

Altered reflex sensitivity after repeated and prolonged passive muscle stretching

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Avela, Janne, Heikki Kyröläinen, and Paavo V. Komi. Altered reflex sensitivity after repeated and prolonged passive muscle stretching. *J. Appl. Physiol.* 86(4): 1283–1291, 1999.—Experiments were carried out to test the effect of prolonged and repeated passive stretching (RPS) of the triceps surae muscle on reflex sensitivity. The results demonstrated a clear deterioration of muscle function immediately after RPS. Maximal voluntary contraction, average electromyographic activity of the gastrocnemius and soleus muscles, and zero crossing rate of the soleus muscle (recorded from 50% maximal voluntary contraction) decreased on average by 23.2, 19.9, 16.5, and 12.2%, respectively. These changes were associated with a clear immediate reduction in the reflex sensitivity; stretch reflex peak-to-peak amplitude decreased by 84.8%, and the ratio of the electrically induced maximal Hoffmann reflex to the maximal mass compound action potential decreased by 43.8%. Interestingly, a significant ($P < 0.01$) reduction in the stretch-resisting force of the measured muscles was observed. Serum creatine kinase activity stayed unaltered. This study presents evidence that the mechanism that decreases the sensitivity of short-latency reflexes can be activated because of RPS. The origin of this system seems to be a reduction in the activity of the large-diameter afferents, resulting from the reduced sensitivity of the muscle spindles to repeated stretch.

neuromuscular fatigue; central fatigue; muscle stretching; stretch reflex; electromyography

SEVERAL STUDIES have demonstrated that exhaustive and intensive stretch-shortening cycle exercise results in an acute reduction in performance with an associated decrease in the neural input to the muscle (30). It has also been shown that these changes occur concomitantly with reduced stretch reflex sensitivity (20). This is in line with the suggestion by Asmussen and Mazin (2) that the origin of the decline in motor unit activation is reflexly dependent on signals from the contracting muscle. As emphasized by Bigland-Ritchie et al. (5), this decline in motor unit activation is advantageous in that it helps to protect peripheral neuromuscular structures from excessive exhaustion and prevent impulse frequencies higher than those needed for a full tetanic activation of the fatiguing muscle fibers.

Two hypotheses have been put forward to account for the decline in reflex output. The first, proposed by Bigland-Ritchie et al. (3) and supported by Garland (14), relies on an inhibitory signal, probably provided by metabolically induced activity in small myelinated

and unmyelinated muscle afferents such as those belonging to groups III and IV. These afferents are mostly polymodal, being sensitive to several parameters associated with either metabolic fatigue or muscle damage (23, 31). It is also known that these receptors make a powerful input to inhibitory interneurons (8). According to Duchateau and Hainaut (10), the fatigue-induced metabolic stimulation of these muscle afferents might lead to presynaptic inhibition of the Ia terminals and/or inhibition of interneurons in the oligosynaptic pathways. This conclusion is supported by their finding that the Hoffmann (H)-reflex decrease does not recover as long as the fatigue-induced metabolic accumulation is maintained by ischemia.

The other hypothesis assumes disfacilitation of the α -motoneuron pool because of a progressive withdrawal of spindle-mediated fusimotor support (6, 18). In these studies, it was hypothesized (1) that, in sustained maximal voluntary contractions (MVCs), fatigue processes occur not only in extrafusal but also in intrafusal fibers and (2) that the intrafusal fatigue leads to a reduction in the voluntary drive conveyed to the α -motoneurons via the γ -loop. Macefield et al. (27) supported the same conclusion by directly measuring the discharge frequencies of the muscle spindle afferents. They demonstrated that in 72% of the measured afferents the discharge frequency declined progressively during submaximal isometric contraction and was inversely related to the change in electromyographic (EMG) activity.

In the present study, we sought evidence for some more direct fatigue effects on the muscle spindle itself by applying repetitive passive stretches on the relaxed muscle. This repetitive stretch could be assumed to cause compliance and/or stiffness changes not only in the tendon, in the extrafusal fibers, but also in the intrafusal fibers. These modifications could then result in changes in Ia afferent response because of changes in stretch to the receptor site in the spindle. These passive stretch conditions were further selected to eliminate as much as possible the fusimotor support to the muscle spindles as well as metabolic changes in the muscle (1).

METHODS

Subjects. Twenty healthy male subjects, aged 21–44 yr (mean 27 yr), participated in the study. None of the subjects had any history of neuromuscular or vascular disease. They were fully informed of the procedures and the risks involved in this study and gave their informed consent (code of ethics of the World Medical Association, Declaration of Helsinki). They were also allowed to withdraw from the measurements at will.

Experimental protocol. All 20 subjects underwent the repeated passive stretching (RPS) of the calf muscles (*proto-*

