ISP5 protocol, we observed a significantly faster decrease in effective contraction amplitudes and, at the same time, a greater difference in amplitude decreases at the end of stimulation, compared to the ISP60 protocol (Figure 5). After 60 s of the ISP60 protocol, the ISP twitch amplitude was statistically unchanged.

The results of basic twitch response measurements before and after the ISP in long and short muscles are shown in Table 1.

The differences between time parameters ( $t_c$ ,  $t_d$ ,  $t_s$ , and  $t_r$  in long [5°] and short [60°] muscles) before the ISP were statistically significant, as shown in Figure 6c and Table 1. The difference between  $D_m$  in long (5°) and short (60°) muscles before the ISP was also statistically significant. For example,  $t_c$  was statistically shorter ( $t_c$  ISP60=26.3±1.1 and  $t_c$  ISP5=24.4±2.7,  $p < 0.05$ ) and  $D_m$  was smaller in long muscles ( $D_m$  ISP60=15.1±4.1,  $D_m$  ISP5=8.4±2.1,  $p < 0.001$ ).

In the basic twitch responses before and after the ISP60 protocol (Figure 6a and Table 1), there were no significant differences in  $t_c$ ,  $t_d$ ,  $t_s$ ,  $t_r$  or  $D_m$ .

In the ISP5 (Figure 6b) protocol, there were significant changes in  $t_d$ ,  $t_s$ , and  $t_r$  and no significant differences in  $t_c$  or  $D_m$ .

The IBTs had different dynamics of changes compared to those observed in twitches similar to contractions (ISP twitches), during both the ISP5 and ISP60 protocols.

From all the observed parameters in the IBTs during the ISP5 protocol, we found statistically significant differences in  $D_m$  only ( $p < 0.05$ ). The same pattern of response/changes was found with the ISP60 protocol in  $D_m$  only ( $p < 0.05$ ).

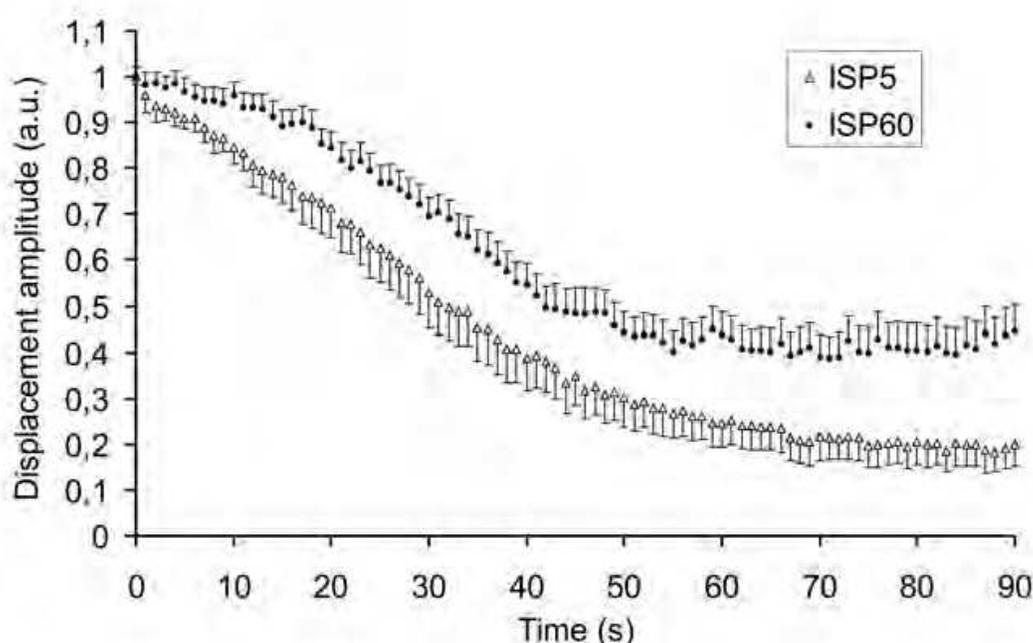


Fig. 5. The time course of the decline of maximal displacement amplitudes, normalized to the initial amplitude, during the 90 s of the stimulation protocol. ISP5 - open triangles  $\pm$  SE; ISP60 - black circles  $\pm$  SE. There was a statistically significant difference ( $p < 0.001$ ) between the two elbow angles (indicated on the figure).

