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Timing of cortical excitability changes during the reaction time of movements superimposed on tonic motor activity

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Schneider, Cyril, Brigitte A. Lavoie, Hugues Barbeau, and Charles Capaday. Timing of cortical excitability changes during the reaction time of movements superimposed on tonic motor activity. *J Appl Physiol* 97: 2220–2227, 2004; doi:10.1152/japplphysiol.00542.2004.—Seated subjects were instructed to react to an auditory cue by simultaneously contracting the tibialis anterior (TA) muscle of each ankle isometrically. Focal transcranial magnetic stimulation of the leg area of the motor cortex (MCx) was used to determine the time course of changes in motor-evoked potential amplitude (MEP) during the reaction time (RT). In one condition the voluntary contraction was superimposed on tonic EMG activity maintained at 10% of maximal voluntary contraction. In the other condition the voluntary contraction was made starting from rest. MEPs in the TA contralateral to the stimulation coil were evoked at various times during the RT in each condition. These were compared to the control MEPs evoked during tonic voluntary activity or with the subject at rest. The RT was measured trial by trial from the EMG activity of the TA ipsilateral to the magnetic stimulus, taking into account the nearly constant time difference between the two sides. The MEPs became far greater than control MEPs during the RT (mean = 332%, SD = 44 %, of control MEPs, $P < 0.001$) without any measurable change in the background level of EMG activity. The onset of this facilitation occurred on average 12.80 ms (SD = 7.55 ms) before the RT. There was no difference in the onset of facilitation between the two conditions. Because MEPs were facilitated without a change in the background EMG activity, it is concluded that this facilitation is specifically due to an increase of MCx excitability just before voluntary muscle activation. This conclusion is further reinforced by the observation that MEPs evoked by near-threshold anodal stimuli to the MCx were not facilitated during the RT, in contrast to those evoked by near-threshold transcranial magnetic stimulation. However, several observations in the present and previous studies indicate that MEP amplitude may be more sensitive to α-motoneuron activity than to motor cortical neuron activity, an idea that has important methodological implications.

magnetic stimulation; motor cortex; cortical excitability

SINCE THE ADVENT OF MAGNETIC brain stimulation developed by Barker et al. (3) some 20 years ago, the question of whether the size of the evoked corticospinal volley depends on the excitability of the motor cortex (MCx) at the time of stimulation remains unanswered. There are many indirect observations in affirmative support of the question, but no single definitive demonstration. The most convincing evidence is the contrast between the effects of threshold magnetic vs. threshold anodal stimuli applied to the hand area of the MCx in different tasks (12, 21). For example, Datta et al. (12) reported that during voluntary abduction of the index finger threshold magnetic stimuli evoked larger motor potentials in the first dorsal interosseus (1DI) than during a power grip, at matched levels of motor activity (but see Flament et al., Ref. 21). By contrast, the response to threshold anodal stimulation was the same in both tasks (12). The interpretation is that threshold magnetic stimuli activate corticospinal neurons in layer V of the motor cortex transsynaptically, or at the initial segment, and consequently the size of the evoked corticospinal volley is sensitive to the level of excitability of the corticospinal neurons (35). Threshold anodal stimuli are thought to activate corticospinal axons in the white matter and initiate D waves (2). Because the site of action potential initiation is relatively distant from the soma, the evoked corticospinal discharge is relatively uninfluenced by the state of intracortical excitability (17, 18, 35). Such observations, however, are mitigated by the fact that the enhancement of a magnetically evoked motor potential accompanying voluntary activation is predominantly the result of the increased excitability of the α-motoneurons and not that of the motor cortical network (28, 30, 18). Furthermore, epidural differential recordings of the corticospinal volleys elicited by magnetic stimulation of the hand area of the MCx show that I waves are increased by only 12–15% during contractions of 100% maximal voluntary contraction (MVC) (18). One potential problem with this method is that the recording electrodes are in the epidural space along the dorsum of the cord and thus electrically distant from the pyramidal funiculi. The electric potential of the moving dipole associated with the action potential decreases with the square of the distance. Thus changes in the amplitude of the corticospinal volleys recorded by distant electrodes will be markedly attenuated and the true change underestimated.

When magnetic stimulation of the leg area of the MCx is considered, other potential difficulties arise. Because the leg area is largely located in the paracentral lobule at the vertex of the head (32), the corticospinal axons may bend sharply in going from the gray to the white matter in the internal capsule. Consequently, the spatial gradient of the electric field will be very high at the bend, promoting excitation at this site (1). The initial experiments showed that at threshold D waves were preferentially elicited (35). However, the more recent paper by Di Lazzaro et al. (17) reports that an anodal stimulus at the vertex preferentially elicits an I wave, whereas a D wave is preferentially elicited with the anode placed 2 cm lateral to the vertex. Higher intensities of magnetic and anodal electric stimuli to the leg area are thought to elicit I waves. The issue of what elements of the cortical network are recruited by

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magnetic and anodal stimuli is separate from the issue of whether and to what extent the size of a motor-evoked potential (MEP) depends on the level of cortical excitability at the moment of stimulation. The two issues are of course related, but the former is a static problem whereas the latter is a dynamic one related to, among other things, the average difference between the membrane potential and threshold in "resting" vs. active motor cortical neurons.