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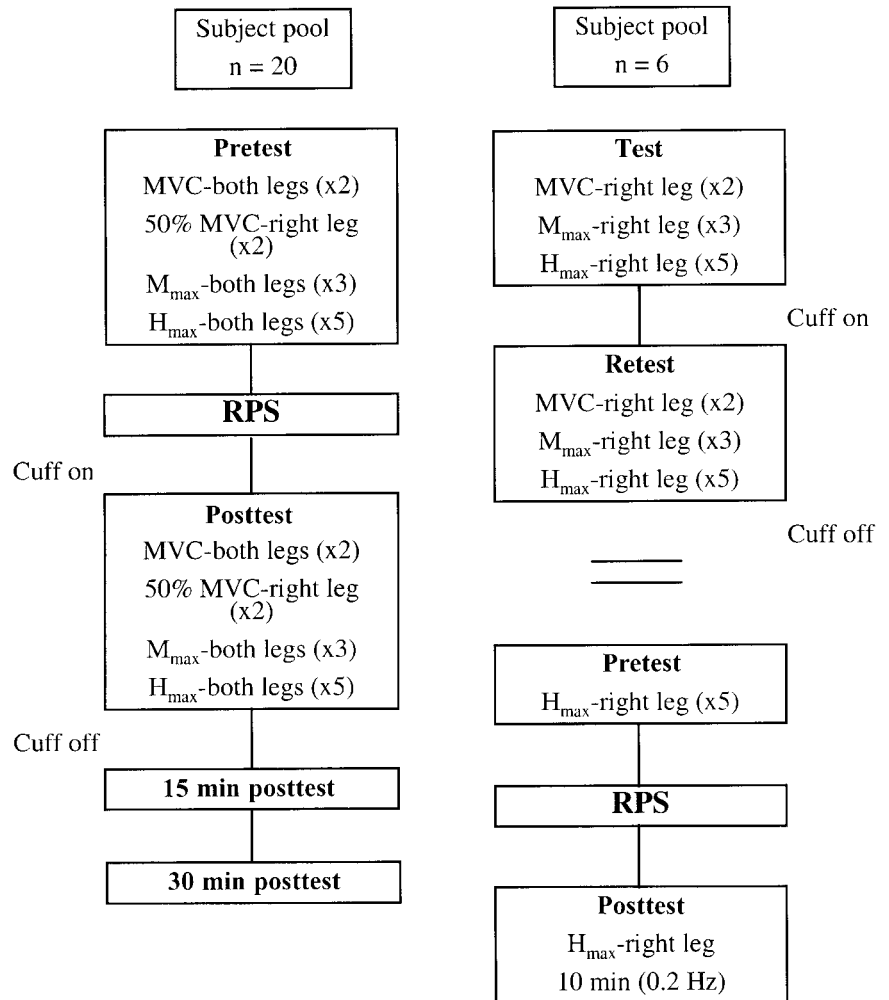
col 1), which lasted for 1 h. In this test the subjects were instructed not to resist the mechanical stretching of the calf muscles. Six of the 20 subjects were also tested for the effect of ischemia (protocol 2) and for the recovery of reflex excitability (protocol 3). The rather complex experimental protocol is summarized in Fig. 1.

In the RPS (protocol 1), pretest measurements included MVC and 50% MVC (2 measurements each). These measurements were followed by the respective maximal mass compound action potential (M wave; 3 measurements) and H-reflex testing (5 measurements). The absence of any background EMG was well controlled. This precaution was important because the H reflex is known to change if the soleus muscle is not fully relaxed (33). The three posttest measurements followed the same protocol; the first of these, however, was performed with ischemia. The complete sequence of pretest and posttest measurements occupied 120–180 s each. Within this period, at least 30 s were allowed to elapse between the MVCs and H-reflex recordings to avoid the problem of postcontraction depression of the H reflex (32). The same time delay was allowed to elapse between the maximal M-wave and H-reflex testing. The comparison of pretest and posttest values for the right leg served to identify the effects of the RPS. The long time interval between these measurements brought up the problem of the effects of any generalized behavioral and environmental factors on reflex excitability. To minimize these factors the contralateral leg was used as a nonstretched control leg, and it was always

measured after the experimental leg before and after the RPS. Therefore, the control leg was exposed to the same long time interval between the tests. The reflex excitability was calculated according to the method used by Garland and McComas (15). Maximal H-reflex peak-to-peak amplitudes were expressed in relation to the maximal M-wave peak-to-peak amplitudes (H/M ratio). Theoretically, the H/M ratios, so determined, should not have been affected by any changes in the peripheral excitability of the muscle fibers consequent on fatigue. The difference between the pretest and posttest H/M ratios was expressed as a percentage of the corresponding pretest value. Any percent change in H/M ratios from the control leg was then subtracted from the percent change on the experimental side so as to give overall reflex excitability. In this way, using a control leg eliminated the effects of any generalized behavioral and environmental factors due to the long RPS.

Stretch reflexes were measured as a unit of 10 consecutive stretches for both legs at the very beginning of the RPS, after every 15 min, and at the very end of the RPS. Recovery was tested 15 and 30 min after the RPS. The signal of RPS also served as a stimulus to induce stretch reflexes. Therefore, the experimental leg underwent continuous stretching, whereas the control leg was treated only during these short bouts of stretch reflex tests. While the final stretch-reflex changes were calculated, the relative change in the control leg was again subtracted from that in the experimental leg.

Fig. 1. Experimental protocol. Left: protocol 1 (repeated passive stretching; RPS). Right: protocol 2 (ischemia control) and protocol 3 (H-reflex recovery). n, No. of subjects; MVC, maximal voluntary contraction; M_{max}, maximum M wave; H_{max}, maximum H reflex.



Blood samples were drawn immediately after the RPS from the ulnar vein for determination of the possible indirect marker of muscle damage [serum creatine kinase (CK) activity]. Furthermore, capillary blood samples from the fingertip were taken for blood lactate determination 5 min after the dynamic stretching. Serum CK was analyzed by using a CK ultraviolet test kit (Boehringer Mannheim), and blood lactate was analyzed enzymatically by using a commercial kit (Biochemica Boeringer).

