Changes in the effect of magnetic brain stimulation accompanying voluntary dynamic contraction in man

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1. The soleus (Sol) H reflex was conditioned by magnetic stimulation of the contralateral motor cortex at rest and during voluntary contraction in healthy human subjects. The intensity of the magnetic stimulus was adjusted so as to have no effect on the H reflex at rest. During tonic voluntary contraction the same magnetic stimulus produced a facilitation with a short latency and a long duration, thus reflecting an increased excitation of Sol motoneurones by the magnetic stimulus during voluntary contraction.

2. The amount of reflex facilitation produced by brain stimulation within the initial 0·5–1 ms after its onset was investigated at different times during dynamic ramp-and-hold plantar flexion. The facilitation was largest at the onset of voluntary activity in the Sol muscle. It then decreased abruptly within 100 ms after the onset of the voluntary contraction. Neither the voluntary Sol activity nor the control H reflex decreased at this time.

3. Electrical stimulation of the brain with the anode placed lateral to the vertex produced a facilitation of the H reflex, which preceded the facilitation evoked by magnetic stimulation by 1–2 ms. The facilitation produced by the magnetic stimulus occurred or increased at the onset of contraction in relation to rest in all experiments. However, this was the case in only two out of eight experiments, when the brain was stimulated electrically.

4. The size of the reflex facilitation measured at the onset of contraction was larger the faster the contraction. Positive correlations were found between the size of the facilitation and the peak of the first and second derivative of the torque and the peak Sol EMG activity.

5. It is suggested that the observed changes in the size of the short-latency reflex facilitation produced by magnetic brain stimulation mainly reflects changes in the excitability of corticospinal cells, since similar changes were not observed in the size of the unconditioned Sol H reflex or in the short-latency reflex facilitation produced by electrical brain stimulation. The data support the hypothesis that fast conducting corticospinal fibres with monosynaptic projections to spinal motoneurones are involved in the initiation of voluntary movement in man.

Following the original study by Barker, Jalinous & Freeston (1985), activation of descending corticospinal tracts through the intact scalp in awake human subjects by painless magnetic stimulation has been used widely. Several studies have suggested that the effect of the magnetic stimulus measured either as evoked responses in the surface electromyogram, as changes in the firing probability of single motor units or as changes in the size of the H reflex, is influenced by the excitability of corticospinal cells in the motor cortex (Datta, Harrison & Stephens, 1989; Day, Riescher, Struppler, Rothwell & Marsden, 1991; Deuschl, Michels, Berardelli, Schenck, Inghilleri & Lücking, 1991; Palmer & Ashby, 1992; Flament, Goldsmith, Buckley & Lemon, 1993; Nielsen, Petersen, Deuschl & Ballegaard, 1993). Nielsen *et al.* (1993) observed that a magnetic stimulus of the motor cortex, which was too weak to have any effect on the soleus (Sol) H reflex when the subject was at rest, produced a long-lasting facilitation with a short latency when the subject performed a tonic voluntary plantar flexion of the foot. As argued by Nielsen *et al.* (1993) only the initial 0·5–1 ms of the facilitation may be attributed with some certainty to activation of cortical cells with direct monosynaptic projections to spinal motoneurones, whereas later parts of the facilitation are likely to be contaminated by activation of non-monosynaptic pathways. The appearance of the initial part of the facilitation during voluntary contraction was suggested by Nielsen *et al.* (1993) to reflect an increased susceptibility to the magnetic stimulus of those cortical

cells which were voluntarily activated during the task. If this is the case, then changes in the initial part of the facilitation evoked by magnetic stimulation may reveal changes in the excitability of cortical cells in relation to different voluntary motor tasks.

The present study was initiated to investigate this possibility further. Corticomotoneuronal cells are known to modulate their firing rate in relation to simple ramp-and-hold contractions in a characteristic way (Fetz & Cheney, 1987). This firing rate modulation may potentially influence the susceptibility of the cells to stimulation and therefore, we studied changes in the effect of magnetic brain stimulation on the Sol H reflex during ramp-and-hold plantar flexion.

METHODS

General experimental arrangement

Seventeen healthy subjects with ages ranging from 20 to 56 years participated in the study. The number of subjects used for each type of experiment is mentioned in Results. All the subjects gave informed consent to the experimental procedure, which was approved by the local ethics committee.

The subjects were seated in an armchair. The subject's right leg was semi-flexed in the hip (120 deg), the knee flexed to 160 deg and the ankle in 110 deg plantar flexion. The foot was attached to a torque meter and the torque was displayed, together with the rectified and integrated electromyogram (EMG) from the Sol and tibialis anterior (TA) muscles, on an oscilloscope placed in front of the subject. All contractions investigated were *isometric*.

Voluntary contractions

At the beginning of each experiment the effect of magnetic brain stimulation on the Sol H reflex was measured at rest and during tonic plantar flexion. The effect of the magnetic stimulus was then investigated prior to, and during, ramp-and-hold contraction of the ankle plantar flexors. The ramp was drawn on an oscilloscope (time base of 2 s across the screen), which displayed the torque exerted on the foot pedal. The oscilloscope was triggered every 8 s at the same time as an auditory warning signal was given. The subject's task was to make the torque follow the prescribed ramp beginning 400 ms after the warning signal. The amplitude of the contraction was 10 N m. The duration of the ramp phase was varied from 300 ms to 1·6 s. In some experiments the subjects were, in addition, instructed to reach the 10 N m torque level as fast as possible ('step' contraction). The EMG signal from the Sol muscle was rectified and connected to a triggering circuit. The EMG onset was then used to trigger the stimulation with various time delays. The triggering level was adjusted throughout the experiment to be just above the background noise. A stimulation could thus be delivered at the very onset of the EMG or at any interval after this onset.

Measurement of background EMG and torque

In approximately every third trial no stimulation was applied. Instead the raw EMG from the TA and Sol muscles were sampled together with the torque exerted on the footplate. The sampling rate was 5 kHz in most experiments but in a few experiments lower and higher rates were used. The EMG was rectified, integrated and averaged off-line on a computer (software package from InfoWest Inc., Winnipeg, Canada). The torque was averaged and differentiated to obtain its first and second derivative.

Test reflexes

Surface electrodes were used for both stimulation and recording. The Sol H reflex was evoked by stimulating the posterior tibial nerve in the popliteal fossa through a monopolar stimulating electrode (1 ms shocks). The indifferent electrode was placed above the patella. The reflex responses were measured as the peak-to-peak amplitude of the H reflex recorded by disc electrodes over the Sol muscle. The size of the maximum motor response (M_{max}) was measured at the beginning of each experiment and the size of the test reflex was expressed as a percentage of this value.

In most experiments the H reflex was used to evaluate the effect of a conditioning stimulation of the motor cortex. In these experiments the tested parameter was the difference between conditioned and unconditioned reflexes. The amplitude of the unconditioned H reflex, however, varies considerably with the different tasks investigated in the study. This causes a complication, since reflexes of different sizes are not similarly sensitive to conditioning effects (Crone, Hultborn, Mazieres, Morin, Nielsen & Pierrot-Deseilligny, 1990). To evaluate the effect of brain stimulation on the H reflex it was, consequently, necessary to adjust the size of the unconditioned control H reflex during the different tasks by changing the intensity of the test stimulus. In this way the test H reflex was maintained constant during all tasks in each experiment.