Given the current interest in the potential role of the MCx during human walking and postural activities (e.g., Refs. 8, 11, 25, 37), we thought it important to provide additional evidence that the size of MEPs depends on the excitability of the MCx at the time of stimulation. We reasoned that if one can show MEP enhancement without an accompanying change in the background level of motor activity, as was done by Flament et al. (21) for the hand, this would add to the evidence sought. To this end, we asked subjects to tonically contract the ankle dorsiflexor tibialis anterior (TA) and on hearing an auditory tone to rapidly increase the ongoing contraction to a higher prescribed value. During the reaction time (RT) of this task (i.e., simple reaction time), the background level of motor activity is essentially constant. Any enhancement of an MEP during the RT implies that the change must occur at the premotoneuronal level. In this task, because the α-motoneurons are tonically active, any increase of spinal interneuron excitability, or that of any other input to the α-motoneurons, would immediately lead to a measurable increase of motor activity (5, 6). The reason for this is as follows. A subthreshold input to a resting motoneuron does not produce a spike; the discharge probability is thus zero. When the motoneuron is active, that same input will produce a spike in a proportion of the discharge cycles; the discharge probability is therefore greater than zero (5, 6). Furthermore, a substantial number of motoneurons in the subliminal fringe (i.e., depolarized relative to rest but inactive) may respond to the input with a probability of one (28). Thus, by contrast to an input that may be subthreshold at rest, during activity the increased discharge probability of the motoneuron pool leads to immediate spike discharge in a proportion of the motoneurons. This is readily detectable in the EMG, which is in effect a multiunit peristimulus time histogram (7). Consequently, in the present task an enhancement in the amplitude of the MEP, occurring independently of the level of background motor activity, is strong evidence that it is the result of increased motor cortical excitability. A summary of some of the results presented here was published as an abstract (36).

METHODS

Subjects. This study was performed on 10 healthy human subjects ranging in age between 22 and 45 yr (mean 33.3 yr, SD 8 yr). All subjects were informed of the nature and purpose of the experiments in accordance with the Declaration of Helsinki, and the study was approved by the local ethics committee.

Behavioral task and instructions. Experiments were done on seated subjects (hip flexed 120°, knee flexed 130°, and ankle dorsiflexed 110°). Subjects were instructed to simultaneously and rapidly contract both TAs in reaction to a 1-kHz tone lasting 200 ms. The tones were delivered at random every 2–4 s. The feet were firmly strapped to an inclined footstep fixed to the ground; the contractions were thus essentially isometric. This simple RT task was performed in two different ways. In one condition the subjects started from rest (i.e., no EMG activity in the TAs). In the second condition subjects maintained a tonic voluntary contraction (i.e., both TAs isometrically contracted) of 10% of their MVC and on hearing the tone rapidly increased the contraction to a prescribed value of 50% MVC. The rectified and filtered surface EMG activity of each TA was displayed on an analog meter; the full-scale deflection of the needle corresponded to the MVC. A translucent mark on each meter's screen indicated the initial contraction level (10% of MVC) and a second mark the level subjects had to reach after hearing the auditory cue (50% of MVC).

Magnetic stimulation may shorten simple RTs when delivered near the time of the "Go" signal, an example of nonspecific intersensory facilitation (38). It can also lengthen the RT when delivered near the expected time of EMG activation (14, 40). Moreover, measurement of the RT from the onset of the TA voluntary EMG activity may be obscured by an MEP or by the post-MEP silent period. To circumvent this problem, we measured at the onset of each experiment the RT difference between the ipsilateral and contralateral TA, trial by trial. This was done by visual inspection of the records and from the cumulative sum (cusum; Ref. 19) of the EMG activities. The two measures were nearly identical. The RT difference between the two sides remained fixed throughout the experiment ($P < 0.05$), as shown in Fig. 1. Thus, to determine the actual onset of the TA contralateral to the stimulating coil (tested side), the measured time difference was added to or subtracted from, depending on the subject, the RT of the ipsilateral TA.

EMG recordings. EMG recordings were obtained from the TA and soleus muscles of each leg with bipolar Ag-AgCl electrodes filled with saline gel. Each electrode had a recording surface of 7 mm in diameter. The electrodes were connected to optically isolated preamplifiers. A large reference electrode connected to the common input of the preamplifiers was placed on the neck just above the shoulder. The EMG signals were amplified, high-passed at 20 Hz, and low-passed at 1 kHz before being sampled at 4 kHz by an analog-to-digital converter. The same signals were also separately amplified, high-pass filtered at 20 Hz, rectified, and low-passed at 100 Hz before being sampled at 4 kHz. The mean level of background EMG activity was measured from the rectified signals 20 ms before the MEP well after the stimulus artifact decayed. Comparison of responses obtained in the different tasks was done for matched levels of background EMG activity. In the RT task starting from rest, this analysis, as well as measurements of the contralateral TA EMG activity, ensured that no EMG activity preceded the MEP.

Magnetic and electric brain stimulation. Magnetic stimuli were applied to the scalp with a focal coil connected to a Cadwell MES-10 electromagnetic stimulator. The coil was coned and double D shaped, each D-shaped half being 7 cm long by 8 cm wide. The coil was positioned just lateral to the vertex at a site having the lowest threshold for activation of the TA at rest. Once the stimulation site was localized, its locus was marked with a cross hair drawn on the scalp. This served as a visual reference against which the coil was positioned and maintained by the experimenter. The magnetic stimulus intensity was expressed as a percentage of the stimulator's maximum output. At the onset of each experiment, the sigmoid-shaped input-output curve relating stimulus intensity to MEP size was measured at rest and during a contraction of 10% MVC (16). Typically 25 different stimulus intensities were used and eight responses averaged at each intensity. From the fitted curve parameters we obtained the stimulus intensity (S_{50}) that evokes a half-maximal MEP and estimated the resting and active motor thresholds (16). The time course of changes in MEP facilitation was determined at several stimulus intensities including threshold stimuli, stimuli of S_{50} intensity, and twice motor threshold.