The measurements in the experimental leg immediately after RPS were performed under ischemia. On average, this lasted ~3 min. The purpose of this procedure was not to induce a nerve block but rather to prevent any premature recovery of possible metabolic changes. Ischemia was induced by a blood pressure cuff wrapped around the middle portion of the right thigh of the subject and was inflated to at least 200 mmHg. The impulse-conduction block progresses according to the fiber size, with large myelinated afferents being affected first (28). Therefore, there is a possibility that the 3-min ischemia itself can affect Ia afferent activity and reduce the reflex excitability. In addition, the cuff pressure can induce pain, leading to inhibition of the α -motoneuron activity. For these reasons, it was important to test the pure effect of a 3-min ischemia itself. Therefore, in the first control test (*protocol 2*), the posttests were measured under ischemic condition and then compared with the nonischemic pretest. These were done in the absence of the RPS. In the second control test (*protocol 3*), the RPS was again included and was preceded only by measurement of the maximal H reflex (5 measurements) in the experimental leg. During the posttest, only the recovery of the maximal H-reflex peak-to-peak amplitude was then measured.

Instrumentation and recording procedures. RPS was induced by repeated dynamic stretching of the calf muscles, performed by an ankle ergometer similar to that of Gollhofer and Schmidtbleicher (17). The stretch reflex responses were measured during the RPS with the same ergometer. In the ergometer, the stretching was applied by a motor torque device (Geisinger, 150 Nm) controlled by a digital feedback system. The torque around the rotational axes of the motor was measured by a piezoelectric crystal transducer (Kistler), and the angular movement of the ankle joint with respect to the plane of the ergometer was monitored by a linear potentiometer. In all the experimental conditions, the subject sat in a chair. Depending on the testing conditions, the thigh of the right leg or left leg was fixed and the foot was mounted on the rotation platform so that the rotation axes of the ankle joint and motor drive coincided. Therefore, only motion around the ankle joint was possible. The initial ankle position was 90° , and the knee angle was fixed to 120° . The stretching amplitude of the dorsiflexion of the ankle joint was 10° , and the corresponding average velocity of the stretch was 3.5 rad/s (Fig. 2). The waveform of the stretching signal was trapezoidal, and the frequency of these stretching units was 1.5 cycles/s. Throughout the experimental procedure, the legs

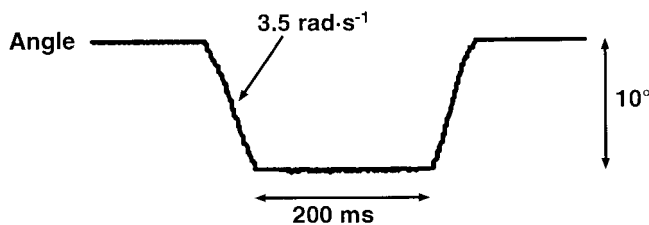


Fig. 2. Example of 1 repetition of repeated dorsiflexions of ankle joint (stretching of calf muscles). It also represents mechanical stimulation of stretch reflex.

were warmed with an infrared lamp, and the temperature of the skin was controlled during every test unit ($30 \pm 0.5^\circ\text{C}$). All the measurements included in this study were performed on the ankle ergometer.

A general methodology was used to record the H reflex. After the skin was prepared, stimulation electrodes (pregelatinized Ag-AgCl electrodes, Niko) were positioned bilaterally for the H-reflex and M-wave testing. The H reflex was evoked by the electrical stimulation of the Ia afferent fibers of the tibial nerve. The M wave (muscle compound action potential) was evoked by the stimulation of the motoneurons of the same nerve. The positions of the stimulus electrodes were tested first in the upright stance, then checked in the experimental position to ensure constant recording conditions. The intensity of the electrical stimulus was set in every testing unit to elicit maximal H response and M response. For each leg, the cathode ($1.5 \times 1.5 \text{ cm}$) was placed over the tibial nerve in the popliteal fossa, and the anode ($5 \times 8 \text{ cm}$) was placed superior to the patella. For H-reflex and M-wave testing, single rectangular pulses of 1-ms duration were delivered from an evoked-potential-measuring system (MEB-5304K, Nihon Kohden).

The recording electrodes for the H reflexes, M waves, stretch reflexes, and the EMGs associated with maximal (MVC) and submaximal (50% MVC) voluntary contractions were bipolar surface electrodes (Beckman 650437 miniature skin electrodes) fixed at a constant interelectrode distance of 20 mm. The electrodes were placed on both legs ~6 cm above the superior aspect of the calcaneus on the soleus muscle and between the center of the innervation zone and distal end of the lateral head of the gastrocnemius muscle.

EMG activity was transferred telemetrically, amplified by an FM microvolt amplifier (Glonner Electronic, Munich, Germany) and finally transferred through an analog-to-digital converter (sampling frequency was 1–3 kHz, depending on the signal) to a microcomputer. Ten consecutive stretch reflexes were averaged, and peak-to-peak amplitudes were analyzed together with latencies and simultaneously with resisting torques and angular displacements of the ankle joint. MVCs and 50% MVCs were analyzed trial by trial. In MVC, integrated EMG was divided by the integration time and considered as average EMG (aEMG). The H-reflex and M-wave recording signals were amplified (bandwidth 10 Hz–1 kHz), stored, and analyzed by the Nihon Kohden measuring system.

EMG activity during MVC and 50% MVC for 12 subjects was also recorded with fine wire electrodes. In the preparation of the wire electrodes, the electrode area was kept constant and clean by removing 2 mm of Teflon insulation from a 50- μm -diameter Evanohm wire (Wilbur B. Driver). The bipolar wire electrode was inserted into the soleus muscle laterally, ~10 cm above the muscle tendon region. The important stable connection between the electrode and the amplifier conductor was achieved with a spring-wire coil connector. The EMG signal was amplified (bandwidth 20 Hz–1 kHz) through a Nihon Kohden measuring system and stored in a microcomputer to analyze the number of times that the amplitude of the signal crossed the zero value of the signal [0 crossing rate (ZCR)] and the median frequency, on the basis of fast Fourier transformation. Individual values were calculated as an average of three overlapping windows (1,024 ms each).

