Sensitivity of H-Reflexes and Stretch Reflexes to Presynaptic Inhibition in Humans

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1 Physiologisches Institut, Christian-Albrechts Universität zu Kiel, 24098 Kiel, Germany; 2 Department of Medical Physiology, Panum Institute, University of Copenhagen, 2200 Copenhagen N, Denmark; and 3 Centre of Sensory-Motor Interaction, Aalborg University, 9220 Aalborg Ø, Denmark

Morita, H., N. Petersen, L.O.D. Christensen, T. Sinkjær, and J. Nielsen. Sensitivity of H-reflexes and stretch reflexes to presynaptic inhibition in humans. J. Neurophysiol. 80: 610 ± 620, 1998. (Nielsen et al. 1994). It also has been reported that the H-reflex during both co-contraction and walking, it must be concluded that the two reflexes either have a different sensitivity to presynaptic inhibition or that the presynaptic inhibition is counteracted by some other mechanism in the case of the stretch reflex (i.e., increased muscle spindle sensitivity). To clarify this it was the purpose of this study to compare the sensitivity of H-reflexes with that of mechanically evoked reflexes in the soleus muscle with conditioning inputs to soleus motoneurons. A preliminary account of some of the data was presented in abstract form (Morita et al. 1997).

METHODS

The experiments were performed on 17 healthy human subjects aged 20–37 yr. The subjects gave informed written consent to the experimental procedure, which was approved by the local ethics committee. The subjects were seated in a reclining armchair with their left leg attached to a foot plate that could be rotated by a motor (Sinkjær et al. 1988).

Reflex responses

Soleus stretch reflexes were evoked by rotating the foot plate in dorsiflexion direction (5°, 40-ms rise time). The electromyographic (EMG) responses were measured by bipolar Ag-AgCl surface electrodes placed over the soleus muscle. During contraction two distinct reflex responses could be evoked. These were named M1 and M2 according to Toft et al. (1991). At rest only M1 was observed in most subjects; therefore we investigated only this response in this study. The soleus H-reflex was evoked by stimulation (1-ms pulses) of the tibial nerve in the popliteal fossa with the use of a monopolar stimulating electrode. The indifferent electrode was placed above the patella. The soleus T-reflex was evoked by a tendon tap applied to the Achilles tendon with the use of a vibration exciter (model 4809; Bruel and Kjær, Skovlunde, Denmark). The tap had a duration of 4 ms and an amplitude of 0.5–4 mm.

Throughout the experiments the reflexes were adjusted to have similar sizes, usually only 2–5% of the maximal direct M-response (Mmax) because of the small size of the T- and stretch reflexes. The size of the reflexes was measured either as peak-to-peak ampli-
tude or as the area under the full-wave rectified signal. At least 20 reflex responses were averaged for each alternative with interstimulus intervals of 4 s. In some experiments the interstimulus interval was 20 s to avoid the influence of the previous reflex discharge on the measurements (Crone and Nielsen 1989; Hultborn et al. 1996).

The mean ± SD of the responses were calculated on-line. Statistically significant differences between control and conditioned reflexes were determined with the use of a paired Student’s t-test.

**Conditioning stimulation**

The soleus reflexes were conditioned by a biceps femoris tendon tap, by stimulation of the common peroneal nerve, and by stimulation of the femoral nerve. The different latencies of H-, T-, and M1 stretch reflexes were taken into account by delaying or advancing the reflexes so that all had the same latency in relation to the conditioning stimulation.

The stimulation of the common peroneal nerve (CPN) was applied through a bipolar stimulating electrode (1-ms rectangular shocks) placed distal to the neck of the fibula. The intensity of the stimulation was adjusted to 1.0 times motor threshold (1.0 × MT) in the tibialis anterior muscle. Care was taken that motor fibers to the peroneal muscle group were not activated. The femoral nerve stimulation was applied through a monopolar ball electrode placed in the femoral triangle. The indifferent electrode was placed on the back of the thigh. The intensity of the stimulation was adjusted to 1.2 × MT in the quadriceps muscle. The biceps femoris tendon tap was applied by a vibration excitor (Bruel and Kjær model 4809). The duration of the tap was 3–4 ms and the amplitude was 0.5–4 mm. Such a mechanical perturbation may easily spread along the leg and possibly activate soleus Ia afferent (Burke et al. 1983). Therefore it was checked that the tap did not evoke a facilitation of the soleus H-reflex at a latency compatible with elicitation of homonymous monosynaptic excitation.

**Poststimulus time histograms**

Histograms of the probability of discharge of single voluntarily activated soleus motor units were constructed after an Achilles tendon tap or stimulation of the tibial nerve (see Fournier et al. 1986 for a detailed description of the technique). To reduce the number of triggers, the stimuli were triggered on the previous discharge of the motor unit. By changing the delay between the trigger and the stimulation the stimulation could thus always be given at an optimum time, i.e., when the unit was not refractory because of the previous discharge. The poststimulus time histogram (PSTH) was constructed for a window between 20 and 70 ms after the stimuli with bins of 1 ms. A histogram was also constructed in a control situation without stimulation. The spontaneous discharge probability of the unit could thus be subtracted from that resulting from the stimulation. The interval between each measurement was 1 s. The onset of peaks in the histograms was determined by the first bin of a series of bins in which a count of more than three times the spontaneous discharge probability was observed. The end of a peak was determined as the last bin in such a series.