The conditions were studied in random order from one subject to another. A single magnetic stimulus was applied during the RT. Stimuli were delivered at intervals of 25 ms starting from the onset of the auditory tone, in a random order. Each interval was tested four times and the MEPs were averaged. The time interval increments were reduced to 5 ms when within 25 ms of the average RT.

In two subjects, electrical stimuli were applied over the scalp through a radio frequency isolation transformer. Two Ag-AgCl electrodes of the same type as the recording electrodes were used. The anode was placed lateral to the vertex over the same scalp position as for magnetic stimulation, and the cathode was placed 3–4 cm more laterally along the motor strip. Square pulses of 200-μs duration were delivered at random between 2 and 4 s. The stimulus intensity that evoked a threshold response in the voluntarily activated TA was first determined and defined as the active motor threshold. The stimulus strength was then adjusted so as to produce MEPs of comparable size to those produced by threshold magnetic stimuli under corresponding conditions. Comparison of MEPs elicited by anodal and magnetic stimuli were made at matched levels of TA EMG activity.

Summary. The time course of changes in MEP size during the RT was determined by comparing the size of MEPs at various times in the RT to the size of MEPs obtained when the subject was instructed not to react to the auditory tone. The latter will be referred to as control MEPs and were obtained for two conditions, at rest and with tonic TA activity of 10% MVC. MEPs elicited during the RT will be referred to as test MEPs.

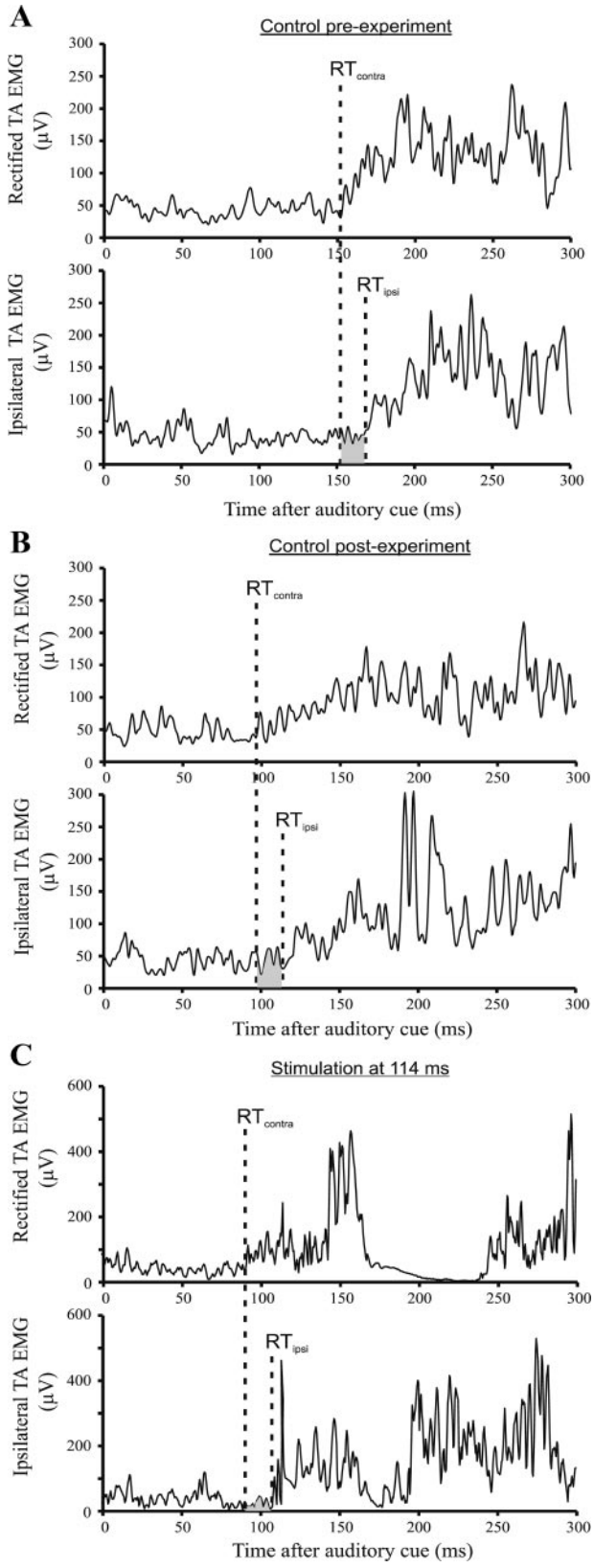

Fig. 1. Difference in reaction times (RTs) between the tibialis anterior (TA) ipsilateral (RT_{ipsi}) and contralateral (RT_{contra}) to the magnetic stimulus remained constant during the experiment. Note the tonic EMG activity on which is superimposed the voluntary increase of activity in response to the auditory cue. *A*: the RT difference at the start of experiment with no stimulation was 18.5 ms ($n = 10$, SD 2.6 ms). *B*: the RT difference at the end of experiment with no stimulation was 17.75 ms ($n = 10$, SD 3.3 ms). *C*: magnetic stimulation applied 114 ms after the auditory cue; the RT difference is nearly the same as in *A* and *B* (18.6 ms, $n = 7$, SD 2.9 ms). Note that the RTs when no magnetic stimulation was applied were measured several times during the course of an experiment to ensure the validity of the estimates.