However, in some experiments separate trials were performed without adjustment of the tibial nerve stimulation (and without any conditioning stimulation). In these experiments the tested parameter was the H reflex itself. These experiments were performed in order to obtain an estimate of changes in the excitability of the motoneuronal pool during the different tasks. In these experiments a small direct motor response, evoked by slightly stronger stimulation than that used to evoke the reflex, was measured during the same tasks. This was done to ensure that the changes in the reflex size was not caused by changes in the efficiency of the stimulus in activating Ia afferents (i.e. movement of the stimulating electrode relative to the nerve) or in the recording conditions (i.e. movement of the recording electrode relative to the muscle) during the different tasks. Measurements of the size of the unconditioned and unadjusted H reflex and M response were randomly alternated with measurements of the effect of the brain stimulation on the (adjusted) test reflex.

Conditioning stimuli

The H reflexes were conditioned by magnetic or electrical stimulation applied over the contralateral motor cortex. The magnetic stimulator was a MagStim 200 (MagStim Co. Ltd, Sheffield, UK). The coil was a prototype of the curved figure-of-eight coil and the maximal stimulator output was 2 T. At the beginning of each experiment the coil was moved to find the optimum position for stimulation of cortical areas projecting to the leg muscles. We found that activation of the leg muscles was obtained with the lowest threshold when the centre of the coil was placed 0–2 cm lateral to the vertex in the different subjects. The coil was placed so that the current in the centre of the coil flowed in the anteroposterior direction. The electrical stimulator was a Digitimer D180A (Digitimer Ltd, Welwyn Garden City, UK) with a maximal output of 1500 V. For stimulation of the

leg, the stimulating anode was placed 2 cm lateral to the vertex (and in some experiments additionally 2 cm posterior to the vertex), whereas the cathode was placed 4–6 cm anterior to the vertex.

At the beginning of each experiment the subject was requested to make a strong voluntary plantar flexion. The threshold for evoking a direct motor-evoked potential (MEP) in the muscle was then determined and the intensity of the stimulation (magnetic or electrical) was decreased below this threshold (approximately × 0·9 threshold). The MEP threshold was checked throughout the study. In most experiments the effect of the magnetic and electrical brain stimulation was investigated in separate trials. It was, however, ensured in three control experiments that similar results were obtained when the two types of stimulation were randomly alternated within the same trial.

Conditioned and unconditioned reflexes were randomly alternated. At least twenty reflexes of each alternative were measured and stored on the computer for later analysis. The mean and the standard error of the mean of each alternative were calculated on-line. The statistical significance of differences in the mean of measurements obtained during the different tasks was tested off-line using an unpaired Student's t test. Regression analysis was used to correlate the effect of the magnetic brain stimulation and the peak voluntary EMG and the peak first and second derivative of the voluntary torque. The Mann-Whitney U test was used to analyse changes in the effect of the brain stimulation and the H reflex at different times in relation to the onset of voluntary contraction.

RESULTS

Time course of the effect of magnetic brain stimulation on the Sol H reflex

As described in Methods the intensity of the magnetic stimulus was adjusted at the beginning of each experiment to be well below the threshold for eliciting a direct motor-evoked potential (MEP) in the Sol muscle even during strong voluntary contraction. A time course of the effect of the magnetic stimulus on the Sol H reflex was then obtained either while the subject was at rest or while performing a voluntary plantar flexion (Fig. 1). If the magnetic stimulus had an effect on the H reflex while the subject was at rest, the intensity of stimulation was decreased still further until no significant effects were observed (Fig. 1; ●). When the brain was stimulated at the same low intensity (45% of maximal stimulator output) during tonic voluntary plantar flexion, a facilitation of the reflex was produced at very short conditioning–test intervals (−5·4 ms for the subject in Fig. 1; ○. The negative conditioning–test interval designates that the conditioning stimulation was applied after the test stimulation). This facilitation was observed in all seventeen subjects tested. Its onset varied between −5·4 and −1·0 ms in the different subjects. This difference in latency is most likely explained by differences in the height of the subjects and in the conduction velocity of their central and/or peripheral fibres.

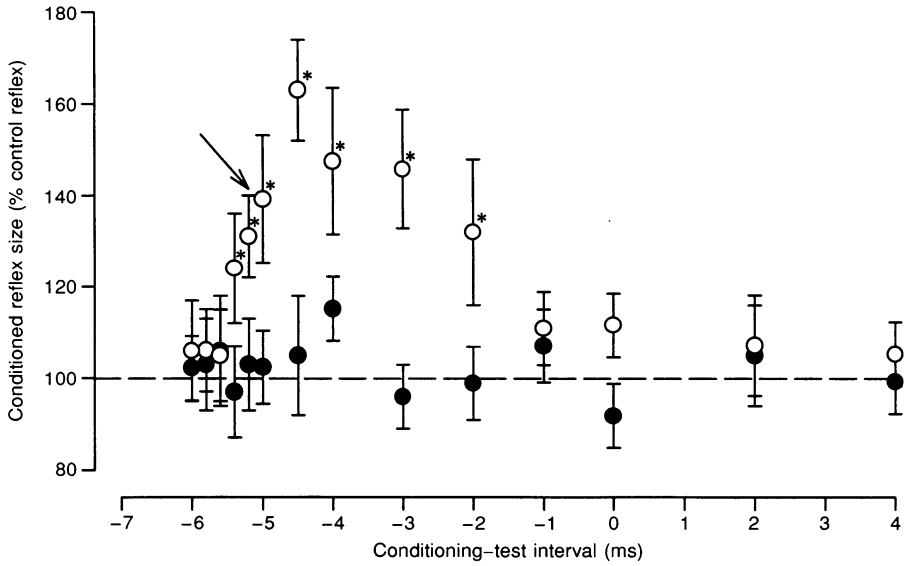

Figure 1. The effect of magnetic brain stimulation on the size of the Sol H reflex

The effect of magnetic brain stimulation on the size of the Sol H reflex at rest (●) and during plantar flexion (○). The data are from a single subject. The ordinate is the size of the conditioned reflex expressed as a percentage of the control reflex size. The abscissa is the interval between the conditioning brain stimulation and the test H reflex stimulation. The control H reflex size was adjusted to 15% of M_{max} in both situations. The intensity of the magnetic stimulus was 45%. The torque level was 10 N m during plantar flexion. Each bar represents one standard error of the mean. * designate that the brain stimulation induced a statistically significant change in the H reflex size ($P < 0·05$). The arrow indicates the conditioning-test interval at which the size of the facilitation was measured.

Nielsen *et al.* (1993) argued that the initial part of the facilitation may be caused by activation of cortical cells with monosynaptic projections to the spinal motoneurones. To avoid contamination from activation of non-monosynaptic descending pathways to the motoneurones, we measured the size of the facilitation within the initial 0·5 ms after its onset in most of the experiments in the present study. In a few experiments the time courses were constructed using bins of 1 ms and the facilitation was therefore determined only with the initial 1 ms after its onset. The average size of the facilitation in the seventeen subjects was 134·9 ± 4·0 (range, 112–162%) during tonic plantar flexion compared with 105·3 ± 1·3 (range, 98–111%) at rest (paired t test; $P < 0·01$).