In each experiment six different alternatives were randomly alternated as follows: 1) control situation without any stimulation, 2) tibial nerve stimulation, 3) Achilles tendon tap, 4) tibial nerve stimulation conditioned by stimulation of the CPN or a biceps femoris tendon tap, 5) Achilles tendon tap conditioned by the same as above, and 6) stimulation of the CPN or a biceps femoris tendon tap. Details of the different stimuli were given above. The Achilles tendon tap was advanced by 4–6 ms in relation to the tibial nerve stimulation to take the different latency of the peaks of increased firing probability following the two stimuli into account.

The statistical significance of the peaks in the histograms was determined with the use of a X² test. This test was also used to determine significant changes in the size of the peaks from individual units following the conditioning stimuli, whereas a paired Student’s t-test was used to determine significant changes in pooled data from different units. Because the size of the peaks in the control situation may influence the estimated absolute change in the size of the peaks following the conditioning stimulation, the size of the peaks in the conditioned situation were also expressed as a percentage of their size in the control situation. When pooling data from different units this calculation was first done for each unit individually, and the percentages from each unit were then averaged.

**RESULTS**

**Different sensitivity of stretch reflex, T-reflex, and H-reflex to presynaptic inhibition**

Figure 1A demonstrates a time course of the effect of a tap applied to the biceps femoris tendon (1-mm amplitude, 3-ms duration) on the short-latency stretch reflex (M1: ●), the soleus T-reflex (□), and the soleus H-reflex (○). All three reflexes were adjusted to have the same size in the control situation (≈2–3% of Mmax), and the tendon tap and muscle stretch were advanced in relation to the tibial nerve stimulation by 5 and 12 ms, respectively, to take the longer latency of the T- and M1 stretch reflex in relation to the H-reflex into account. The biceps femoris tendon tap evoked a clear and highly significant depression of the H-reflex at an interval of 40 ms, which lasted for 150 ms. This depression is most likely caused by presynaptic inhibition of the Ia afferents that mediate the reflex (e.g., Nielsen and Petersen 1994). A tendency for a similar depression of the T-reflex was also observed, but it never reached a statistically significant level. No depression of the stretch reflex was seen. Although the three reflexes had a very similar size and shape (see Fig. 1A, inset) they were nevertheless very differently affected by presynaptic inhibition. H- and T-reflexes were compared in 17 subjects (Fig. 1B), and H- and M1 stretch reflexes were compared in 6 subjects (Fig. 1C).

In all experiments the H-reflex was significantly (P < 0.05) more depressed than the M1 stretch reflex and the T-reflex at an interval of 70 ms after the biceps femoris tendon tap, taking the different latencies of the reflexes into account. At this interval the H-reflex was on average depressed to 48.4 ± 22.5% (SD) of the control reflex size (P < 0.001), whereas the M1 stretch reflex was not depressed at all (100.5 ± 8.3%; P > 0.1). The T-reflex was depressed to 92.8 ± 7.6% of the control reflex size, which was a statistically significant change (P < 0.05). The H-reflex was thus significantly more depressed than the other two reflexes (P < 0.001). On average the H-reflex lasted 15.2 ± 2.9 ms, the T-reflex 16.1 ± 3.6 ms, and the M1 stretch reflex 23.2 ± 5.5 ms. The reflexes had average latencies of 32.7 ± 2.4, 38.2 ± 2.6, and 45.8 ± 1.5 ms, respectively. In three subjects it was ensured that a similar difference in the amount of inhibition was observed when the reflexes were evoked every 20 s instead of the usual 4 s.

In 14 experiments on 12 subjects the effect of a conditioning stimulation of the CPN on the H- and T-reflexes was compared (Fig. 2). The time course of the depression of the H- and T-reflexes in a single subject is shown in Fig. 2A. At a conditioning-test interval of 1–3 ms the CPN stimulation...
FIG. 1. Effect of a biceps femoris tendon tap on the H-, T-, and M1 stretch reflex. A: time course of the effect of a biceps femoris tendon tap (1-mm amplitude, 2-ms duration) on the soleus H- (○), T- (△), and M1 stretch reflex (●) in a single subject. The reflexes were adjusted to have the same size (2–3% of maximal direct M-response, $M_{\text{max}}$) in the control situation. The T- and M1 stretch reflex were advanced by 5 and 12 ms in relation to the H-reflex to take their longer latency into account. The abscissa is the interval between the conditioning biceps femoris tendon tap and the test stimulation evoking each of the 3 reflexes. The ordinate is the size of the conditioned reflex as a percentage of the control reflex size. Each bar is 1 SE. Sample traces (average of 5 sweeps) of each of the 3 reflexes are shown as an inset above the graph.