RESULTS

Characteristics of the ipsilateral and contralateral RTs. The average RT to the auditory cue was 127.3 ms (SD 26.5 ms, $n = 10$ subjects). No statistical difference in RTs was found in starting from rest vs. tonic preactivation (respectively, mean 128.1 ms, SD 25 ms vs. 126.1 ms, SD 30 ms, $P = 0.6$). An important and consistent feature of the RTs was that there was a constant time difference between the two sides (Fig. 1). The average time difference was 18.5 ms (SD 2.6 ms, $n = 10$ subjects) when tested at the start of the experiment and 17.8 ms (SD 3.3 ms, $n = 10$ subjects) when tested at the end of the experiment. Moreover, close scrutiny of the records revealed that when magnetic stimulation was applied this time difference remained essentially the same (Fig. 1). We could thus confidently use the RT measured from the TA ipsilateral to the magnetic stimulus to determine the RT of the contralateral one when measurement of the latter was obscured by the elicited MEP.

Facilitation of MEPs elicited by magnetic stimuli during the RT. In the RT task starting from tonic voluntary activity, the TA MEPs were facilitated on average 12.5 ms (SD 7.3 ms) before the increase of TA EMG activity. In the example shown in Fig. 2, the MEP elicited at 153 ms during the RT is superimposed on the control MEP elicited during tonic voluntary contraction of the TA. Note the increase of the TA MEP elicited during the RT task, as well as the nearly identical background level of EMG activity between the artifact at 125 ms and the MEP onset 28 ms later. The mean value of the TA EMG in this time interval was 51.8 μV (SD 25 μV) in the RT task and 61.2 μV (SD 17 μV) during tonic voluntary contraction; the difference was not significant (*t*-test, $P > 0.5$). The MEP amplitude increased from 290 μV during tonic voluntary activity to 759.3 μV in the RT task (i.e., 161% increase). The CUSUM of the ipsilateral TA EMG activity, shown at the

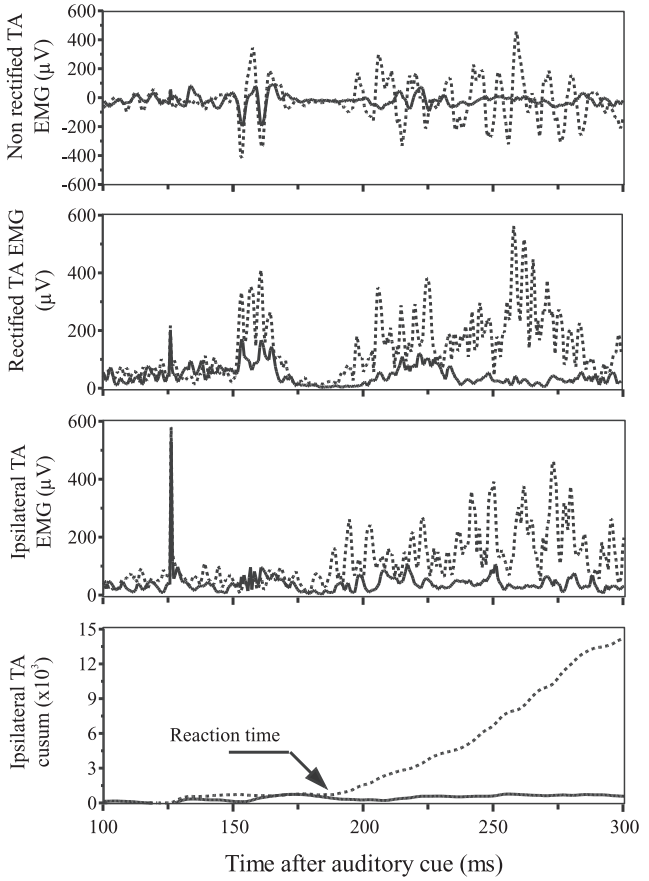

Fig. 2. Example of TA motor-evoked potentials (MEPs; *top* 3 graphs) during tonic voluntary TA activity (continuous traces, mean of 4 trials) and when the subject reacted to the auditory cue by increasing activity starting from a tonic level (dotted traces, mean of 4 trials). The magnetic stimulus intensity was set at the intensity that evokes half-maximal MEP (S_{50}) in both tasks and delivered at the same time (125 ms) after the auditory cue. Note that the TA MEP peak-to-peak amplitude increased from 290.2 to 759.3 μV during the RT task without any significant change in TA EMG background activity (61.2 ± 17 μV during tonic activity vs. 51.8 ± 25 μV during the 20 ms preceding the MEP in the RT task). The RT, estimated from the cumulative sum (cusum) of the EMG activity of the TA muscle ipsilateral to the stimulating coil, is indicated at *bottom*.

bottom of Fig. 2, begins to increase 173 ms after the auditory signal, 22 ms after MEP facilitation.

An example, taken from a single subject, of the time course of changes in MEP amplitude during the RT is shown in Fig. 3*A* along with the time course of the changes in TA EMG activity. Each data point is the average of four trials. The data are aligned with respect to the RT, indicated as *time zero* in the graphs. Note that the MEP increases above the control value < 15 ms before the increase of EMG activity. Figure 3*B* provides a summary of the onset of significant MEP facilitation for all 10 subjects tested (*S1* to *S10*). The onset of facilitation was determined by a two-way ANOVA (subjects and time intervals) followed by Tukey's post hoc test. The data points are expressed in terms of the amount of change from the control value (test MEP − control MEP). Note that the onset of MEP facilitation varied between 2.5 and 25.6 ms before the RT. For the group of subjects, at the onset of facilitation (i.e., 12.5 ms, SD 7.3 ms) the MEP increased on average by 58.5% (SD 11.30%), and it increased by 224% (SD 134%) at the peak of facilitation 4.35 ms before the TA RT (SD 5.57 ms).