The size of the reflex facilitation during the ramp phase of voluntary contraction

Figure 2 shows the size of the reflex facilitation in two different subjects (conditioning–test interval −4·0 and −3·0 ms, respectively) at different times during a ramp-and-hold plantar flexion. The ramp phase of the contraction lasted 300 ms and the final torque level was 10 N m. The data from the two subjects are shown with a short (Fig. 2*A*) and a long (Fig. 2*B*) time base. In both subjects the magnetic stimulus had no effect on the H reflex at rest, but produced a facilitation of 135 and 125%, respectively, during tonic plantar flexion. At the very onset of contraction the facilitation was, however, much larger in both subjects; 170 and 230%, respectively. A similar observation was made in all eight subjects tested

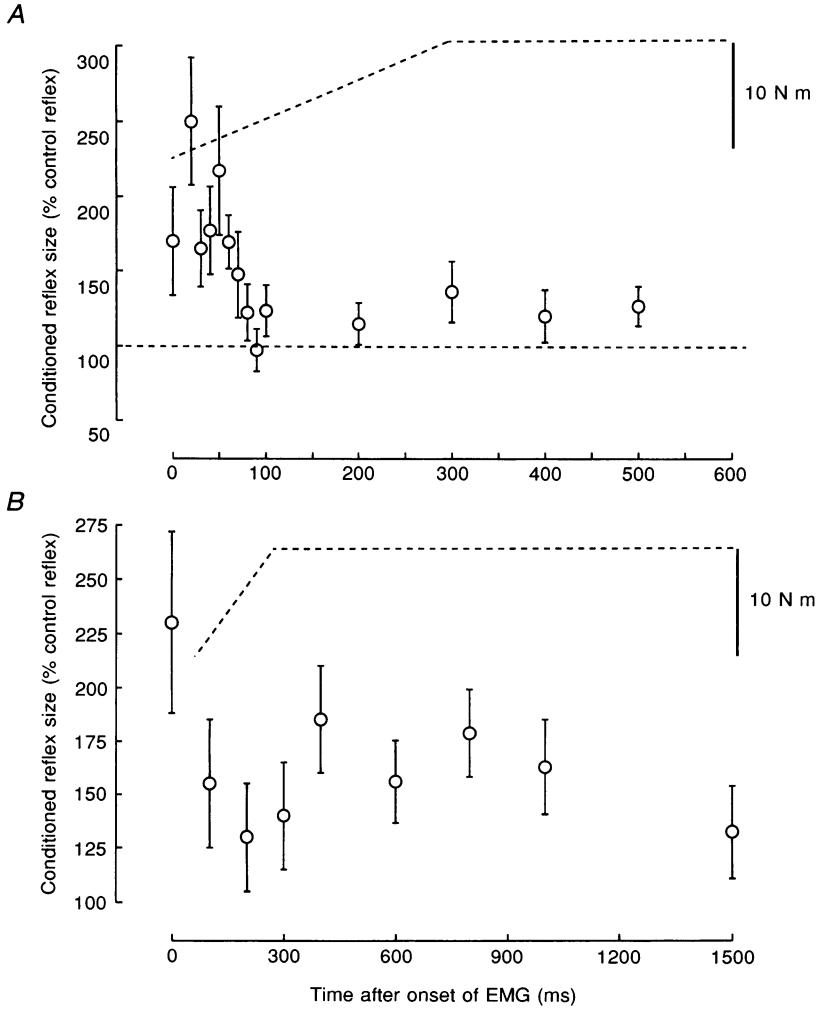

Figure 2. The size of the short-latency facilitation at different times during a ramp-and-hold plantar flexion in two different subjects (*A* and *B*)

In each case the duration of the ramp was 300 ms and the torque level was 10 N m. The ordinate is the size of the conditioned reflex as a percentage of the control H reflex. The abscissa is the time in relation to the onset of EMG activity in the Sol muscle. Each vertical bar represents one standard error of the mean.

where the average size of the reflex facilitation was 165% at the onset of contraction compared with only 122% during tonic contraction ($P < 0{\cdot}01$). As seen from Fig. 2*A* the facilitation decreased abruptly between 80 and 100 ms after the onset of contraction. In the other six subjects, in whom the facilitation was tested with similar short time intervals, it was found to decrease between 30 and 80 ms after the onset of contraction. In the remaining subjects it was confirmed that the facilitation decreased before 100 ms after the onset of contraction as seen from Fig. 2*B*. With the long time base in Fig. 2*B* it is also seen that a secondary increase of the facilitation occurred just after the end of the ramp phase. This secondary increase of the facilitation was a less common finding than the decrease of the facilitation, but was still seen in five of the subjects. Added data from fourteen experiments with eight subjects showed it to be statistically significant (i.e. the reflex facilitation was significantly larger around the end of the ramp (300–400 ms after the onset of contraction) than in the middle of the ramp and during tonic contraction; Mann–Whitney *U* test; $P < 0{\cdot}05$).

The size of reflex facilitation during voluntary contractions of different speed

To investigate whether the decrease of the reflex facilitation shortly after the onset of contraction was influenced by the speed of the contraction, eight subjects were requested to perform voluntary ramp-and-hold plantar flexions of differing speed in the same experimental session. In Fig. 3 data from one of these experiments are shown. The subject had to reach and maintain a torque level of 10 N m within either 150 ms ('step' contraction; ■), 300 ms (○) or 600 ms (△). The size of the facilitation at the same stimulus intensity was much smaller at the beginning of the slow contractions than at the beginning of the fast contractions (see below). To be able to compare changes occurring during the three types of contraction, the intensity of the conditioning magnetic stimulus was adjusted, so that the size of the facilitation was approximately the same at the onset of all three contractions.

In all three cases, the size of the reflex facilitation decreased 50–100 ms into the contraction as already described. For the very fast contraction ('step' contraction; dynamic phase lasting approximately 150 ms) the decrease occurred abruptly 50 ms into the contraction, whereas it occurred around 80 ms after EMG onset for the 300 ms contraction and around 100 ms after EMG onset for the 600 ms contraction. In the latter two cases the decrease was more gradual and less clear. Changing the duration of the dynamic phase of the contraction thus had little effect on the time of the decrease of the reflex facilitation. This was also the case in the experiments on the other seven subjects.When adding data from all eight subjects there was no significant difference in the time of the decrease of

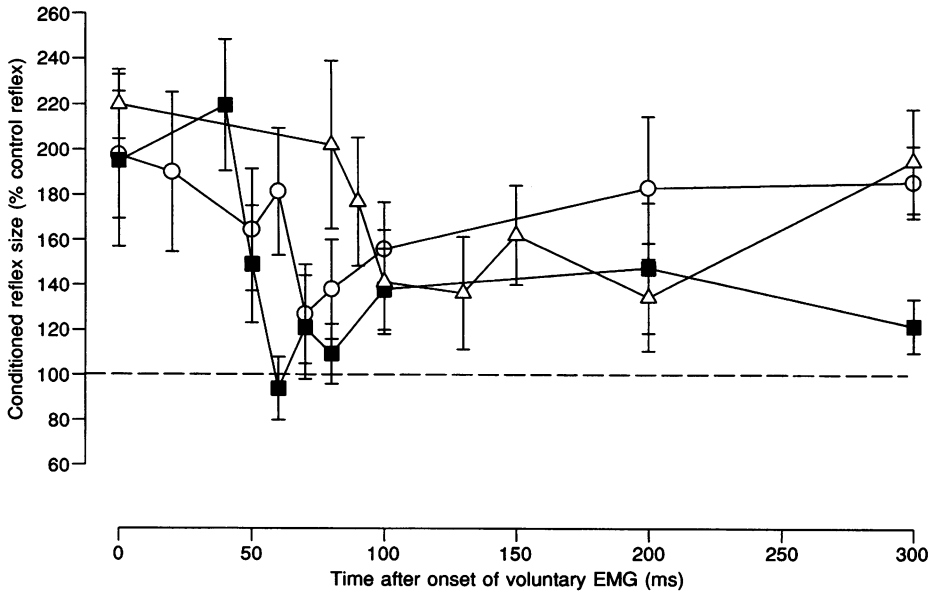

Figure 3. The size of the short-latency reflex facilitation during ramp-and-hold plantar flexions of different speed