B and C: comparison of the depression of the H- and T-reflex and H- and M1 stretch reflex, respectively, in all tested subjects at an interval of 70 ms after the conditioning biceps femoris tendon tap, taking the different latencies of the reflexes into account. In all experiments the control reflexes were adjusted to have the same size ($\approx 2$–5% of $M_{\text{max}}$). The inhibition is expressed as the size of the conditioned reflex in percent of the control reflex size in all cases. In 17 subjects the H- and T-reflex and in 6 subjects the H- and M1 stretch reflex were compared. The control reflexes were in general smaller in the six subjects in whom H- and M1 stretch reflexes were compared than in the subjects in whom H- and T-reflexes were compared. Because small reflexes will appear to be more inhibited than larger reflexes (Crone et al. 1985), the H-reflex on average was more inhibited in C than in B. Each symbol and line represent 1 subject.

evoked a depression of the H-reflex, which has been demonstrated to be caused by disynaptic reciprocal Ia inhibition (Crone et al. 1987; Tanaka 1974). At conditioning test intervals $>10$ ms a second period of inhibition was observed. This inhibition was termed D1 by Mizuno et al. (1971) and is likely caused by presynaptic inhibition of the Ia afferents, which mediate the soleus H-reflex (Capaday et al. 1995; Faist et al. 1996). Although the T-reflex was as depressed as the H-reflex at the interval of the disynaptic reciprocal Ia inhibition, it was not significantly depressed at the interval of the D1 inhibition.

It can be seen from Fig. 2B that a clear short-latency inhibition of the T- and H-reflexes could be evoked in 7 of the 14 subjects (at a conditioning test interval of 2.0 ms the H-reflex was on average inhibited to $71.7 \pm 10.0\%$, whereas the T-reflex was inhibited to $66.7 \pm 12.5\%$). The small difference in the size of this depression of the two reflexes was not significant ($P = 0.07$). In contrast, at the latency of D1 (Fig. 2C; conditioning test interval 15 ms) the H-reflex was significantly more depressed than the T-reflex (depression of H-reflex to $62.9 \pm 14.3\%$ as compared with $91.4 \pm 13.4\%$ for the T-reflex; $P < 0.01$). In the remaining five subjects it was not possible to evoke a short-latency inhibition of either the H- or the T-reflex. However, in these subjects a significantly larger depression of H-reflex than of T-reflex at the D1 latency was also observed ($P < 0.01$). In two experiments the M1 stretch reflex and the soleus H-reflex were compared in the same way. In both experiments the two reflexes were equally depressed at the latency of the disynaptic inhibition, although only the H-reflex was depressed at the D1 latency. Thus it appears that the three reflexes have a different sensitivity to presynaptic but not to postsynaptic inhibition.

It generally was not possible to obtain T- and M1 reflexes >5% of $M_{\text{max}}$. However, in three subjects T-reflexes as large as 15% of $M_{\text{max}}$ were obtained. In these subjects the H-reflex...
FIG. 2. The effect of a common peroneal nerve (CPN) stimulation on the H- and T-reflex. A: time course of the effect of a CPN stimulation [1.0 × motor threshold (MT); single shock] on the H- (○) and T-reflex (●) in a single subject. The reflexes were adjusted to have the same size (∼2–3% of M_max) in the control situation. The T-reflex was advanced by 6 ms in relation to the H-reflex to take its longer latency into account. The abscissa is the interval between the conditioning CPN stimulation and the tibial nerve stimulation or the Achilles tendon tap. The ordinate is the size of the conditioned reflex as a percentage of the control reflex size. Each bar is 1 SE.

B and C: comparison of the depression of the H- and T-reflex for reciprocal Ia inhibition (B) and D1 (C), respectively, in 7 subjects who showed both reciprocal Ia inhibition and D1. In all experiments the control reflexes were adjusted to have the same size (∼5% of M_max). The inhibition is expressed as the size of the conditioned reflex in percent of the control reflex size in all cases. Each symbol and line represent one subject.

was always found to be significantly more depressed than the T-reflex by the CPN stimulation at the latency of D1 regardless of the size of the control reflex.

Sensitivity of H-reflex and T-reflex to heteronymous Ia facilitation

In five experiments on four subjects the effect of stimulation of the femoral nerve (1.2 × MT) on H- and T-reflexes was compared. Data from one of these experiments are shown in Fig. 3. As can be seen from the figure, the femoral nerve stimulation induced a clear facilitation of both reflexes at a short latency (∼7.5 ms; the negative conditioning test interval designates that the test stimulation of the tibial nerve was applied before the conditioning stimulation of the femoral nerve). This facilitation has been shown to be caused by monosynaptic facilitation of soleus motoneurons from Ia afferents in the femoral nerve at least within the initial 0.5 ms after the onset of the facilitation (Hultborn et al. 1987).