The size of the MEPs evoked by a stimulus applied simultaneously with the auditory tone in the RT task was 23% greater than the control MEPs obtained during tonic voluntary activity (*t*-test, $P < 0.005$). The background level of TA EMG activity, however, did not differ significantly between the two tasks (mean 34.91 μV, SD 14.39 μV in the RT task and mean 35.09 μV, SD 13.9 μV during voluntary tonic activity, *t*-test $P > 0.5$).

The onset of MEP facilitation in the RT task starting from rest was 15 ms (SD 8.9 ms) before the onset of TA EMG activity, and the range was between 2 and 26.1 ms. These values did not differ significantly from those obtained in the RT task superimposed on tonic voluntary activity as determined by a two-way ANOVA. The more remarkable observation, however, was that in five subjects MEPs could be as strongly facilitated during the RT in the absence of motor activity, as they were when subjects maintained tonic voluntary activity (Fig. 4). It can be seen in Fig. 4 that MEPs elicited ~10 ms before the RT could reach the same size as MEPs elicited on tonic voluntary activity.

One of the consequences predicted by the nonlinear nature of the MEP input-output curves is that the amount of facilitation is related to MEP amplitude by a bell-shaped relation, having its peak at S_{50} (5, 16). In other words MEPs of half-maximal amplitude should be the most facilitated and those near threshold, or on the plateau of the relation, least. The experimental results we have obtained corroborate these predictions. The MEP input-output curve obtained in one subject during tonic voluntary activity is shown in Fig. 5*A*, with the estimated S_{50} stimulus intensity indicated on the abscissa. In Fig. 5*B* we show the extent of facilitation for MEPs near motor threshold and MEPs of half-maximal amplitude when elicited 20 ms before the RT, respectively. Clearly, the amount of facilitation is greatest for MEPs of half-maximal amplitude. A more complete presentation of the dependence of MEP facilitation on MEP amplitude is shown in Fig. 5*C*. As can be seen, the amount of facilitation is greatest at S_{50} and decreases for weaker and stronger stimuli.

Lack of facilitation of MEPs elicited by anodal stimuli. In two subjects, while they maintained a tonic voluntary activation, anodal stimuli of 1.1 × active motor threshold were applied 12–15 ms before the TA RT. No significant facilitation of the MEPs was observed (Fig. 6, *bottom*). By contrast, magnetically evoked MEPs of comparable size elicited at the same relative time in the RT were facilitated by ~50% (Fig. 6, *top*). In the example shown in Fig. 6, the size of the control MEP evoked by anodal stimulation was 250 μV (SD 87 μV) and the test MEP was 246 μV (SD 96 μV). The size of the control MEP evoked by magnetic stimulation was 249 μV (SD 98 μV) and the test MEP was 374 μV (SD 102 μV). There was no statistical difference in the level of tonic TA EMG activity between conditions. The mean value of the background EMG during magnetic stimulation was 65 μV (SD 23 μV) and 62 μV (SD 20 μV) during anodal stimulation.

DISCUSSION

Three cogent and mutually reinforcing observations were made demonstrating that the size of MEPs depends on the

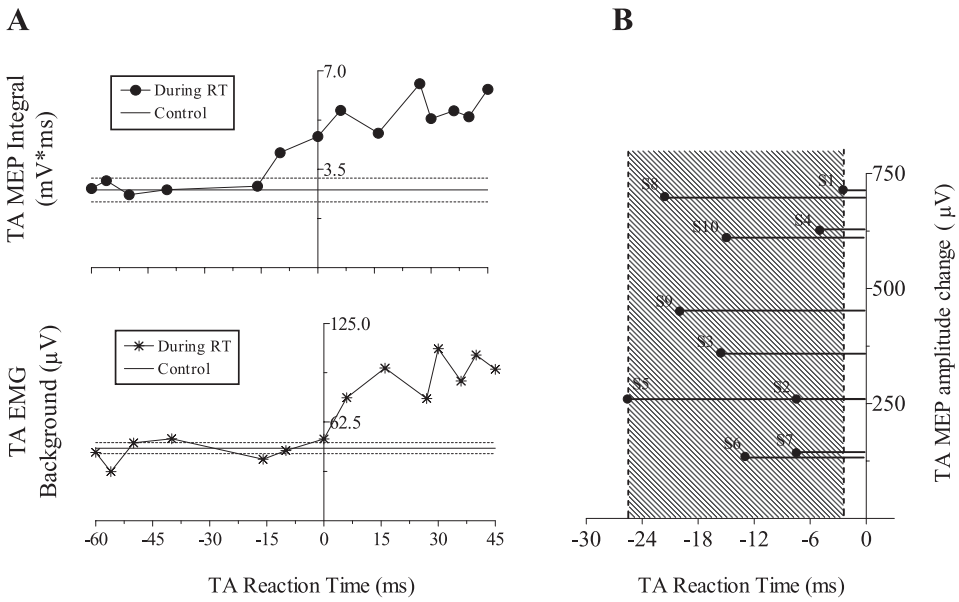

Fig. 3. *A*: time course of TA MEP facilitation in a subject during the RT starting from a tonic level of TA activity. The integral of the rectified TA MEPs is shown at *top* and the background level of TA EMG activity at *bottom*. The origin of the *x*-axis corresponds to the subject's RT determined from the TA ipsilateral to the stimulus and adjusted for the time difference in RTs between the ipsilateral and contralateral TA, as described in the text. The mean value of the control MEP (i.e., when the subject simply maintained a tonic level of activity) is shown as a solid line with the dotted lines representing ±2 SE around the mean. Note that the onset of MEP facilitation occurred <15 ms before RT without any significant change in background EMG activity. *B*: summary of the onset of MEP facilitation for all subjects studied (*S1* to *S10*). The time of occurrence of the first facilitated MEP is plotted against the amount of change relative to the control MEP. Note that the onset of TA MEP facilitation varied across subjects between 2.5 and 25.6 ms (hatched area).