	$t_c(\text{ms}) \pm \text{SD}$	$t_s(\text{ms}) \pm \text{SD}$	$t_r(\text{ms}) \pm \text{SD}$	$t_d(\text{ms}) \pm \text{SD}$	$D_m(\text{mm}) \pm \text{SD}$
ISP60 before	$26.34 \pm 1.18$	$153.02 \pm 33.47$	$104.33 \pm 36.00$	$23.75 \pm 1.81$	$15.14 \pm 4.22$
ISP5 before	$24.44 \pm 2.67$ *	$205.71 \pm 48.74$ ***	$169.64 \pm 47.88$ *	$22.31 \pm 2.25$ *	$8.50 \pm 2.06$ ***
ISP60 before	$26.34 \pm 1.18$	$153.02 \pm 33.47$	$104.33 \pm 36.00$	$23.75 \pm 1.81$	$15.14 \pm 4.22$
ISP60 after	$27.01 \pm 2.51$	$136.54 \pm 27.73$	$104.21 \pm 27.55$	$25.47 \pm 2.12$	$13.63 \pm 3.06$
ISP5 before	$24.44 \pm 2.67$	$205.71 \pm 48.74$	$169.64 \pm 47.88$	$22.31 \pm 2.25$	$8.50 \pm 2.06$
ISP5 after	$26.80 \pm 5.96$	$173.48 \pm 27.04$ §	$112.31 \pm 26.07$ §§	$23.84 \pm 1.44$ §	$7.69 \pm 1.70$

Contraction parameters from ISP60 are significantly different from ISP5 contraction parameters, \*  $P < 0.05$  and \*\*\*  $P < 0.001$   
 Contraction parameters from ISP5 before are significantly different from ISP5 after contraction parameters, §  $P < 0.05$  and §§  $P < 0.01$

Table 1. Results of basic twitch response measurements before and after the ISP in long and short muscles.

The dynamics (direction and size of changes) of IBT response during the ISP were not significantly different when comparing the ISP60 and ISP5 protocols.

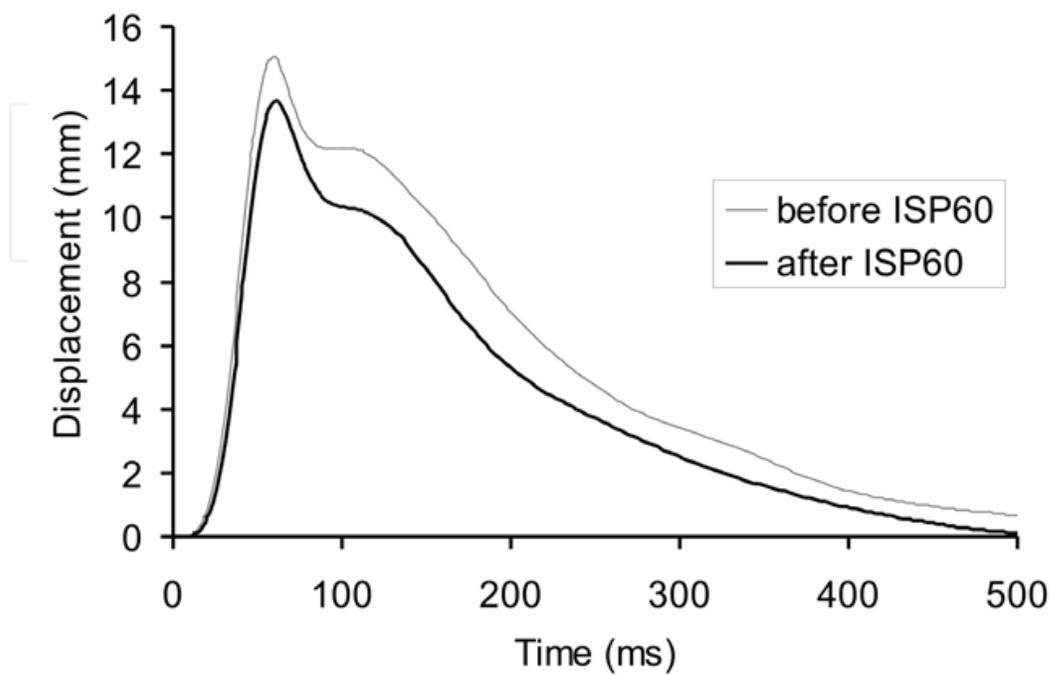


Fig. 6. (a) Twitch responses of the BB before and after ISP60. No statistically significant changes for any of the measured parameters were observed.