The size of the short-latency reflex facilitation during ramp-and-hold plantar flexions of different speed. The data are from a single subject. The subject was instructed to perform a ramp-and-hold plantar flexion reaching a torque level of 10 N m within either 150 ms (step contraction, ■), 300 ms (○) or 600 ms (△). The conditioning–test interval was −3 ms and the size of the facilitation is expressed as the size of the conditioned reflex as a percentage of the control reflex size. The abscissa is the time in relation to the onset of EMG.

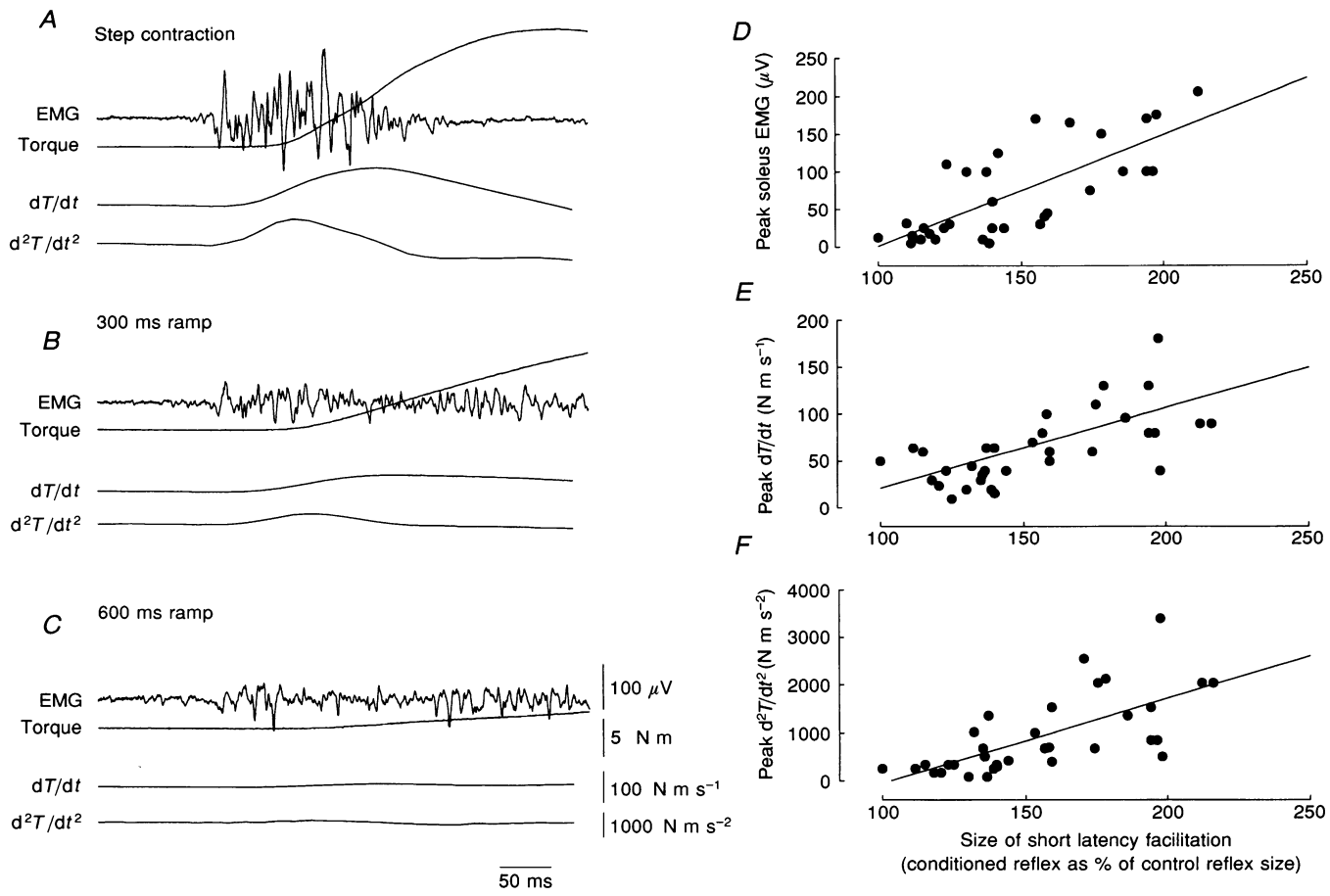

Figure 4. Relation of the short-latency reflex facilitation to the EMG and torque changes during voluntary plantar flexions of different speed

The data in *A–C* are from a single subject. In *A* the subject was instructed to reach the 10 N m target level as fast as possible, in *B* within 300 ms and in *C* within 600 ms. The first trace in each figure is the voluntary Sol EMG (μV), the second trace is the torque (N m), the third trace is the first derivative of the torque (N m s^{-1}) and the fourth trace is the second derivative of the torque (N m s^{-2}). All traces are the average of twenty trials. The size of the short-latency facilitation of the Sol H reflex evoked by the magnetic stimulus was measured at the onset of each of the three contractions. The intensity of the magnetic stimulus was 50% of the maximal stimulator output and the conditioning–test interval was −3 ms. In *A* the conditioned reflex was 156·8% of the control reflex size, in *B* it was 130% and in *C* it was 108%. *D–F*, correlation of the short-latency facilitation expressed as the conditioned reflex in percentage of the control reflex size (abscissa in all figures) and the peak voluntary Sol EMG (ordinate in *D*), the peak first (ordinate in *E*) and second derivative of the torque (ordinate in *F*). The data are from 33 measurements in 8 different subjects. Each subject performed at least 3 different contractions. The amplitude of the contractions was 10 N m in all cases. The subjects were requested to reach this torque level either as fast as possible or to follow a ramp with a duration varying between 200 and 1600 ms. The intensity of the magnetic stimulus was between 35 and 55% of the maximal stimulator output in the different subjects.The conditioning–test interval was between −4·0 and −2·8 ms. The peak EMG was measured from the rectified and integrated voluntary Sol EMG. For the slowest contractions a distinct peak was not observed. The highest EMG level measured within 200 ms after the onset of contraction was used in these cases. The straight line is the regression line calculated for the data. *D*, $y = 1{\cdot}5x - 147$, $r = 0{\cdot}74$ and $n = 33$; *E*, $y = 0{\cdot}85x - 64$, $r = 0{\cdot}58$ and $n = 33$; *F*, $y = 17{\cdot}7x - 1825$, $r = 0{\cdot}68$ and $n = 33$; where r is the correlation coefficient and n the number of replicates.