excitability of the MCx at the time of stimulation. First, the size of MEPs increased independently of any change in the level of background motor activity. We have shown that this increase occurs for threshold as well as suprathreshold stimuli and that the amount of change was larger for MEPs of half-maximal amplitude, as predicted theoretically (5, 16). This implies that a task-dependent change in MEP amplitude would be most obvious for stimuli near S_{50}, rather than near threshold as is commonly assumed. This finding extends the preliminary data of Flament et al. (21), suggesting that stronger stimuli resulted in more statistically robust task-dependent differences. Second, when subjects intended to react, MEPs elicited at the time of the auditory cue were 23% greater than those elicited during tonic voluntary activity; i.e., the size of MEPs was greater when subjects intended to react. However, this was only seen when subjects maintained a tonic level of background motor activity. The implications of this finding are dealt with below. Lastly, it was shown that MEPs elicited by near-threshold anodal stimuli were not facilitated during the RT, in marked contrast to MEPs elicited by near-threshold magnetic stimuli.

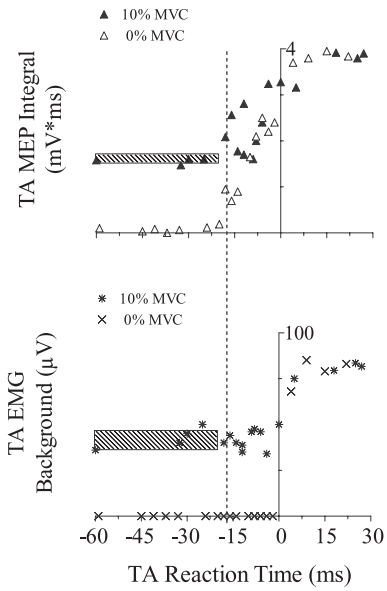

Fig. 4. *Top*: comparison, in a subject, of the onset of MEP facilitation (dashed vertical line) during the RT starting from tonic activity of 10% maximal voluntary contraction (MVC; ▲), or from rest (△). *Bottom*: level of background TA EMG activity. Each of the data points is the mean of 4 trials. Note that close to the RT the size of TA MEPs starting from rest were in some cases comparable to those obtained on a background of tonic activity. Note also that the onset of MEP facilitation is nearly the same in the 2 conditions. Hatched horizontal bars in each graph represent the mean and SE of the control MEP and of the tonic TA EMG activity.

It should be noted that although the studies of Flament et al. (21) and Datta et al. (12) showed task-dependent changes in MEP amplitude independent of changes in EMG activity, the results of these two studies on the 1DI muscle are contradictory. In the study of Datta et al., MEPs were largest during voluntary abduction of the index finger and smallest during fist clenching; the opposite was found by Flament et al., leaving the issue unsettled. We studied a single task under different conditions, whereas the studies on the 1DI involved different tasks. The difference is important because the recruitment gain of the 1DI motor pool may not be the same in different tasks (15, 22). For example, the rank order of motor unit recruitment in the 1DI is not identical in different directions of action (39). The results may thus reflect recruitment gain changes in addition to, or rather than, differences in motor cortical excitability. In our study, we compared the MEPs evoked in the isometrically contracted TA during a simple RT task vs. the MEPs evoked when the same muscle was simply isometrically contracted at the same level of EMG activity. We could thus be confident that we were measuring the output of the same population of α-motoneurons at the same recruitment gain.

An unexpected observation was that the onset of MEP facilitation during the RT was significantly longer than reported in all but two previous papers (13, 27). It is generally reported that in simple RT tasks MEP facilitation commences some 80–100 ms before EMG onset (see Ref. 9 and references therein). These values stand in marked contrast to what we report here (12.8 ms). Consistent with our observation, Davey et al. (13) reported an increase of MEP size occurring on