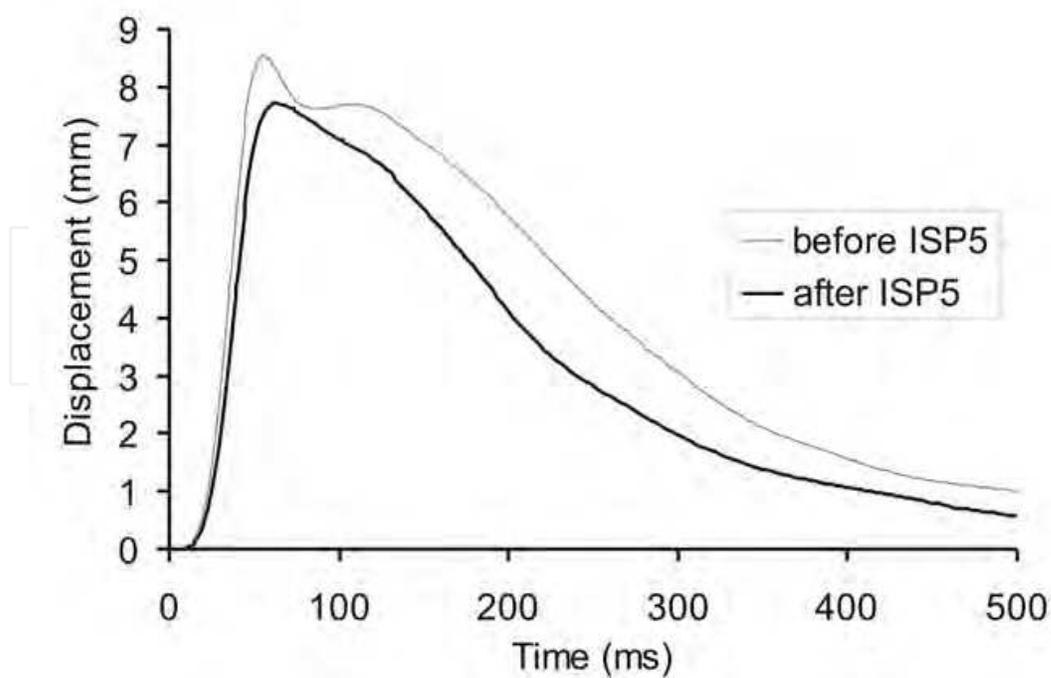


Fig. 6. (b) Twitch responses of the BB before and after ISP5. Statistically significant differences were observed for  $t_r$  and  $D_m$  ( $p < 0.05$ ).