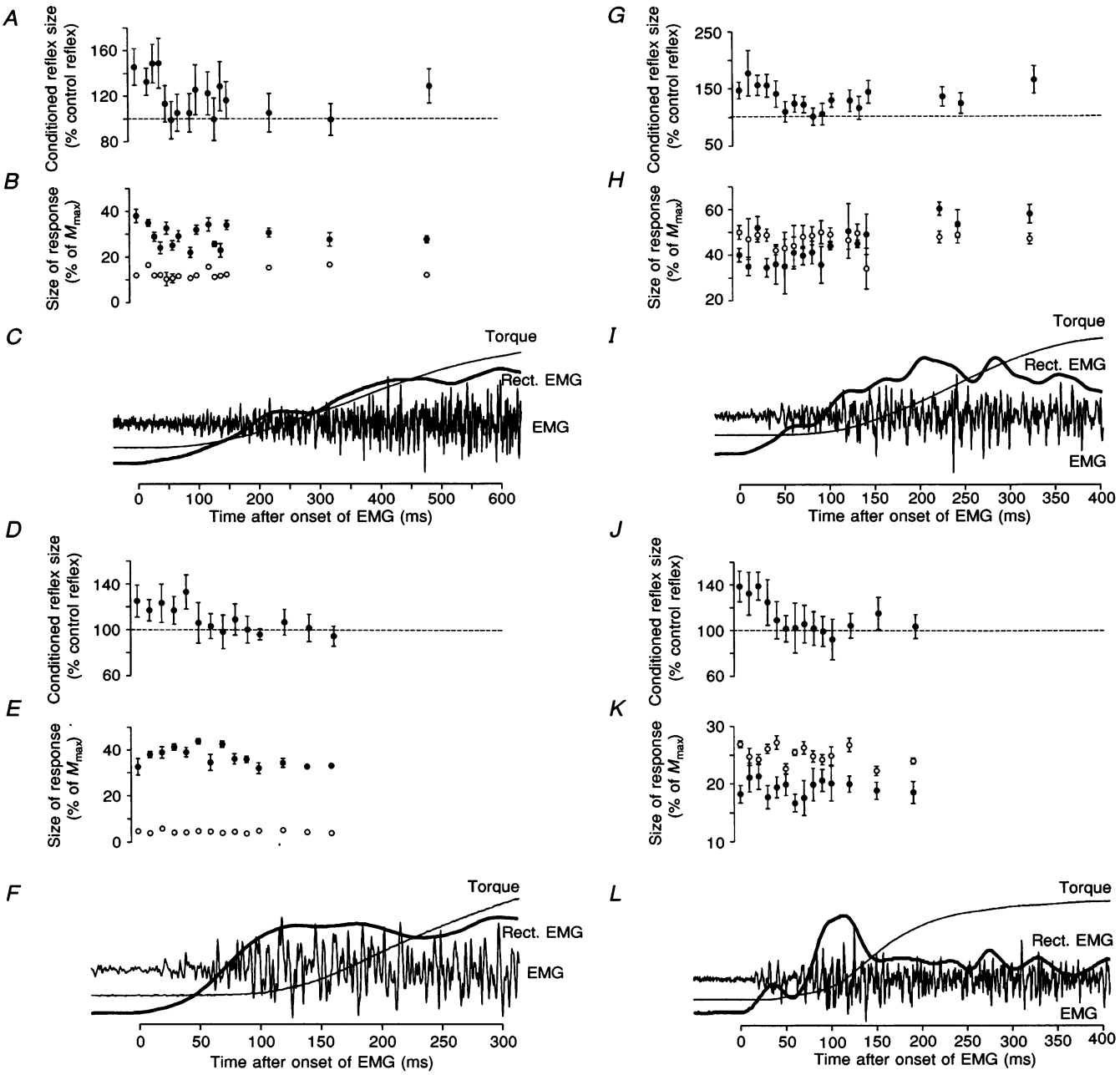

Figure 5. Comparison of the size of the short-latency facilitation, the Sol H reflex and the voluntary Sol EMG activity during a voluntary ramp-and-hold plantar flexion in four different subjects (*A-C*, *D-F*, *G-I* and *J-L*)

All subjects were requested to reach a torque level of 10 N m within either 600 ms (*A–C*), 300 ms (*D–I*) or as fast as possible (step contraction; *J–L*). In the latter case the dynamic phase of the contraction lasted approximately 200 ms. In all graphs the abscissa is the time in relation to the onset of EMG activity in the Sol muscle (note, however, the different scale for each of the four experiments). *A*, *D*, *G* and *J*, the Sol H reflex was conditioned by magnetic stimulation of the brain at a conditioning–test interval of −4·0, −3·0, −4·2 and −2·5 ms, respectively. The intensity of the stimulation was 40–50% of the maximal stimulator output. The ordinate is the size of the conditioned reflex as a percentage of the control reflex size. The size of the unconditioned reflex was adjusted in all cases to approximately 20% of M_{max}. Each vertical bar is one standard error of the mean. *B*, *E*, *H* and *K*, the size of the unconditioned (and unadjusted) Sol H reflex (●) and M response (○) as a percentage of M_{max}. The two responses were evoked by two separate stimulations to the tibial nerve, the stimulus evoking the M response being somewhat stronger than the stimulus evoking the H reflex (adjusted to evoke a reflex of approximately 50% of the maximal H reflex at the onset of contraction). Each vertical bar is one standard error of the mean. *C*, *F*, *I* and *L*, the voluntary Sol EMG is shown either unrectified or rectified and integrated (thick continuous line) together with the voluntary torque exerted by the subjects (thin continuous line). All traces are the average of approximately twenty trials.

the reflex facilitation between the step contraction and the 300 ms ramp contraction or between this contraction and the 600 ms ramp contraction (Mann–Whitney U test; $P > 0{\cdot}1$). However, the time of the decrease of reflex facilitation was significanly longer for the 600 ms ramp contraction than for the step contraction (Mann–Whitney U test; $P < 0{\cdot}05$).

The size of the reflex facilitation at the onset of voluntary contractions of different speed

Figure 4 *A–C* demonstrates averaged traces of the voluntary Sol EMG, the torque, the first (dT/dt) and the second derivative of the torque (d^2T/dt^2) from series in which the subject was requested to perform 10 N m contractions of varying velocity as in the previous experiment. In contrast to the experiments illustrated in the previous figure, the size of the reflex facilitation was measured at the very onset of EMG without any adjustment of the conditioning stimulation. The test stimulation, however, had to be adjusted as the control H reflex was very much larger at the onset of the fast contractions than at the onset of the slow contractions. The subject was instructed to perform the 10 N m contraction either as fast as possible ('step' contraction; Fig. 6*A*) or by making the torque follow a prescribed torque ramp lasting either 200 (Fig. 6*B*) or 600 ms (Fig. 6*C*). In the fast 'step' contraction, the voluntary EMG consisted of an initial burst lasting 130 ms followed (after a silent period) by a more sustained EMG activity during the hold phase of the contraction (not illustrated). At the same time as the burst of EMG, d^2T/dt^2 displayed a distinct peak, whereas dT/dt only displayed a peak 50 ms later. Similar, although smaller, bursts of EMG and peaks in d^2T/dt^2 and dT/dt were also seen for the 200 ms, but not for the slow 600 ms ramp contraction. Likewise, the short-latency facilitation was much larger at the onset of the step contraction (conditioned reflex 158% of control reflex size) than at the onset of the slow ramp contractions (130 and 108%, respectively). There was, thus, a significant positive correlation between the size of the short-latency facilitation measured at the onset of contraction and the speed of the contraction ($P < 0{\cdot}01$).