It was important to ensure that the EMG responses came from the examined muscle only. Therefore, EMG cross talk measurements, similar to those reported by Moritani et al. (29), were performed. Near-maximal percutaneous stimulations (Neuropack, 4 miniature, 30–50 mA, 1-ms rectangular

pulse wave) were delivered to evoke compound mass action potentials (i.e., M waves) in gastrocnemius. The extent of cross talk was determined by the relative amplitudes of the M wave recorded from the soleus. In these recordings, the mean peak-to-peak M-wave amplitude was 9.20 ± 3.61 mV for the gastrocnemius and 0.42 ± 0.35 mV for the soleus, resulting in a cross talk of $4.76 \pm 4.45\%$. This value was lower than the 6% reported by Moritani et al. It can therefore be assumed that the extent of cross talk between these muscles was negligible in the present experiment.

Statistical analysis. Mean and SD values were calculated for the various parameters in all the different tests. In the RPS (protocol 1; $n = 20$), the statistical significances of the different parameters between tests and between legs (experimental vs. control) were determined by using double multivariate analysis of variance. When a significant *F*-ratio occurred for the main effects, profile analysis was carried out by multivariate analysis of variance to locate the source of the difference. Correlation coefficients were calculated to determine the relationships between selected parameters. For protocols 2 and 3 ($n = 6$), only descriptive statistical methods were employed, including some regression plots.

RESULTS

RPS ($n = 20$). After 1 h of repeated stretching of the triceps surae muscle, maximal voluntary plantar flexion torque in the subjects decreased on average by $23.2 \pm 19.7\%$. This was also the case for the amount of neural input to the gastrocnemius and soleus muscles as expressed by the relative reduction in the average EMG values of 19.9 ± 29.4 and $16.5 \pm 24.4\%$, respectively. These reductions resulted in nonsignificant changes in the EMG-to-force ratio. The follow-up tests revealed total recovery 15 min after the RPS (Fig. 3).

The recordings of the stretch reflexes showed a dramatic peak-to-peak reduction 15 min after the beginning of the RPS. This reduction continued along with RPS (Fig. 4). The maximal H reflex, which was not measured during the RPS, declined by $46.1 \pm 38.3\%$ immediately after the RPS. This reduction was not associated with a decrease in the maximal M wave, indicating that there was no failure in muscle fiber excitation or slowing in the impulse conduction in the muscle fibers. Therefore, the changes in the maximal H reflex resulted in a reduced maximal H/M ratio (mean decline $43.8 \pm 41.4\%$), suggesting impaired excitation of the α -motoneuron pool. Figure 5 was constructed to reveal all the above-mentioned changes in the original analog-signal mode in one subject. In the H-reflex recordings, it can easily be seen that the appearance of the maximal H reflex in relation to the M-wave level has changed.

There was also a moderate fall (nonsignificant) in the stretch reflex peak-to-peak amplitude and maximal H/M ratio of the control leg. This was probably due to crossover afferent influences from the experimental leg or changes in any generalized behavioral or environmental factors as suggested by Garland and McComas (15). To minimize nonspecific influences in the final calculations of reflex excitability (Fig. 4), the effect of the control leg was subtracted from that of the experimental leg, as described in METHODS.

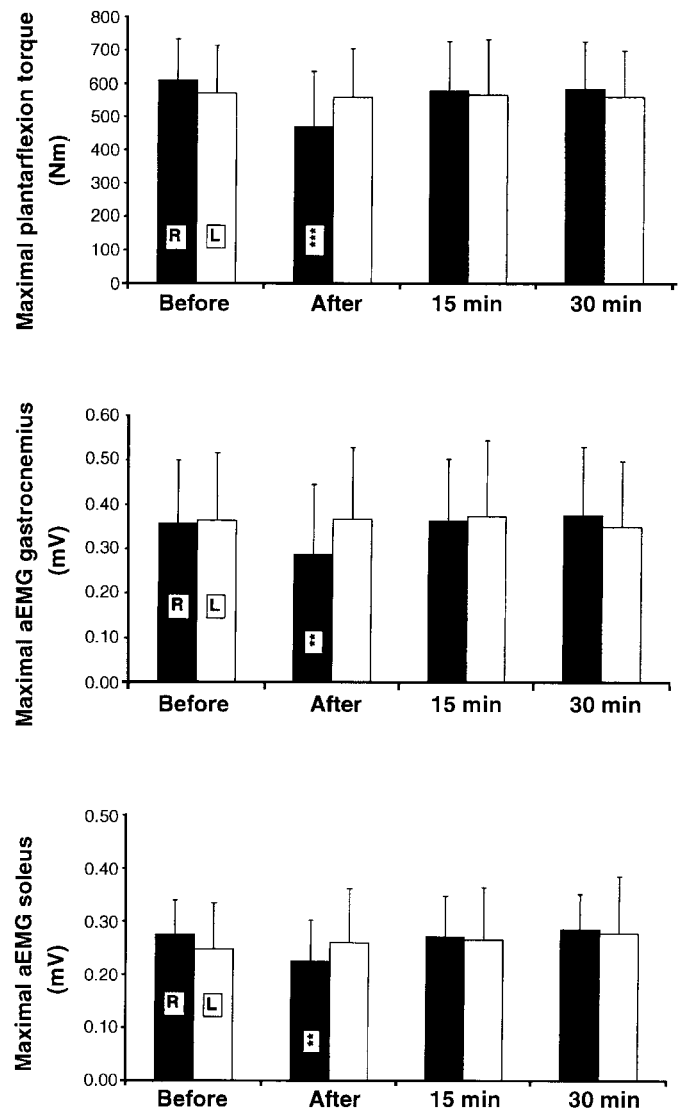


Fig. 3. Mean values (\pm SD) for all 20 subjects in maximal plantar flexion torque (top) and maximal gastrocnemius and soleus average EMG (aEMG; MVC) resulting from RPS (middle and bottom, respectively). R, experimental leg; L, control leg. Statistically significant difference compared with before-RPS condition: ** $P < 0.01$, *** $P < 0.001$ ($n = 20$).