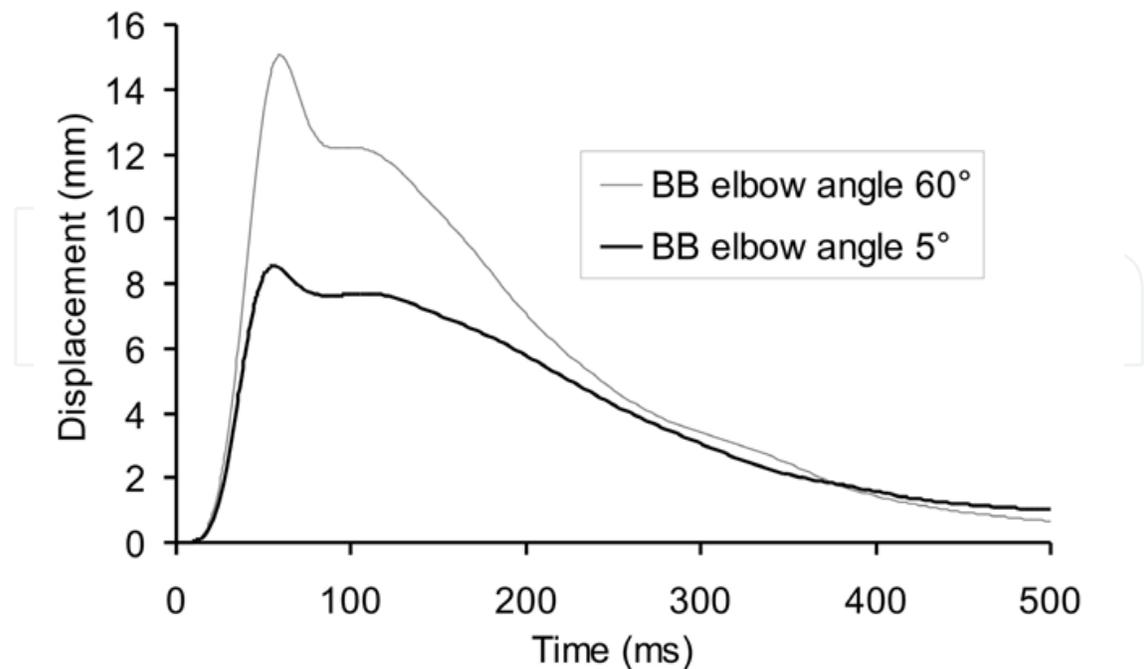


Fig. 6. (c) Twitch responses of the BB in the long muscle (elbow angle 5°) and short muscle (elbow angle 60°). The differences between  $t_c$  and  $D_m$  in the long (5°) and short (60°) muscles were statistically significant ( $t_c [60^\circ] = 26.3 \pm 1.1$ ,  $t_c [5^\circ] = 24.4 \pm 2.7$ ,  $p < 0.05$ )

#### 4. Discussion

TMG has been used in previous studies in which linear correlations were found between  $t_c$  and the percentage of type I muscle fibers (Dahmane et al. 2001) and between relative axial force and muscle belly radial displacement (Djordjevic et al. 2005).

The initial muscle length (length at the beginning of a contraction) is an extremely important modulator of muscle action. It has been established previously that several skeletal muscle physiological parameters depend on initial muscle length. Examples include the production of force/tension (Rassier et al. 1999), changes in motor unit activity (Ballantyne et al. 1993; Van Zuylen et al. 1988; Kennedy 2001),  $Ca^{2+}$  sensitivity (Stephenson, 1984) and the development of fatigue (Sacco et al., 1994; Fitch et al., 1987; Gauthier et al., 2000; McKenzie et al. 1987; Rassier, 2000, Lee et al., 2007).

All the cited authors studying the development of fatigue (except Fitch et al. 1987; McKenzie et al. 1987) showed that shortened muscles fatigue more quickly than extended muscles. At first glance, our data were not consistent with most of these studies; however, these studies are difficult to compare, as different measuring approaches were used as well as different muscle groups and stimulation protocols. These differences may explain the inconsistency of the obtained results.

In all previously mentioned studies, tetanic-type electrical stimulation was applied to induce fatigue. For example, in the study by Sacco et al. (1994), a fatigue protocol consisting of 6 15-s tetanic stimulations at 30 Hz (on the tibialis anterior) was applied. Rassier (2000) used a muscle fatigue protocol of nine tetanic contractions (50 Hz, 5 s in duration), with 5-s

intervals between contractions, on the quadricep muscle. Gauthier et al. (1993) used tetanic stimulation (train duration = 500 ms, duty cycle = 0.25) decreasing from 100 to 50 Hz with a 250-s duration on the rat diaphragm. In Fitch and McComas's (1993) study, the fatiguing procedure consisted of either indirect tetanic stimulation at 20 Hz or maximal voluntary contractions; each procedure lasted 90 s. Here, the observed muscle was the human ankle dorsal flexor muscle.