This is confirmed from Fig. 4*D–F*, in which data from eight subjects in whom these experiments were performed, has been pooled.

The short-latency facilitation was correlated to both the peak EMG (Fig. 4*D*), the peak dT/dt (Fig. 4*E*) and the peak d^2T/dt^2 (Fig. 4*F*) with correlation coefficients ranging from 0·58 to 0·74 ($P < 0{\cdot}01$). The facilitation was slightly better correlated to the peak EMG and the peak d^2T/dt^2 than to the peak dT/dt, but this difference was not statistically significant ($P > 0{\cdot}1$).

What is the cause of the changes in the reflex facilitation?

To investigate whether the described changes in the size of the reflex facilitation were caused by changes in motoneuronal excitability or could be attributed to changes in the excitability of cortical cells, the voluntary Sol EMG and the Sol H reflex were recorded and compared with changes in the size of the facilitation produced by the brain stimulation. This kind of experiment was performed in eight subjects. Representative data from experiments in four of the subjects are shown in Fig. 5. As in the previous figures the subjects performed a ramp-and-hold plantar flexion of 10 N m. The ramp phase lasted either 600 (Fig. 5*A–C*), 300 (Fig. 5*D–I*) or 200 ms (Fig. 5*J–L*).

The effect of the magnetic stimulus decreased around 50 ms into the contraction in all the subjects although the duration of the ramp phase varied from 200 to 600 ms (Fig. 5*A*, *D*, *G* and *J*). With the fastest contraction (Fig. 5*J*), a burst of EMG activity was observed in the beginning of the contraction followed by a silent period and a second burst around the end of the dynamic phase. In this case the decrease in the reflex facilitation coincided with the silent period in the EMG. However, the voluntary EMG increased gradually throughout the ramp phase of the relatively slow ramp-and-hold contractions without any evidence of a silent period (ramp phase lasting 300 to 600 ms; Fig. 5*C*, *F* and *I*). Despite this, the reflex facilitation still decreased. Furthermore, in none of the subjects did the decrease in the reflex facilitation coincide with a similar decrease in the size of the unconditioned (and unadjusted) reflex. In the subject with the slowest ramp contraction (Fig. 5*A–C*) the unadjusted H reflex decreased 20 ms into the contraction after which it maintained a constant amplitude. A similar pattern was also seen when this subject performed faster contractions. In the two subjects who performed a ramp contraction lasting 300 ms (Fig. 5*D–I*), the reflex increased gradually throughout the ramp phase of contraction. This was also the case when the subjects performed slower contractions, but when they performed faster contractions a decrease of the reflex size was often observed immediately after the onset of contraction, as seen for the subject illustrated in Fig. 5*A–C*. In the final subject (Fig. 5*J–L*) the reflex maintained a constant size throughout the ramp phase during both this fast contraction and slower contractions. Despite this variation in the regulation of the H reflex size in the different subjects, the decrease in the size of the reflex facilitation was always observed. When adding data from all eight subjects the reflex facilitation was found to decrease significantly around 60 ms after the onset of contraction (Mann–Whitney U test; $P < 0{\cdot}001$), whereas this was not the case for the H reflex (Mann–

Whitney U test; $P = 0{\cdot}9$). From these observations we find it unlikely that changes in motoneuronal excitability are responsible for the decrease of the reflex facilitation.

This was further strengthened from a comparison of the effect of magnetic and electrical stimulation of the brain. Nielsen *et al.* (1993) demonstrated that the electrically induced short-latency facilitation of the Sol H reflex was less dependent on changes in cortical excitability than the magnetically induced facilitation, probably because it primarily activates the axons of the cortical cells at some distance from the cell soma. In contrast, the magnetic stimulation probably activates the cells either indirectly or directly close to the cell soma. If the increase of the short-latency facilitation evoked by magnetic stimulation at the onset of contraction is caused by changes in cortical excitability, a similar increase should therefore not be seen for the short-latency facilitation evoked by electrical stimulation. The effect of magnetic and electrical brain stimulation was compared in eight experiments in four subjects. Data from these experiments are presented in

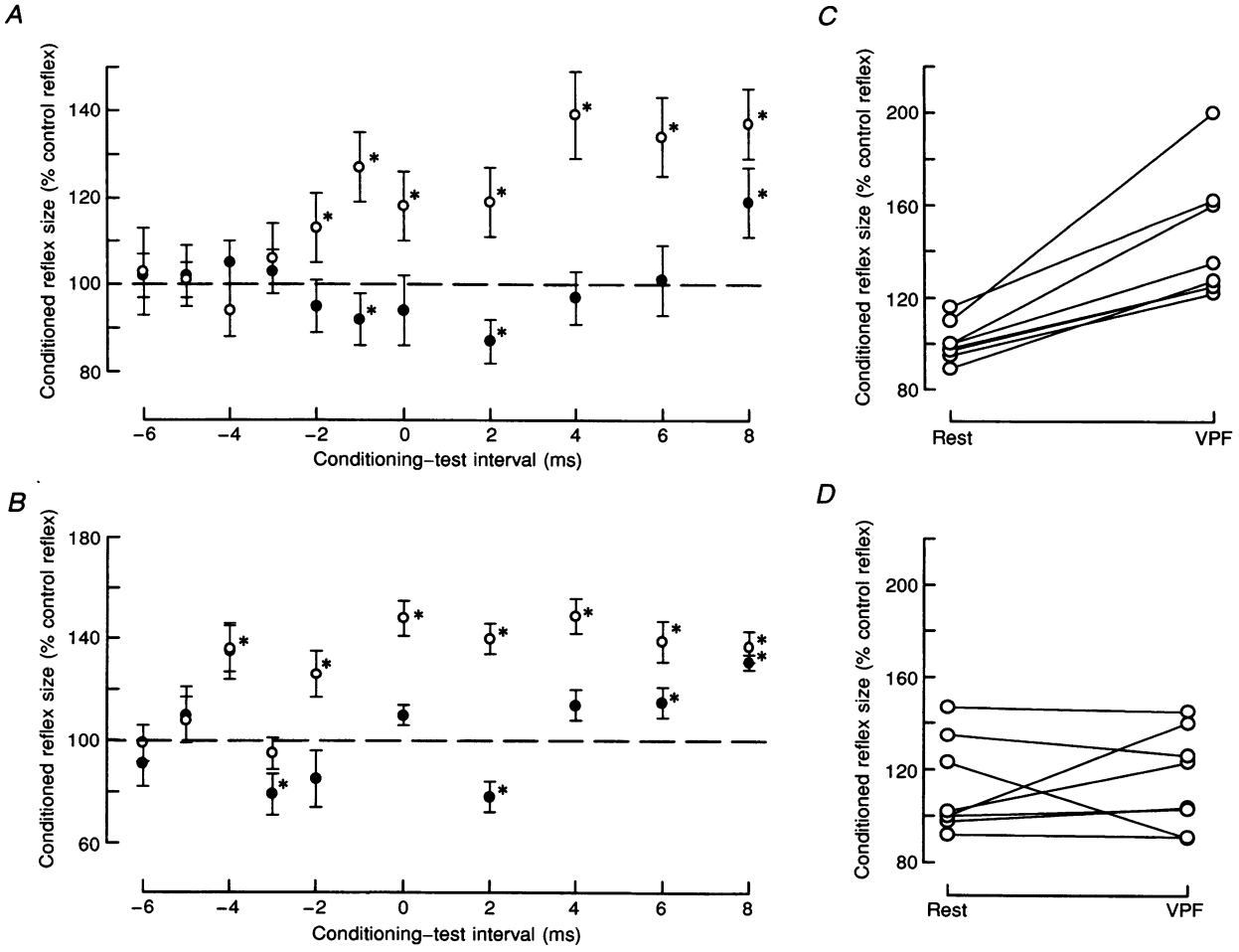

Figure 6. Comparison of the short-latency facilitation evoked by electrical and magnetic brain stimulation