The metabolic parameters revealed a slight but nonsignificant post-RPS increase. Mean peak lactate concentration increased from 1.84 ± 0.3 to 2.12 ± 0.6 mmol/l (nonsignificant). For serum CK concentration, this increment increase was from 262.7 ± 164.5 to 284.2 ± 172.4 U/l (nonsignificant). These small changes could be possibly explained by the metabolic activity due to MVC and 50% MVC. However, the nonsignificant nature of these changes is in line with the study by Armstrong et al. (1).

Motor unit firing rates ($n = 12$). Changes in the motor unit firing rates were estimated indirectly from the 50% MVC. The EMG data were recorded with fine wire electrodes to increase the selectivity of the recording, and the ZCR was then analyzed. The data showed a $12.2 \pm 11.4\%$ ($P < 0.01$) decrease in the ZCR immediately after the RPS. According to Lindström et al. (26),

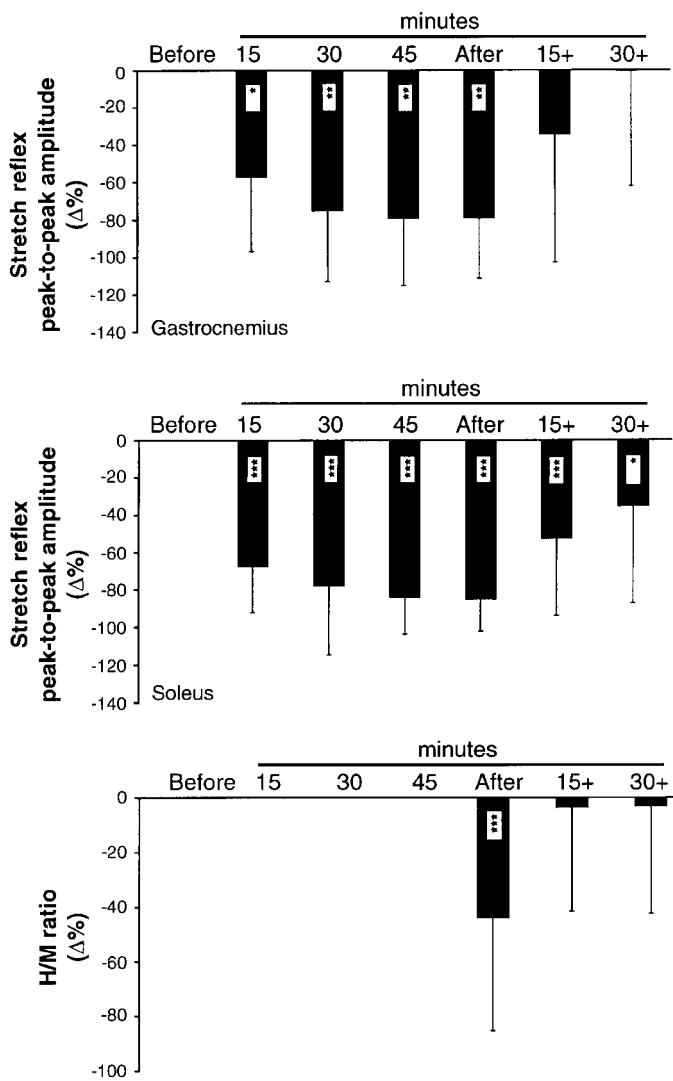


Fig. 4. Relative post-RPS changes (Δ) for experimental leg in stretch reflex EMG peak-to-peak amplitude of gastrocnemius (*top*) and soleus (*middle*) muscles and ratio of electrically induced maximal Hoffmann reflex to maximal mass compound action potential (H/M ratio; *bottom*; mean and SD). H reflex was not measured during RPS. Statistically significant difference compared with before-RPS condition ($n = 20$): * $P < 0.05$, ** $P < 0.01$, and *** $P < 0.001$.

the relationship between the ZCR and motor unit action potentials is linear for low- and constant-level contractions. Therefore, it could possibly be suggested that there was a reduction in the motor unit firing rates due to the RPS. Thus the possibility of increased synchronization of the motor unit firing, which could also be seen as a reduction in the ZCR, seemed to be a less plausible explanation, because there was no increase in the EMG power spectrum, especially in its low-frequency range (4), as shown by an only $1.1 \pm 1.8\%$ (nonsignificant) reduction in median frequency immediately after RPS. According to Hägg (19), a decrease in the firing frequency has correspondingly little effect on the median frequency. The recovery of the ZCR was complete 15 min after the RPS.

H-reflex recovery ($n = 6$). H-reflex peak-to-peak amplitude recovered almost completely within 4 min and

then leveled off to its post-RPS value (Fig. 6). At least part of this incomplete recovery in the post-RPS value could be explained by the changes in the general alertness of the subject, as was proposed earlier.

Effect of 3-min ischemia ($n = 6$). The results showed clearly that none of the mean values of the measured parameters showed any significant changes because of ischemia. Figure 7, which shows the plotted values and correlation coefficients for the selected parameters, demonstrates in all cases good reproducibility. Most importantly, the unaffected maximal H/M ratio implies that the Ia afferent activity was not altered by the ischemia. Thus it can be suggested that the 3-min ischemia was unimportant as a factor in the RPS measurements.