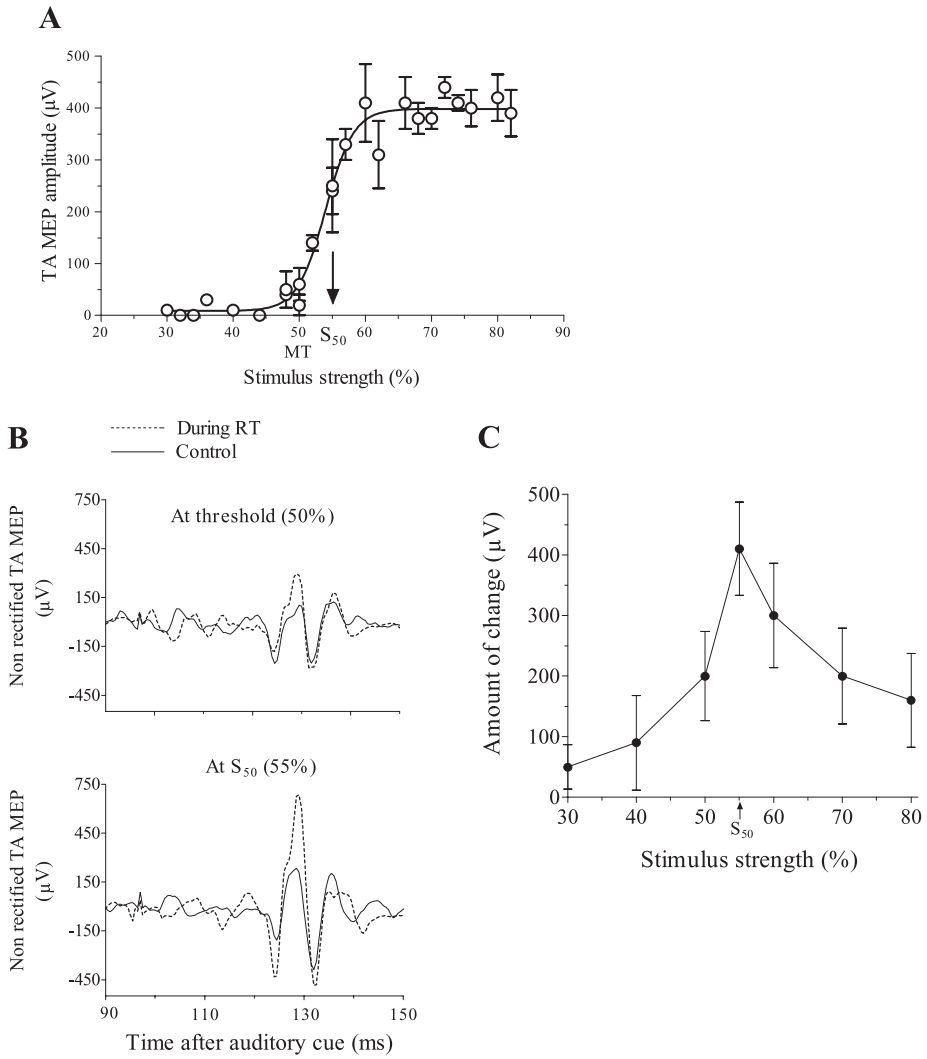

Fig. 5. *A*: example of a MEP input-output curve obtained in 1 subject during tonic activation of TA at 10% MVC. Each symbol represents the mean of 4 TA MEPs ± SD. The data points were fitted with a Boltzmann sigmoid function ($r^2 = 0.99$). The estimated S_{50} stimulus intensity is indicated by the arrow. *B*: for the same subject, TA MEPs in response to a threshold stimulus (*top* graph) and a stimulus of S_{50} intensity (*bottom* graph) during a tonic TA activation. The control MEPs are shown as continuous traces; those obtained at 122 ms during the RT are shown as dashed traces. Note the larger MEP facilitation when stimulated at S_{50}. *C*: plot of the amount of MEP change (test MEP − control MEP obtained at 122 ms in the RT) vs. magnetic stimulus intensity, same subject as in *A*. Note that the maximal MEP change was obtained at S_{50} stimulus intensity.

average some 15 ms before EMG onset, and MacKinnon and Rothwell (27) reported a value of ~10 ms. However, in these studies the movements in response to the auditory cue were made starting from rest. Consequently, the possibility that the MEP increase was attributable to subthreshold depolarization of the α-motoneurons cannot be ruled out (28). For example, in ankle muscles, changes in H-reflex amplitude precede the onset of EMG activity in a simple RT task by some 14 ms (26). Our study and those of Davey et al. and MacKinnon and Rothwell report the actual time of MEP facilitation before the onset of EMG activity rather than the time of stimulation, the two measures may differ by some 30 ms for ankle muscles. Nonetheless, even by taking the conduction time of the MEPs into account, there is a 50–70 ms discrepancy between previous reports and the values reported by Davey et al., MacKinnon and Rothwell, and us. MEPs are very sensitive to changes of the background level of motor activity. Consequently unless the EMG activity immediately before the MEP is closely scrutinized, MEPs may be falsely reported as facilitated, when in fact they are simply enhanced by an increase of α-motoneuron activity. This was also suggested by MacKinnon and Rothwell to explain the discrepancy between their results and those in previous reports.

The very late onset of MEP facilitation during a simple RT task has two possible interpretations with important physiological implications. It may imply that in this task motor cortex excitability increases indeed very shortly before the motor command is dispatched to the motoneurons. This may seem at odds with direct measurements of neural activity preceding movements or EMG activity. Values of ~80–100 ms are often cited in review articles (e.g., Ref. 9). However, close scrutiny of the data reveals that there is in fact a wide range of onset times reported in different studies. For example, when movements are made against a spring load, identified corticomotoneurons discharge some 60 ms before EMG activity in wrist muscles (10). For wrist movements made against an inertial load moved against gravity, most MCx neurons discharge ~60 ms before movement onset (20). For unloaded ballistic elbow movements, MCx neurons discharge ~104 ms before the onset of movement (23, 24). Allowing for a 50–60 ms delay between EMG onset and movement, this implies that neurons began to discharge some 40–50 ms before the onset of EMG. Finger-related corticomotoneurons begin to discharge nearly simultaneously with finger muscle EMG (4, 31). Thus the onset of increased neural activity in MCx before movement may be related to the type of movement and the animal's strategy,