There was no statistically significant difference in the amount of the facilitation of the two reflexes at any of the conditioning test intervals at which the facilitation was seen in this subject (from −7.5 to −6.0 ms; P > 0.1). There was also no statistically significant difference in the amount of facilitation of the two reflexes in any of the other subjects (measured within the initial 0.5 ms after the onset of the facilitation; P > 0.1).

This demonstrates that H- and T-reflexes are not only similarly sensitive to postsynaptic inhibition from the CPN, but also to postsynaptic excitation from the femoral nerve. They thus seem to differ only in their sensitivity to presynaptic inhibition.

Single unit experiments

PSTHs of the discharge of single voluntarily activated soleus motor units were constructed after subthreshold electrical stimulation of the tibial nerve and subthreshold Achilles tendon tap. Both stimuli elicited clear peaks in the PSTH of the motor units, an example of which is shown in Fig. 4, A and D. The latency of the peak induced by the tibial nerve stimulation was 42 ms, whereas that of the peak induced by the Achilles tendon tap was 46 ms. However, the Achilles tendon tap was advanced by 4 ms, so that the peaks occurred in the histograms with the same latency. The peak evoked by the Achilles tendon tap lasted 5 ms, whereas the peak induced by the electrical nerve stimulation lasted 3 ms. The peak evoked by the electrical stimulation was very strongly depressed when it was preceded by a biceps femoris tendon conditioning test interval of 60 ms (Fig. 4B). In contrast to this the peak evoked by the Achilles tendon tap was only weakly depressed (Fig. 4E). This was visualized in Fig. 4, C and F, by subtracting from each other the histograms in Fig. 4, A and B and D and E, respectively. When comparing the
first bins of the mechanically induced peak (marked by dotted vertical lines) to the last bins (marked by dashed vertical lines), it is seen that only the first part of the peak was depressed. The last part of the peak even tended to be facilitated. The biceps femoris tendon tap had no effect on the discharge probability of the motor unit at the latency of the peaks induced by the tibial nerve stimulation and Achilles tendon tap (Fig. 4G).

This experiment was performed in 53 motor units from 10 subjects. In these 53 motor units the electrically evoked peaks lasted on average 2.7 ms, whereas the mechanically evoked peaks lasted 5.0 ms. For all units the difference in the size of the peaks in the control and conditioned situations was visualized by subtracting the histograms in the control and conditioned situations from each other, thereby obtaining a new histogram as shown in Fig. 4, C and F. The difference between the peaks could then be calculated as the number of counts in the peaks or troughs in this new histogram (i.e., number of counts between the dotted and dashed vertical lines in Fig. 4, C and F). The results of this calculation in each unit are shown in Fig. 5A for the electrically evoked peaks and in Fig. 5B for the mechanically evoked peaks. On average the biceps femoris tendon tap depressed the peaks induced by the tibial nerve stimulation by 13.1 counts per 100 triggers, whereas the peaks induced by the Achilles tendon tap were depressed by 10.0 counts per 100 triggers. This difference was statistically significant ($P < 0.01$). The size of the control peaks was very similar (30 counts per 100 triggers for the electrically evoked peak and 33 counts per 100 triggers for the mechanically evoked peak). When expressing the size of the conditioned peak as a percentage of the size in the control peak for each individual unit, it was found that the biceps femoris tendon tap on average depressed the tibial nerve–evoked peak to $55 \pm 27\%$, whereas the peak evoked by the Achilles tendon tap was depressed to $76 \pm 53\%$.

When making a bin-to-bin analysis of the effect of the biceps femoris tendon tap on the mechanically induced peaks, it was found that the last bins in the peak were generally less depressed than the first bins, but the most marked difference was found between the first and last two bins. Therefore Fig. 5, C and D, demonstrates the effect of the biceps femoris tendon tap on the first (Fig. 5C) and last (Fig. 5D) two bins of the mechanically induced peaks. When pooling units from each of the 10 subjects individually, the last two bins of the mechanically evoked peaks were found to be significantly less depressed than the first two bins in all subjects ($P < 0.05$). On average the biceps femoris tendon tap depressed the first two bins of the mechanically induced peaks by 5.0 counts per 100 triggers, whereas the last two bins were depressed by only 2.7 counts per 100 triggers. This difference was statistically significant ($P < 0.001$). The number of counts within the first two bins of the control peaks was very similar to the number of counts in the last two bins (13.8 vs. 11.8 counts). When expressing the size of the bins in the conditioned peak as a percentage of their size in the control peak for each individual unit, it was found that the initial two bins of the peaks were depressed to 69 ± 48%, whereas the last two bins were not depressed (99 ± 92%). In contrast to this, all bins of the electrically induced peaks were found to be equally depressed.