DISCUSSION

This study demonstrated that RPS of a muscle can cause considerable impairment of its force output. The mean reduction was 23.2%, and this was clearly higher than the 13% reduction reported by Lieber et al. (25) for the rabbit tibialis anterior muscle. In their study, the maximal tension was induced by electrical stimulation (100 Hz) of the isolated peroneal nerve. They proposed, therefore, that the origin of the force deficit could be impaired force transfer from the muscle fibers to the tendon. This could be caused by stretch-induced damage in the portions of the myotendinous junction, a location that has been shown to be susceptible to acute injury because of stress concentration at the ends of the tapered muscle fibers (16). However, they did not observe any abnormalities within the muscle fibers. From the viewpoint of their study, the other likely explanations for the reduction in force output because of passive stretching could be either a failure in excitation-contraction coupling or weakened neuromuscular propagation. A possible failure in excitation-contraction coupling can be divided into several phases. Most of them are related to changes in Ca^{2+} metabolism inside the muscle fiber. Armstrong et al. (1) found that static stretching of an isolated rat soleus muscle caused an elevation in muscle Ca^{2+} concentration via Ca^{2+} influx from the extracellular space. This was associated with a reduction in the ability of the muscles to produce force. Despite the difference between the applied passive stretches, this mechanism cannot be disregarded in the present RPS condition. The possibility of weakened neuromuscular propagation is excluded by the nonsignificant changes in the maximal M wave in the present study.

According to Lieber et al. (25), 13% of the reduction in the force output in a similar condition might be because of a failure in the contractile properties of the muscle or in force transfer from the muscle fibers to the tendon. This mechanism does not, however, cover the whole loss of force observed in the present study but leaves part of the force impairment to be explained by some other mechanisms. As our aEMG and ZCR values suggest, these other mechanisms seem to be related to decreased neural drive to the muscle. The reduced neural input could imply the occurrence of central fatigue (12),

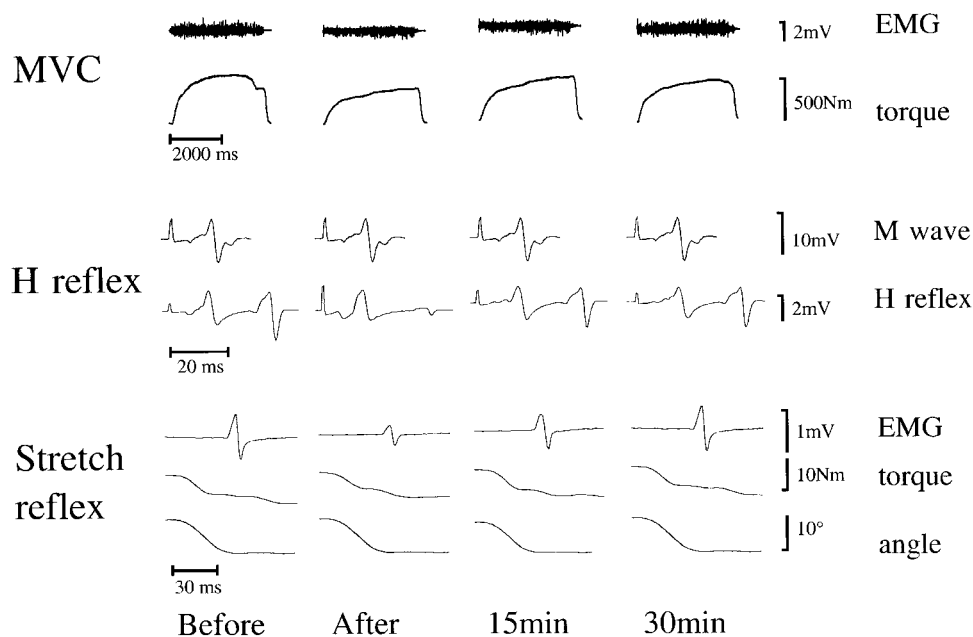


Fig. 5. Data from 1 subject illustrating effectiveness of RPS and subsequent recovery. All signals are from experimental leg. *Top:* soleus EMG and isometric plantar flexion torque from MVC. *Middle:* maximal M waves and H reflexes. *Bottom:* soleus EMG, plantar flexion torque, and ankle angle displacement from stretch reflex recordings.

which can be caused either by supraspinal fatigue (7) or by changes in the inhibitory as well as disfacilitatory signals originating from the contracting muscle (3, 18).

The weakness of the present study was that the effect of possible supraspinal fatigue on the changes in the central drive was not measured. However, we believe that if such an effect could have occurred, it would also have appeared in the contralateral side (control leg), which was not the case, as demonstrated by the nonsignificant changes in the MVC of that side. Thus it would be difficult to explain how a condition in which the muscles are not activated could induce supraspinal fatigue. It seems attractive to suggest that in the RPS condition any central fatigue could be mediated by signals from the involved muscle. The clear reduction in stretch reflex sensitivity and the decreased α -motoneuron pool excitability found in the present study strongly support this hypothesis.