An important factor that may be responsible for the effects of stimulation is the recruitment order because this factor would affect the dynamics of fatigue. In our study, we used an intermittent type of transcutaneous electrical stimulation. Bursts of 100 Hz for 100 ms were given twice per second with two types of rest intervals (400 ms in between each pulse train and 1000 ms every 30 stimuli; Figure 3b), which was different from any of the previously published protocols. Our idea was to simulate the moderate cyclic activity of the muscles during fast walking, jogging or cycling. In this type of muscle activity, it is important that there is no overlapping of the contraction and the relaxation phases during fatigue development. Using the intermittent type ES, we wanted to avoid the effects of HFF, which can produce very dramatic losses of force/amplitude of contraction, and it is questionable whether HFF is a "normal" (physiological) fatigue mechanism (Jones 1996).

We believe that twitch intermittent-type ES has similar recruitment patterns to those found during voluntary action. A few studies have supported our contention that the recruitment order due to transcutaneous ES-induced contractions is non-selective (normal recruitment order versus reverse recruitment order) (Adams et al. 1993; Bickel et al. 2003; Binder-Macleod et al. 1995; Dahmane et al. 2005; Feiereisen et al. 1997; Knaflitz et al. 1990; Slade et al. 2003).

Nevertheless, with the ISP5 protocol, we observed an almost immediate drop in the twitch amplitude (Figure 3c), while with the ISP60 protocol, a decrease occurred after 10-20 s. This last observation could be attributed to a usual HFF response (Jones 1996). In the time frame of 10-20 s, the difference between the two protocols was most evident. After 60 s with the ISP60 protocol, the twitch amplitude was unchanged, while the twitch amplitude with the ISP5 protocol continued to decrease (Figures 5 and 7).

A statistically significant difference was observed between the two protocols regarding the twitch response of the biceps brachii, while the differences in basic twitches before and after the ISP were not significant, except for  $t_d$ ,  $t_s$ , and  $t_r$  when comparing the ISP5 and ISP60 protocols.

This finding can be explained by the different electrical stimuli and by the delay (1 s) at the end of the stimulation protocols before the 1-ms twitch. The reasoning for incorporating the 1-s delay into the protocol following the ISP, as opposed to the normal 400-ms delay between twitches, was to prevent the possibility of twitch fusion. The stimulation protocol is depicted in Figure 4b.

This study showed that the high data acquisition rate during the ISP resulted in a better assessment of the temporal components of the fatigue process. It also revealed that twitch measurements every 15 s (IBT) did not alone show statistically significant differences between the ISP protocols (it is true that the conditions were slightly different: 1000 ms versus 400 ms rest between 100 ms stimulus durations). The applied procedure and the recording method enabled a higher sampling rate (180 ISP twitches versus 7 IBTs), which resulted in a much

more precise (statistically significant) detection of changes occurring during the fatigue process. A summary of important results/ differences is shown in Figure 7.