Figures 6 and 7 demonstrate that essentially the same observations were made when presynaptic inhibition was evoked by stimulating the CPN. The CPN stimulation was advanced with respect to the test tibial nerve stimulation and the test Achilles tendon tap so that the period of D1 occurred at the same time as the peaks induced by the two test stimuli (in the illustrated example a delay of 60 ms was used). As can be seen from Fig. 6, A–C and D–F, the CPN stimulation depressed all parts of the electrically induced peak equally, whereas only the initial part of the mechanically induced peak was depressed. As seen from Fig. 7A, a depression of the electrically induced peaks was seen for all 34 motor units from seven subjects studied in this way (average decrease of counts: 9.6 per 100 triggers; the size of the conditioned peak was 65 ± 27% of the control peak; the size of the control peak was 30 counts
FIG. 4. Effect of biceps femoris tendon tap on peaks evoked by tibial nerve stimulation and Achilles tendon tap in the poststimulus time histogram of a single voluntarily activated soleus motor unit. A and D show the histograms after stimulation of the tibial nerve and an Achilles tendon tap, respectively. The Achilles tendon tap was advanced in relation to the tibial nerve by 6 ms so that the peaks induced by the 2 stimuli occurred at the same time. In B and E a tendon tap was applied to the biceps femoris tendon (3-ms duration, 1-mm amplitude) 60 ms before the 2 test stimuli. C and F demonstrate the histograms that were obtained by subtracting the histograms in B from A and in E from D, respectively. In G is shown the effect of the biceps femoris tendon tap on the discharge probability of the motor unit when it was applied separately. The dotted vertical lines mark the onset of peak, whereas the dashed vertical lines mark the end of peak. The ordinate is the number of counts expressed as a percentage of the total number of triggers. The abscissa is the time after the test stimulation in milliseconds. The number of triggers per alternative was 100.

per 100 triggers). The mechanically induced peaks were significantly ($P < 0.01$) less depressed (Fig. 7B; average decrease of counts: 6.3 per 100 triggers; the size of the conditioned peak was $83.3 \pm 28\%$ of the control peak; the size of the control peak was 37 counts per 100 triggers), and when making a bin-to-bin analysis of the peaks it was found that this was mainly explained by a lack of depression of the last two bins of the peaks (Fig. 7D; average increase of counts = 2.2 per 100 triggers), whereas the first two bins were strongly depressed (Fig. 7C; average decrease of counts = 9.8 per 100 triggers). This difference was statistically significant ($P < 0.001$). When expressed in percent for each unit individually, the two first bins in the conditioned peak were on average found to be $85 \pm 24\%$ of their size without conditioning, whereas the last two bins were facilitated to $133 \pm 88\%$. The number of counts in the first and last two bins of the control peaks were 23 and 14, respectively. The difference between the first and last two bins of the peaks was also statistically significant in all subjects when pooling data from each subject individually ($P < 0.05$). It was ensured in all motor units that the biceps femoris tendon tap when applied separately had no effect on the discharge probability of the motor unit at the latency at which the peaks induced by the tibial nerve stimulation and Achilles tendon tap were measured.

**DISCUSSION**

It was demonstrated that electrically evoked H-reflexes are more sensitive to presynaptic inhibition than mechanically evoked T- and M1 stretch reflexes. However, the three types of reflexes were equally sensitive to postsynaptic inhibition and facilitation.

This finding provides a possible explanation for the discrepancy in the modulation of H- and stretch reflexes, which was reported in previous studies. Nielsen and Kagamihara (1993) thus described that H-reflexes are strongly depressed during co-contraction compared with an isolated plantar flexion at a matched background EMG-level, and they found that this was caused by an increase of presynaptic inhibition of the Ia afferents that mediate the reflex. Surprisingly in a later study short-latency stretch reflexes turned out not to be as depressed as the H-reflexes during the co-contraction task (Nielsen et al. 1994). Similarly, Sinkjaer et al. (1996) found that short-latency stretch reflexes are not depressed in the stance phase of walking compared with a tonic soleus contraction, although H-reflexes are strongly depressed (Ca-
afferents by either the biceps femoris tendon tap or the CPN neurons different from that in the electrically evoked reflex, were explained by a lower recruitment gain (Kernell and Hultborn 1990) or by recruitment of a population of α-motor axons, which through collaterals may lead to activation of Renshaw cells. Long-lasting changes in the muscle spindle sensitivity such as those described by Gregory et al. (1990) after muscle length changes are also not likely to explain our observations. We took precautions to ensure that the soleus muscle was not activated by any of the conditioning stimuli and it was checked that similar observations were made whether the test reflexes were evoked at short (every 3–4 s) or long (every 15 s) intervals.