Comparison of the short-latency facilitation evoked by electrical (D) and magnetic brain stimulation (C) at rest and at the onset of voluntary plantar flexion (VPF). A and B demonstrate the time course of the effect of the two types of stimulation on the H reflex in a single subject at rest (●) and during tonic voluntary plantar flexion (○) in a single subject. Same ordinate and abscissa as in Fig. 1. * designate that the brain stimulation induced a statistically significant change in the H reflex size. C and D, the data are from 8 experiments in 4 different subjects. Each line and circle represents one experiment. The intensity of the magnetic stimulus varied between 40 and 55% of the maximal stimulator output and the conditioning–test interval between −5·0 and −2·8 ms. The intensity of the electrical stimulus varied between 35 and 45% of the maximal stimulator output and the conditioning–test interval between −6·0 and −4·0 ms. The ordinate in the figures is the size of the conditioned reflex as a percentage of the control reflex size.

Fig. 6. In the beginning of the experiments a time course of the effect of the magnetic and electrical brain stimuli on the H reflex was obtained while the subject was at rest or performing a tonic voluntary plantar flexion. This is shown for one of the subjects in Fig. 6*A–B*. At rest both stimuli evoked an inhibition (●; notice that the inhibition evoked by the electrical stimulus preceded that evoked by the magnetic stimulus by 1–2 ms). The intensity of the two stimuli was adjusted so that the inhibitions produced by the two stimuli were of almost equal size (see also Nielsen *et al.* 1993). In the illustrated subject an earlier facilitation was evoked by the electrical stimulus both at rest and during contraction (Fig. 6*B*, ● and ○; conditioning–test interval, −4 ms), whereas the magnetic stimulus only evoked a facilitation during contraction (Fig. 6*A*, ○; conditioning–test interval, −2 ms). For the illustrated subject these conditioning–test intervals were then used for the rest of the experiment. In the majority of subjects, however, the electrical stimulation failed to evoke a facilitation. In these subjects a conditioning–test interval 1 ms prior to the onset of the inhibition was used. In Fig. 6*C–D* a comparison is made between the effect of the two stimuli at these conditioning–test intervals at rest (left side) and at the onset of a voluntary ramp-and-hold plantar flexion (right side). Each line and symbol represent data from one experiment. In all eight experiments the magnetic stimulus had no effect at rest (Fig. 6*C*, left side; average size of the conditioned reflex in relation to the control reflex, 100·6 ± 3·0%) but produced a significant facilitation at the onset of contraction (Fig. 6*C*, right side; average size, 144·6 ± 9·7%). The difference in the effect of the magnetic stimulus between the two situations was highly statistically significant (paired *t* test; $P < 0{\cdot}01$). The electrical stimulus also failed to evoke a facilitation at rest except in three of the experiments (Fig. 6*D*, left side; average size, 112·0 ± 7·1%). However, in contrast to the magnetic stimulus it also failed to evoke a facilitation at the onset of contraction in the majority of the experiments. The average effect of the electrical stimulus on the reflex was thus 115·2 ± 7·5% at the onset of contraction, which was not significantly different from rest (paired *t* test; $P > 0{\cdot}5$). This suggests that the increase (or appearence) of the reflex facilitation evoked by magnetic stimulation at the onset of contraction is caused mainly by an increased excitability of the cortical cells that become active in relation to the voluntary contraction.

DISCUSSION

Can H reflex testing of the effect of magnetic stimulation of the brain be used to investigate the excitability of corticomotoneuronal cells in man?

Corticomotoneuronal cells of the monkey have been shown to modulate their firing rate during voluntary contraction (Fetz & Cheney, 1987). The underlying changes in excitability of the cells may influence their susceptibility to stimulation provided that the stimulation excites the cells either indirectly or sufficiently close to the cell soma. This may be the case for magnetic stimulation of the intact human brain (Day *et al.* 1989; Nielsen *et al.* 1993) and changes in firing rate of cortical cells may, therefore, result in changes in the descending volley evoked by this kind of stimulus. The basic idea of the present study is that the H reflex technique may be used to monitor these changes. The validity of the protocol rests on the assumption that the degree of H reflex facilitation produced by brain stimulation reflects the size of the descending volley, i.e. the number of corticomotoneuronal cells activated by the stimulus. Therefore, it should firstly be questioned whether this is a valid assumption.

Some alternative possibilities might be proposed. Firstly, it is well documented that monosynaptic reflexes may be used to monitor the effect of conditioning inputs to quiescent motoneurones (Lloyd, 1941). However, in the present study the effect of the conditioning stimulus was investigated during voluntary contractions, i.e. when the background excitability of the motoneurones was not constant. This may introduce non-linearities in the summation of conditioning and test inputs to single motoneurones or in the recruitment of successive motoneurones (recruitment gain; Kernell & Hultborn, 1990) and thus interfere with the interpretation of the results. Secondly, in order to evaluate the effect of the conditioning stimulus it was necessary, in a number of cases, to adjust the size of the control H reflex by changing the intensity of the test stimulus eliciting the reflex (Crone *et al.* 1990). The stimulus, therefore, probably activated a different number of afferents in the different tasks. Furthermore, although the constant size of the control reflex indicates that a comparable proportion of the motoneuronal pool was activated, we cannot be sure that the test stimulus recruited exactly the same motoneurones in the different tasks. The susceptibility of the H reflex to the effect of the conditioning volley evoked by the brain stimulus may, therefore, be very different in the different tasks.

However, these possibilities do not adequately explain several of the findings in the present study. The short-latency facilitation decreased 30–80 ms into the movement without any similar changes in the size of the unconditioned Sol H reflex or in the voluntary EMG activity at the same time. The test stimulus eliciting the H reflex was thus constant in this case and there was no apparent decrease in the excitability of the motoneuronal pool. Furthermore, we did not find any task-related changes in the facilitation of the H reflex evoked by electrical stimulation of the cortex. Nielsen, Petersen & Ballegaard (1995) suggested that electrical stimulation of the brain with the anode placed lateral to the vertex as in

the present study may preferentially activate the axons of the corticospinal cells at a site below the cortex. This explains not only the earlier occurrence of the facilitation evoked by electrical stimulation, but also the lack of task-dependent changes. Had the changes in the size of the magnetically induced facilitation been caused by changes in motoneuronal excitability, we would have expected similar changes for the electrically induced facilitation. Finally, Meunier & Pierrot-Deseilligny (1988) have demonstrated that the monosynaptic facilitation of the Sol H reflex evoked by stimulation of femoral nerve Ia afferents does not decrease until the middle of the ramp phase during similar ramp-and-hold plantar flexion as in the present study. (This finding was also confirmed in relation to the present study; J. Nielsen & N. Petersen, unpublished observations.) Had the decrease in the reflex facilitation evoked by the brain stimulation shortly after the onset of contraction been caused by excitability changes at a motoneuronal level, a similar decrease should have been expected for the facilitation evoked by the femoral nerve stimulation.