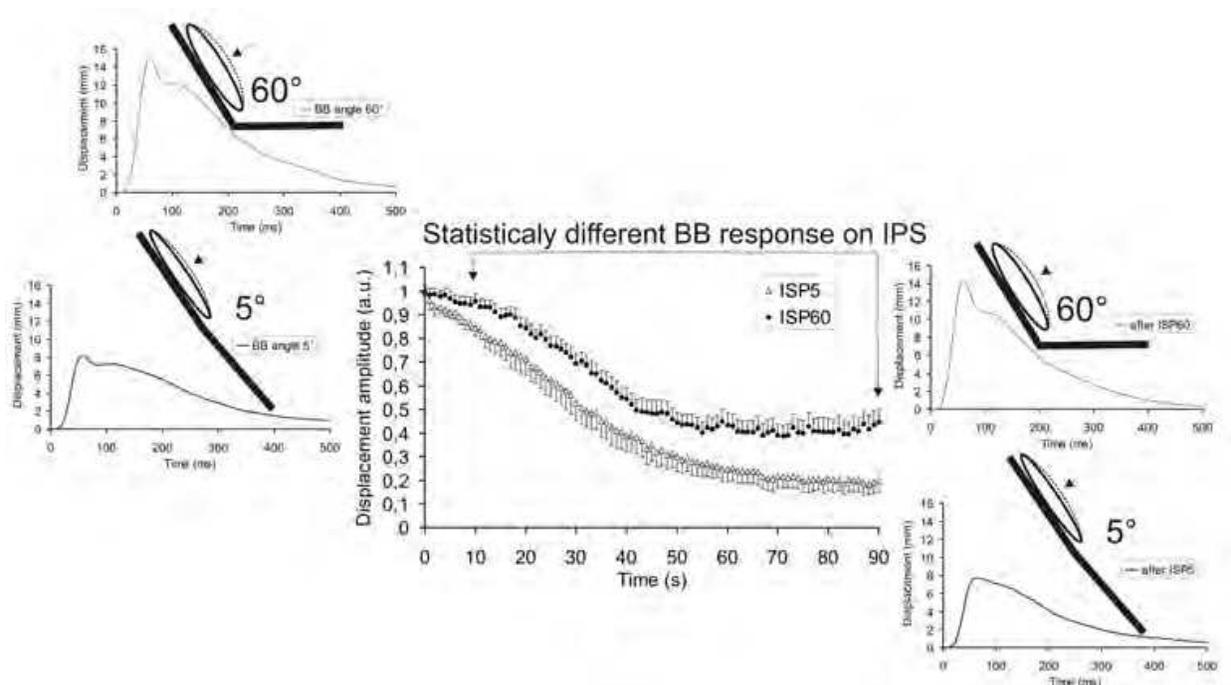


Fig. 7. The IBTs before (a and b) and after the ISP protocol (c and d) were measured at two different lengths of the BB muscle (elbow angle 5 and 60°). Shorter contraction time and faster fatigue (decline of contraction amplitude) were observed in the long muscle (elbow angle 5°). A statistically significant difference in fatigue development during the ISP was observed after 10 s for the two elbow angles (e). During ISP60, after 50 s, the BB displacement achieved a steady state; however, this finding was not observed during ISP5. The influence of muscle length was reflected in muscle contractions and twitch conditions and during fatigue conditions produced by the ISP.

The crucial question that we wanted to answer in this study was whether the shorter contraction times at smaller angles (longer muscle) were related to changed muscle activation patterns (motor unit recruitment order) or whether there were other mechanisms involved. Hence, we expected the fatigue process to be more pronounced if faster twitch fibers were recruited during contraction. Although this finding would not have been definite proof, it would have indicated that such a hypothesis could not be rejected. The results confirmed our working hypothesis. They showed a higher fatigue rate and a different time course in long muscles using the same ISP protocol. If we accept that the shorter  $t_c$  in long muscles means a greater percentage recruitment of fast twitch fibers, then a faster and more pronounced onset of fatigue during the ISP5 protocol seems to be a logical consequence.

## 5. Conclusion and future directions

Prolonged or repeated contractions of skeletal muscles lead to impaired muscle action or to a decrease in force-generating ability. The present study explores the twitch-to-twitch effect of an intermittent stimulation protocol at variable muscle lengths (different elbow angles) on

the muscle twitch response of the human biceps brachii muscle. Results showed a higher fatigue rate and a different time course in long muscles compared to short muscle using the same ISP protocol. For a better understanding of the detected changes and underlying processes, further research, in combination with other methods (EMG ...) and conditions, e.g., normalize(load) cyclic voluntary muscle activation is required.

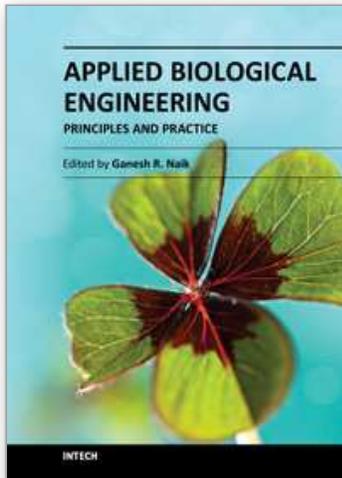
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Biological engineering is a field of engineering in which the emphasis is on life and life-sustaining systems. Biological engineering is an emerging discipline that encompasses engineering theory and practice connected to and derived from the science of biology. The most important trend in biological engineering is the dynamic range of scales at which biotechnology is now able to integrate with biological processes. An explosion in micro/nanoscale technology is allowing the manufacture of nanoparticles for drug delivery into cells, miniaturized implantable microsensors for medical diagnostics, and micro-engineered robots for on-board tissue repairs. This book aims to provide an updated overview of the recent developments in biological engineering from diverse aspects and various applications in clinical and experimental research.

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