The reason for the different sensitivity of the different reflexes is more likely to be found in the different afferent volleys evoked by the three stimuli. The electrical nerve stimulation activates the Ia afferents almost simultaneously and therefore elicits a volley that is only little dispersed in time on its arrival at the spinal cord (Burke et al. 1983). After a tendon tap and especially a slow muscle stretch the Ia afferents are on the other hand activated at very different times, and the same Ia afferents may discharge several times. The volleys are therefore considerably dispersed in time on their arrival at the spinal cord (Burke et al. 1983). This distinction was also clear from the different duration of the peaks in the PSTH evoked by the electrical and mechanical stimuli. As noted also by Burke et al. (1984) the filtering properties of the motoneurons nevertheless ensure that the reflexes have almost the same shape, size, and duration (cf. Fig. 1A, inset). Only the M1 stretch reflex had a significantly longer duration than the other two reflexes, whereas the H- and T-reflexes had almost the same shape, size, and duration, although the peaks in the PSTH evoked by the tendon tap lasted almost twice as long as the peaks evoked by the electrical stimulation.

The afferent volleys evoked by the three stimuli likely also differ in the contribution from other afferents than Ia afferents. Both the electrical and mechanical stimulation may activate group Ib, group II, and cutaneous afferents and the electrical stimulation may additionally activate efferent α-motor axons, which through collaterals may lead to activation of Renshaw cells.

The question is then how this different composition and shape of the afferent volleys can explain the different sensitivity to presynaptic inhibition of the evoked reflexes. It seems evident that postsynaptic mechanisms can be rejected. If the lower sensitivity of the mechanically evoked reflexes were explained by a lower recruitment gain (Kernell and Hultborn 1990) or by recruitment of a population of motoneurons different from that in the electrically evoked reflex, the reflexes would also have been expected to have a different sensitivity to postsynaptic effects. However, we found that postsynaptic inhibition and facilitation were equally pronounced for the three reflexes. Furthermore, it has been demonstrated in several studies that electrical stimulation of Ia afferents and a muscle stretch both recruit the motoneurons according to Hennemann’s size principle (Burke 1981). Finally, we were able to demonstrate a difference in the sensi-

Why are mechanically evoked reflexes less sensitive to presynaptic inhibition than electrically evoked reflexes?

There is a remote possibility that the stimulation of Ia afferents by either the biceps femoris tendon tap or the CPN activated soleus γ-motoneurons, thereby changing the sensitivity of the muscle spindles to the soleus muscle stretch or the Achilles tendon tap, it could be that this effect then counteracted the depression of transmission in the Ia afferent synapses caused by presynaptic inhibition. However, it has been found that group Ia afferents have either very weak or no effects on γ-motoneurons in the cat hind limb (Appelberg et al. 1983). The difference in the depression of the electrically and mechanically evoked reflexes could furthermore be seen for >100 ms following the biceps femoris tendon tap. It is unlikely that the effects on the γ-motoneurons would have such a long duration. Finally, it seems difficult to explain the differential effect of the conditioning biceps tendon tap and CPN stimulation on the first and last part of the mechanically evoked peaks in the PSTH by an effect on γ-motoneurons.

Long-lasting changes in the muscle spindle sensitivity such as those described by Gregory et al. (1990) after muscle length changes are also not likely to explain our observations. We took precautions to ensure that the soleus muscle was not activated by any of the conditioning stimuli and it was checked that similar observations were made whether the test reflexes were evoked at short (every 3–4 s) or long (every 15 s) intervals.

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FIG. 5. Summary of the effect of biceps femoris tendon tap on peaks evoked by tibial nerve stimulation and Achilles tendon tap in the poststimulus time histogram from all investigated motor units. Each line and dot represent data from a single motor unit. A total number of 53 motor units from 10 subjects were studied. In A and B the difference between the total number of counts contained in the electrically evoked (A) and mechanically evoked peaks (B) with and without the conditioning biceps femoris tendon tap is shown. In C and D the same calculation is shown for the first (C) and last 2 ms (D) of the mechanically evoked peaks. In all experiments the Achilles tendon tap was advanced by 4–6 ms in relation to the tibial nerve stimulation so that the peaks evoked by the 2 stimuli occurred at the same time.

Why are mechanically evoked reflexes less sensitive to presynaptic inhibition than electrically evoked reflexes?

There is a remote possibility that the stimulation of Ia afferents by either the biceps femoris tendon tap or the CPN activated soleus γ-motoneurons, thereby changing the sensitivity of the muscle spindles to the soleus muscle stretch or the Achilles tendon tap, it could be that this effect then counteracted the depression of transmission in the Ia afferent synapses caused by presynaptic inhibition. However, it has been found that group Ia afferents have either very weak or no effects on γ-motoneurons in the cat hind limb (Appelberg et al. 1983). The difference in the depression of the electrically and mechanically evoked reflexes could furthermore be seen for >100 ms following the biceps femoris tendon tap. It is unlikely that the effects on the γ-motoneurons would have such a long duration. Finally, it seems difficult to explain the differential effect of the conditioning biceps tendon tap and CPN stimulation on the first and last part of the mechanically evoked peaks in the PSTH by an effect on γ-motoneurons.
tivity to presynaptic inhibition also for the peaks induced in the PSTH by the mechanical and electrical stimuli. The peaks in the PSTH were shown to reflect a combination of the first derivative of the underlying excitatory postsynaptic potential (EPSP) and the EPSP itself (Fetz and Gustafsson 1983; Gustafsson and McCrea 1984; Kirkwood and Sears 1982). A decrease in the size of the EPSP caused by presynaptic inhibition is therefore reflected as a decrease in the size of the PSTH peaks. As the mechanically evoked peaks were less depressed than the electrically evoked PSTH peaks, this must reflect that presynaptic inhibition caused a smaller decrease in the EPSP induced by the mechanical stimulus than in the EPSP induced by the electrical stimulus. This contrast suggests that the reason for the different sensitivity of the reflexes to presynaptic inhibition is to be found at a presynaptic rather than postsynaptic level.