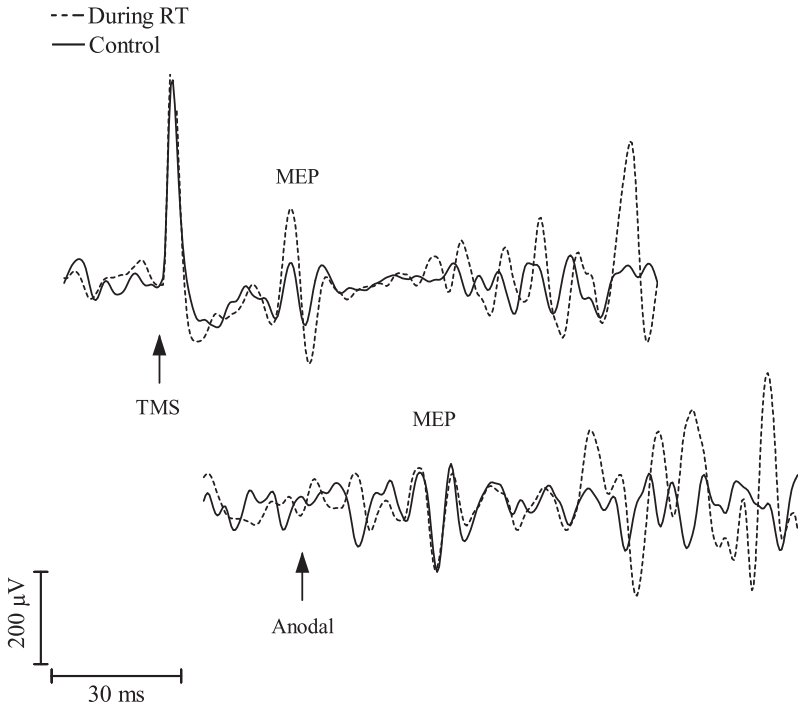

Fig. 6. TA MEPs elicited by a near-threshold magnetic stimulus were facilitated during the RT starting from tonic activity. By contrast, TA MEPs elicited by near-threshold anodal stimuli were not. The arrow under each set of traces indicates the time of stimulation. Each trace represents the mean of 8 trials. The magnetically evoked MEPs occurred 15 ms before the RT, whereas the MEPs evoked by the anodal stimulation occurred 13 ms before the RT (TA RT was determined from the ipsilateral TA, see METHODS and Fig. 1).

which may in turn depend on the load applied and to the mechanical characteristics of the moved limb. There is thus no plain answer to the seemingly simple question of how long do MCx neurons discharge before the movement related EMG activity. Our results and those of Davey et al. (13) and MacKinnon and Rothwell (27) may thus be more accurate measures of the onset of increased corticospinal neuron excitability in the human MCx in a simple RT task. However, in more complex protocols, such as choice or precued RT tasks (e.g., Refs. 29, 34) the time course of cortical excitability may be longer than for simple RT tasks. Furthermore, other neural processes occurring in the MCx, such as changes of intracortical inhibition (33), may lead changes in excitability. The relation between these processes requires further study. Thus the timing of excitability changes is not of necessity a measure of the timing of inputs to the MCx.

An alternative interpretation of the late onset of MEP facilitation during the RT is that MEP amplitude is much more sensitive to the level of α-motoneuron activity than it is to the level of motor cortical neuron activity. This implies that cortical neuron activity needs to be increased significantly more than α-motoneuron activity to affect MEP size. This is because when α-motoneurons are at rest, their membrane potential is relatively far from threshold. By contrast, cortical neurons are spontaneously active even when the subject is at rest. Indeed evidence of spontaneous activity in human MCx was obtained from EEG rhythms as far back as the 1940s (see Ref. 9). Thus the membrane potential of motor cortical neurons is probably on average much closer to threshold than that of the α-motoneurons, even when the subject is at rest. Consequently, the difference between the mean membrane potential and threshold for cortical neurons may not differ between rest and activity by as much as it does for α-motoneurons. In other words, the excitability of cortical neurons increases far less than that of α-motoneurons in going from rest to activity. This idea is consistent with the lack of MEP enhancement tested at the time of presentation of the auditory cue in the RT task starting from rest. In this condition, a potential increase of cortical excitability is masked by the lack of activity of the α-motoneurons. Furthermore, the observation that MEPs elicited in the RT starting from rest could reach the same amplitude as those evoked in the RT superimposed on tonic voluntary activity reinforces the idea. Observations made in previous studies are also consistent with this idea. As mentioned in the introduction, epidural differential recordings of the corticospinal volleys elicited by magnetic stimulation of the hand area of the MCx show that I waves are increased by only 12–15% during contractions of 100% MVC (18). Studies of task-related changes in motor cortical activity have consistently found that the plateau value of the input-output curves is the only parameter that changes significantly and consistently (15, 8). Thus high stimulus intensities, or high levels of activity, appear to best reveal changes of cortical excitability. In any case, the idea that MEPs may be more sensitive to the level of α-motoneuron activity than to that of MCx neurons is worthy of further study.

In summary, in the simple RT task we have studied a measurable increase of MCx excitability that occurs some 12–15 ms before the motor command is issued. Additionally, associated with the intention to move, there occurs a modest increase of excitability, which enhances MEPs by some 23%, but this is only observed when there is ongoing motor activity. We suggest that this activity of the α-motoneurons allows expression of cortical excitability to be reflected in MEP size. The present data also raise the possibility that MEP amplitude is much more sensitive to the level of activity of the α-motoneurons than it is to motor cortical neuron activity. This needs to be a central consideration in the interpretation of experiments aimed at understanding the task-dependent changes of motor cortex activity using magnetic brain stimulation.

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