The hypothesis of the peripheral inhibition of the α -motoneuron pool resulting from stimulation of mechanoreceptors and nociceptors (group III and IV) (14, 13) cannot be totally disregarded. These muscle afferents

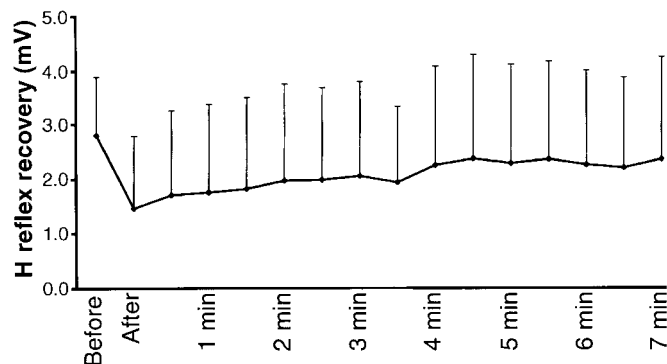


Fig. 6. Mean recovery curve of the H-reflex peak-to-peak amplitude. Values are means \pm SD; $n = 6$ subjects. No statistical significances were tested because of a small sample size ($n = 6$).

have been found to be polymodal, being sensitive to several parameters associated with either metabolic fatigue or chemicals released because of muscle damage (23, 31). However, in the RPS condition it seems to be very difficult to identify the agent that could trigger the discharge of both group III and IV muscle afferents. Our results (blood lactate and serum CK), as well as the literature (1, 25), do not support the occurrence of metabolic fatigue or muscle damage due to RPS. In addition, the rather fast recovery of the neuromuscular parameters favors this conclusion.

If the theory of the presynaptic inhibition of the α -motoneuron pool via the small muscle afferents is less plausible, the possibility of disfacilitation due to reduced Ia afferent activity must be discussed. In several studies of sustained MVC, muscle fatigue has been associated with a decreased inflow of autogenetic excitatory impulses mediated to the α -motoneurons via the γ -loop (6, 27), a phenomenon that also results in reduced reflex sensitivity. However, the exact mechanism inducing the reduced Ia afferent activity has not yet been thoroughly explained. Two major possibilities have been presented: 1) withdrawal of the fusimotor support to the muscle spindles and/or 2) intrafusal fiber fatigue itself (18, 6). Bongiovanni and Hagbarth (6) induced a reduction in MVC motor unit firing rates by a partial anesthetic block of the deep peroneal nerve, which they could counteract by muscle vibration. This could be taken as evidence for the reduced fusimotor role. However, in active muscle fatigue it seems very difficult to separate the pure function of the fusimotor system from that of the muscle spindles. Therefore, in the present study we tried to isolate the pure effect of the muscle spindles by inducing muscle fatigue passively. This was based on the presumption that, although intrafusal fibers are stimulated only by external stretching force, Ia afferent activity is induced without assistance from the fusimotor system. How-

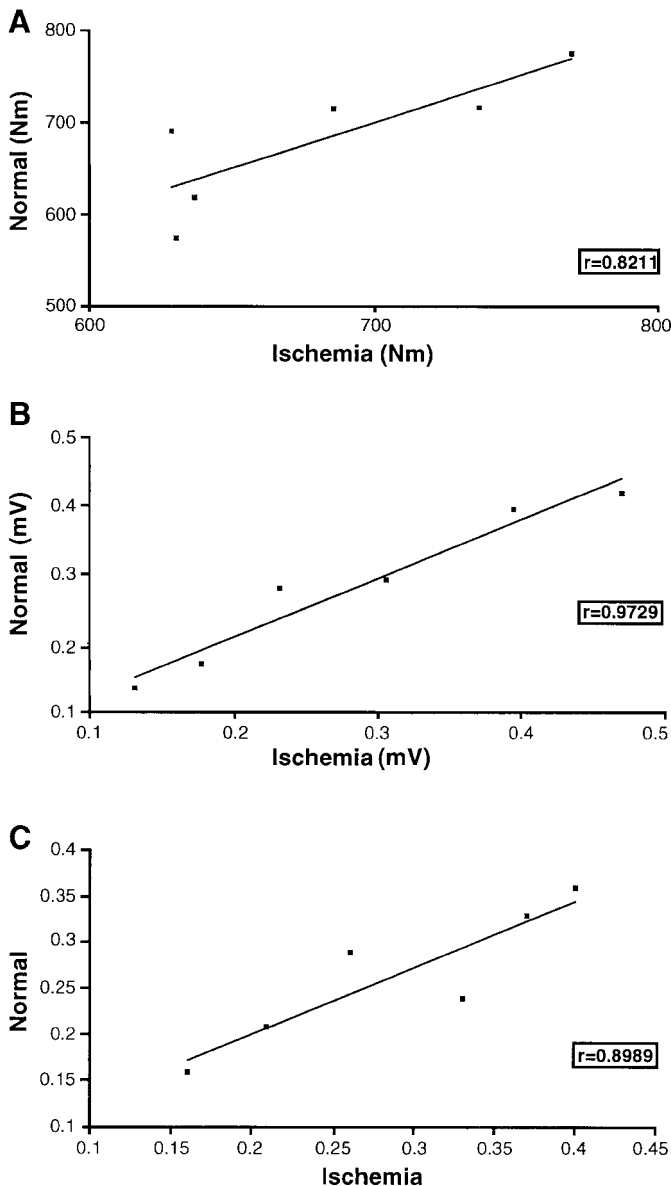


Fig. 7. Similarity between individual parameters before (normal) and at end of 3-min ischemia. *A*: maximal plantar flexion torque on MVC. *B*: maximal soleus aEMG on MVC. *C*: maximal H/M ratio ($n = 6$).

ever, the purpose was not to try to disregard the possible role of the withdrawal of fusimotor support but rather to reveal some more direct effects on the muscle spindle itself.

The possibility that metabolic fatigue processes occur not only in extrafusal but also in intrafusal muscle fibers has not been well demonstrated. Some signs of intrafusal fiber fatigue have been observed after prolonged stimulation of static γ -axons in cats (9) or during the prolonged swimming of mice (35). Such an explanation would be ideal for the results of the present study, especially if signs of metabolic fatigue had been demonstrated. However, this was not the case.