The possibility that the different sensitivity of the reflexes is caused directly by the different temporal dispersion of the underlying afferent volleys is also made less likely by the same arguments. A proper timing between the test and conditioning stimulation must be assumed to be much more important for the short-lasting disynaptic reciprocal inhibition or heteronymous monosynaptic facilitation than for the much longer-lasting presynaptic inhibition. Therefore the weaker synchronization of the afferent volley underlying the mechanically evoked reflexes should have been expected to make the demonstration of the short-lasting postsynaptic inhibition more difficult than when the H-reflex was used as a test, but this was not the case.

The question is then which mechanism located at a presynaptic level may explain the different sensitivity of the reflexes. When analyzing the PSTH peaks we found pronounced differences in the depression of the first and the last part of the peaks evoked by the Achilles tendon tap. Whereas the first part of the peaks (initial 2 ms) was strongly depressed, the later part (last 2 ms) was only weakly depressed or not depressed at all. If it is accepted that the first part of the PSTH peaks reflects the initial part of the underlying EPSP, whereas the later part reflects later parts of the EPSP, this signifies that only the very initial part of the EPSP evoked by the Achilles tendon tap was depressed by presynaptic inhibition, whereas all of the EPSP evoked by electrical stimulation was equally depressed.

Two alternative explanations may be provided to explain why the later parts of the mechanically evoked PSTH peaks are less depressed than the earlier part. First, it may be that the EPSPs evoked by the mechanical stimuli are more contaminated by nonmonosynaptic effects than the EPSPs

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**Fig. 6.** Effect of CPN stimulation on peaks evoked by tibial nerve stimulation and Achilles tendon tap in the poststimulus time histogram of a single voluntarily activated soleus motor unit. The figure is arranged in the same way as Fig. 4 except that the conditioning stimulation was applied to the CPN (1.1 × MT) at an interval of 60 ms before the test stimulation. The Achilles tendon tap was advanced in relation to the tibial nerve stimulation by 6 ms. The total number of triggers per alternative was 100.
Furthermore, even if the synapses on the interneurons are based on observations of the modulation of H-reflexes should synaptic inhibition, but if not this might also contribute to conclusions regarding the modulation and functional significance of stretch reflexes during voluntary movement based on observations of the modulation of H-reflexes should

Figure 7. Summary of the effect of CPN stimulation on peaks evoked by tibial nerve stimulation and Achilles tendon tap in the poststimulus time histogram from all investigated motor units. A total number of 34 motor units from 7 subjects were studied. The arrangement is the same as in Fig. 4 except that the conditioning stimulation was applied to the CPN. The interval between the conditioning and test stimulation used for each subject was determined from a time course of the effect of the CPN on the soleus H-reflex. The interval between the largest inhibition of the H-reflex within the period 15–80 ms after the CPN stimulation was chosen. In most subjects an interval of 15–20 ms was used, but in 2 subjects no or only weak inhibition was observed at this interval, and an interval of 60 ms was therefore used instead. The subject used for the illustration in Fig. 5 was one of these subjects. In all experiments the Achilles tendon tap was advanced by 4–6 ms in relation to the tibial nerve stimulation so that the peaks evoked by the 2 stimuli occurred at the same time. On average the electrically evoked peaks lasted 2.5 ms, whereas the mechanically evoked peaks lasted 4.9 ms.

evoked by the electrical stimulation (Burke et al. 1983, 1984) and the two types of stimuli may activate different afferents (such as group Ib, group II, and cutaneous afferents) to a different extent. Both group Ia and group II afferents are known to have nonmonosynaptic excitatory projections to homonymous motoneurons (Cavallari et al. 1987; Jankowska et al. 1981a,b). Such projections would contribute mainly to the later parts of the EPSPs underlying the PSTH peaks, and because group II afferents are not subjected to presynaptic inhibition from group Ia afferents (Baldissera et al. 1981) this might explain the lack of depression of the later part of the EPSPs underlying the PSTH peaks. It is unknown whether the terminals of Ia afferents on interneurons in the oligosynaptic Ia pathways are subjected to presynaptic inhibition, but if not this might also contribute to the lack of depression of the last part of the PSTH peaks. Furthermore, even if the synapses on the interneurons are presynaptically inhibited, the transmission through several interneuronal relays is likely to mask the effect of presynaptic inhibition of the input to the first order interneurons.