It could also be argued that the facilitation was in fact contaminated by activation of an indirect pathway to the motoneurones. In this case the changes in the facilitation could be caused by changes in the excitability at an interneuronal level. It should be noticed, especially, that the decrease of the facilitation after the onset of contraction coincided with the arrival of peripheral feedback from the contraction at the level of the spinal cord (Vallbo, 1971). Although we tried to maintain isometric conditions as far as possible, some stretching of ankle dorsiflexors may, nevertheless, have occurred and the contraction may thus have caused a burst of activity in Ia afferents from the dorsiflexors. This must be assumed to result in an increased activity of the Ia inhibitory interneurones, which mediate reciprocal inhibition of Sol motoneurones. Since the same interneurones also receive input from descending fibres, which are activated by the magnetic stimulus (Nielsen *et al.* 1993), the decrease of the facilitation could be explained by an increased excitability of the Ia interneurones. In the study by Nielsen *et al.* (1993) the inhibition was always found at an interval at least 1 ms longer than the facilitation. The facilitation evoked by the magnetic stimulus was, therefore, always measured within the initial 0·5–1 ms after its onset in the present study. We argue that this interval is too short for a volley in non-monosynaptic pathways, such as the disynaptic inhibitory pathway, to arrive at the motoneurones and that the facilitation measured within this interval may thus be regarded as a purely monosynaptic effect. Furthermore, the decrease of the size of the reflex facilitation still remains when activity in the peroneal nerve is blocked reversibly by lidocaine injection (J. Nielsen, N. Petersen & C. Crone, unpublished observations).

A final concern could be that our findings might reflect changes in presynaptic inhibition of the terminals of the descending fibres. This possibility is unlikely, however, since descending fibres in man and the cat seem to be free from presynaptic inhibition (Eide, Jurna & Lundberg, 1968; Rudomin, Nunez & Madrid, 1975; Berardelli, Day, Marsden & Rothwell, 1987; Nielsen & Petersen, 1994).

Consequently, we suggest that changes in the short-latency facilitation of the Sol H reflex during voluntary movements may indeed be used to monitor changes in the excitability of corticomotoneuronal cells. If so, our observations should resemble previous observations on the discharge of single corticomotoneuronal cells in the monkey.

Do the changes in the facilitation resemble the discharge profile of corticomotoneuronal cells?

Unfortunately, only little is known of the natural activity of corticomotoneuronal cells projecting to leg motoneurones. Data from corticomotoneuronal cells projecting to forearm muscles in the monkey are, however, well in line with our findings. In the monkey, the majority of forearm corticomotoneuronal cells are thus most active just before and at the onset of contraction, but then decrease their firing rate after movement onset (Fetz, Cheney, Mewes & Palmer, 1989). It is a possibility that the changes in excitability accompanying these changes in firing rate explain the decrease of facilitation in the present study. In the monkey, corticomotoneuronal cells increase their firing rate as a function of the first derivative of torque (Cheney, Mewes & Fetz, 1988). This probably explains the correlation between the size of the facilitation and the first and second derivative of the torque in the present study. It should finally be mentioned that Nielsen & Petersen (1992), found that the facilitation of the Sol H reflex evoked by the magnetic brain stimulation increased 70–100 ms prior to the onset of EMG. Although corticomotoneuronal cells begin to discharge at very different times in relation to the onset of contraction, the most notable change in the population activity occurs around 100 ms prior to contraction (Cheney & Fetz, 1980).

Functional considerations

The observation that the short-latency corticospinal facilitation increased prior to contraction and was largest at the very onset of contraction is in accordance with previous studies, which have suggested that the corticomotoneuronal cells are involved in bringing the motoneurones to threshold and thus initiate the movement (Porter, 1970; Cheney & Fetz, 1980; Lemon & Mantel, 1989). Corticomotoneuronal cells have been shown to discharge at a very rapid rate around the onset of contraction (Lemon & Mantel, 1989) and a significant potentiation of two EPSPs arriving with such short intervals has been demonstrated (Porter, 1970; Porter & Muir, 1971). Thus, the corticomotoneuronal projection

seems especially well suited to bring the motoneurones to their firing threshold at the onset of contraction.

One of the few motor deficits seen following lesion of the pyramidal tract in monkeys (in addition to the well-known impairment of fine manipulative skills; Lawrence & Kuypers, 1968) is a reduction in the speed of phasic voluntary contraction (Tower, 1940, 1944; Hepp-Reymond, Trouche & Wiesendanger, 1974; Hepp-Reymond & Wiesendanger, 1976). It has also been shown that corticomotoneuronal cells discharge vigorously in relation to phasic changes in force output (Evarts, 1968; Cheney & Fetz, 1980) and it has therefore been suggested that the large diameter component of the pyramidal tract may be necessary for rapid phasic contractions. Our findings that the short-latency facilitation was larger in phasic than in tonic contractions and that it increased with the speed of contraction are well in line with this.

Ghez & Gordon (1987) have shown that fast isometric contractions consist of an initial agonist burst, followed by a burst of the antagonist and finally a second burst of the agonist. They suggested that this triphasic pattern is caused by a central programme controlling the trajectory of the force output (pulse-step protocol). This supports the long-held view that voluntary movements are controlled by an initial motor command (initial adjustment; Woodworth, 1899) which determines most of the movement of the limb and acts independently of peripheral feedback. The final adjustments of the contraction before reaching the goal on the other hand may be determined by an interaction between the central commands and the peripheral feedback (current control; Woodworth, 1899). In our experiments we never observed bursts in the antagonist EMG, but during the fastest contractions a distinct agonist burst was seen at the beginning of the contraction followed by a later, more gradual, and maintained increase of EMG. Like the agonist EMG, the short-latency corticospinal facilitation also displayed two phases during ramp-and-hold plantar flexions, i.e. the initial increase of facilitation before the onset of contraction (which decreased again 30–80 ms into the contraction) and the secondary increase seen around the end of the ramp phase.

Gordon & Ghez (1987*a*, *b*) suggested that the initial burst of EMG determined most of the limb's trajectory towards the target and further argued that the trajectory was determined by setting the amplitude of this burst, rather than its duration (pulse–height control). Similarly, we observed that the size of the short-latency facilitation at the onset of contraction was larger the faster the contraction, whereas the duration of the facilitation changed only very little. Consequently, we suggest that activity of the corticomotoneuronal cells, which we believe to be responsible for the facilitation, may at least contribute to the EMG and torque changes studied by Ghez and co-workers and shown also in the present study. Corticomotoneuronal cells may thus have a role in rapid initiation and execution of fast contractions ensuring a powerful output from the motoneuronal pool independent of peripheral feedback to the central nervous system from the contracting muscles.

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