The theory of intrafusal fatigue relies on a fatigue-induced decline in intrafusal contraction force, which reduces the afferent discharge. In the present study the

incompleteness of the explanation regarding metabolic fatigue raises the question of whether the reduced intrafusal contraction force could be induced by mechanical factors. In the study of repeated passive stretch of the rabbit tibialis anterior muscle, Lieber et al. (25) measured the peak force that passively resisted the muscle stretch. This force declined by 19.5% after 30 min of stretches. In the present study, the passive stretch-resisting force was measured differently. We analyzed the average plantar flexion force for the first 40 ms after the onset of the pedal movement during the stretch reflex tests. This average force was obtained by integrating the force-time curve and then dividing the integral by the integration time. During the 40-ms period, the stretch reflexes are triggered but do not yet contribute to the force. The behavior of the stretch-resisting force was very similar to that of the stretch reflexes, and the reduction after the RPS was $\sim 16\%$ (Fig. 8). These results suggest that RPS of a muscle modifies the muscle tissue so that its compliance increases. This results in an impaired external force response of the muscle to stretch and can lead to a reduced stretch response of the muscle spindle. The resulting decrement in the intrafusal force would then decrease the inflow of autogenetic excitatory impulses mediated to the α -motoneurons via the Ia afferents. It would be attractive to speculate that this mechanical modification could also increase intrafusal fiber compliance. In such a case, passive stretching of a muscle could lead to a direct decrease in intrafusal force. Thus, in the presence of γ -motoneuron activation, the contractile properties of these fibers would also have been impaired, leading to reduced intrafusal force. In both cases, the final result would be disfacilitation of the α -motoneuron pool.

It is interesting that a significant reduction in the H-reflex peak-to-peak amplitude could be seen in a resting condition after repeated and prolonged passive stretching of the muscle. There indeed was a rather high correlation coefficient between the stretch-resisting force and the maximal H/M ratio ($r = 0.70$, $P <$

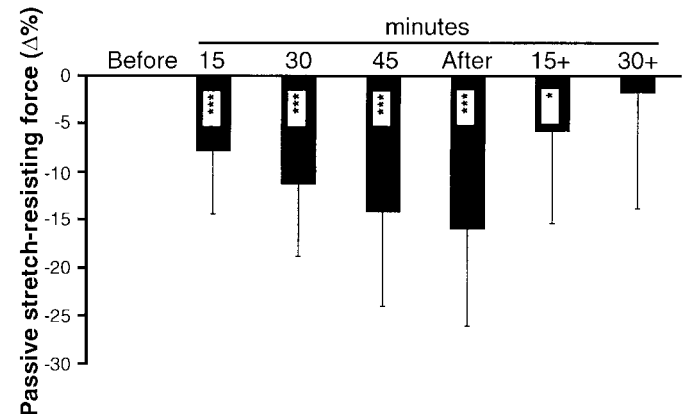


Fig. 8. Relative changes (Δ) resulting from RPS in passive stretch-resisting force (mean and SD) for experimental leg. Force was measured from stretch reflex tests. Statistically significant compared with before-RPS condition: * $P < 0.05$, ** $P < 0.01$, and *** $P < 0.001$ ($n = 20$).

0.01). Therefore, it seems obvious that the increased compliance of the muscle also plays some role in altering the H reflex. In general, the size of the H reflex is affected by the ongoing net excitatory drive onto the α -motoneurons. If the H reflex is depressed in size, then the excitatory drive onto the α -motoneurons has been reduced or the effect of some inhibitory mechanisms has been enhanced. Although, as discussed earlier in this section, the presynaptic inhibition of the Ia-afferent terminals due to stimulation of the group III and IV muscle afferents seems not to be a valid explanation, some other forms of inhibition could be involved. However, we believe that the most likely explanation for the depressed H reflex is a reduction in the excitatory drive from the Ia afferents onto the α -motoneurons, the origin of which is possibly the decreased resting discharge of the muscle spindles because of increased compliance of the muscle.

It would be of interest to discuss the exact origin of this possible modification in muscle tissue. Unfortunately, our results only permit indirect speculations. However, it is most likely that the strain is directed to several elements in the muscle tissue, the total effect depending on the compliance characteristics of the element. Edman and Tsuchiya (11) studied the strain on passive elements during force enhancement by stretch in frog muscle fibers. They suggested that the origin of the elastic elements affected by the stretch is in the longitudinal filaments that link together the Z and M lines. These filaments have been termed titin (also known as connectin) (24) and nebulin (34). The role of titin is of especial interest. Horowitz and Podolsky (21) proposed that titin is responsible for maintaining the central location of the myosin filaments inside a sarcomere in relaxed muscle. Therefore, modification of titin could result in some irregularities of filament overlapping. This could lead to increased compliance of the sarcomere and also to a decrease in the number of attached cross bridges. If this also happens in intrafusal fibers, the direct effect will be reduced intrafusal contraction force. However, in RPS the logical result of the modification of titin is a reduced external force response to stretch and, therefore, a decrease in the mechanical effect on the muscle spindles.

In conclusion, a mechanism to reduce reflex sensitivity, which is known to be present in active muscle fatigue, can also be activated because of repeated and prolonged passive stretching of the muscle. The origin of this system is probably not the small-diameter afferents but rather the reduced activity of the large-diameter ones, resulting from the reduced sensitivity of the muscle spindles to stretch. It is suggested that in this situation of passive stretches the decreased spindle sensitivity is not chemical (metabolic accumulation or deprivation of energy substrate) in nature but mechanical, because of some modification (increased compliance) of the extrafusal and/or intrafusal fibers.

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