The second possible explanation of the differential effect of presynaptic inhibition on the first and last part of the PSTH peaks comes from the observation that the tendon tap evokes a burst of discharges in the Ia afferents, whereas the electrical stimulation evokes a single synchronized discharge from a large number of Ia afferents. Therefore the EPSP evoked by the electrical stimulation is caused mainly by spatial summation of EPSPs from a relatively large number of Ia afferents, whereas the EPSP evoked by the tendon tap is evoked to a larger extent by temporal summation in a relatively low number of Ia afferents. Previous activation of a synapse is known to influence the probability of transmitter release from the synapse (Mendell 1984). Within the initial ~50 ms after a previous activation a facilitation of transmitter release is generally seen, whereas a long-lasting depression is seen at longer intervals. It has been demonstrated that during high-frequency stimulation the facilitation is increased and the depression is decreased when the transmitter output from the synapse is reduced by changing the external Ca²⁺ concentration or applying the γ-aminobutyric acid (GABA) agonist baclofen (Pinco and Lev-Tov 1993; Seebach and Mendell 1996). Stuart and Redman (1991) also demonstrated that the initial facilitation evoked by paired stimulation was greatly increased by presynaptic inhibition. This interaction between presynaptic inhibition and the changes in transmitter release induced by previous activation of the synapse may explain the relatively small depression of the last part of the PSTH peaks evoked by the mechanical stimuli. The first part of these and all the peaks evoked by the electrical stimulus are in contrast evoked by synapses that were not previously activated at this relatively short interval, and presynaptic inhibition of these peaks may therefore easily be demonstrated.

In other words what we propose is that presynaptic inhibition may effectively modulate EPSPs evoked by low-rate Ia afferent discharges, but at higher rates the interaction between the facilitatory effect of the previous discharges of the Ia afferents and presynaptic inhibition will counteract the depression of the EPSPs. Ongoing experiments in the cat may reveal whether this interpretation is correct.

**Methodological implications**

In the original studies by Burke et al. (1983, 1984) the popular comparison of H- and T-reflexes with the aim of elucidating changes in γ-motor activity was questioned because it was argued that the afferent volleys and composite EPSP rise times evoked by electrical and mechanical stimuli were too different and that therefore the two reflexes would be influenced by nonmonosynaptic pathways to a different extent. We have now demonstrated that the concern expressed in these earlier studies was indeed justified. Our data not only confirm that a comparison of electrically and mechanically evoked reflexes cannot be used to deduce changes in muscle spindle sensitivity but also that direct conclusions regarding the modulation and functional significance of stretch reflexes during voluntary movement based on observations of the modulation of H-reflexes should...
be made with some caution (Capaday and Stein 1986, 1987).

The recording of H-reflexes is a valuable tool in investigating and understanding the modulation of central neuronal pathways, including presynaptic inhibition of Ia afferents, but our data stress that it is not straightforward to make a functional interpretation of H-reflex data in terms of stretch reflex behavior. The H-reflex data do demonstrate how presynaptic inhibition and thereby the access of the Ia afferents to the motoneurons is modulated during voluntary movement, but our point is that they say nothing about how this modulation affects the stretch reflex or the afferent stretch reflex volley because the afferent volleys underlying the H- and stretch reflex and the central processing of these volleys are not comparable. When functional conclusions are made, it should be kept in mind that both the application of electrical stimuli to nerves and artificial external perturbations to muscles, as we have used here, are experimental techniques, which only to a certain extent mimic the normal physiological Ia afferent feedback.

Functional significance

Functional interpretations of the findings in this study should therefore also be made with considerable caution only. It should be pointed out that we tested the influence of presynaptic inhibition evoked by a rather synchronized discharge of peripheral afferents, which is not necessarily directly comparable to the modulation of tonically active presynaptic inhibition during different motor tasks. Furthermore, most of the experiments in this study were performed at rest, and it is not certain that they would also apply during natural voluntary motor tasks. We would nevertheless like to suggest the following hypothesis as one possible functional interpretation of our findings. If it is true that presynaptic inhibition is pronounced when the Ia afferent discharge is low, and small when the discharge is high, it might mean that the effect of presynaptic inhibition is automatically adjusted according to the discharge rate of the Ia afferents. This mechanism would ensure that presynaptic inhibition provides an efficient gating mechanism of the afferent inflow to the spinal cord when the Ia afferent discharge is low. However, a sudden perturbation of the limb would lead to a high discharge rate in the Ia afferents, and because presynaptic inhibition would depress the synaptic effects of this discharge only to a limited extent the perturbation could be adequately counteracted by the evoked stretch reflex. Presynaptic inhibition would thereby at the same time modulate the central effect of the normal low rate Ia afferent discharge and allow compensatory reflexes to exert their full action. However, the existence of this simple mechanism still has to be established experimentally.

This study was supported by grants from The Danish Research Council, Danish Research Foundation, and The Danish Society of Multiple Sclerosis.

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Received 28 October 1997; accepted in final form 22 April 1998.

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