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RESEARCH ARTICLE

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Spread of electrical activity at cortical level after repetitive magnetic stimulation in normal subjects

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Abstract In normal subjects, focal repetitive transcranial magnetic stimulation (rTMS) of the hand motor area evokes muscle potentials (MEPs) from muscles in the hand (target muscles) and the arm (non-target muscles). In this study we investigated the mechanisms underlying the spread of MEPs induced by focal rTMS in non-target muscles. rTMS was delivered with a Magstim stimulator and a figure-of-eight coil placed over the first dorsal interosseus (FDI) motor area of the left hemisphere. Trains of 10 stimuli were given at a suprathreshold intensity (120% of motor threshold) and at frequencies of 5, 10 and 20 Hz at rest. Electromyographic (EMG) activity was recorded simultaneously from the FDI (target muscle) and the contralateral biceps muscle and from the FDI muscle ipsilateral to the side of stimulation (non-target muscle). rTMS delivered in trains to the FDI motor area of the left hemisphere elicited MEPs in the contralateral FDI (target muscle) that gradually increased in amplitude over the course of the train. Focal rTMS trains also induced MEPs in the contralateral biceps (non-target muscle) but did so only after the second or third stimulus; like target-muscle MEPs, in non-target muscle MEPs progressively increased in amplitude during the train. At no frequency did rTMS elicit MEPs in the FDI muscle ipsilateral to the site of stimulation. rTMS left the latency of EMG responses in the FDI and biceps muscles unchanged during the trains of stimuli. The latency of biceps MEPs was longer after rTMS than after a single TMS pulse. In conditioning-test experiments designed to investigate the cortical origin of the spread, a single TMS pulse delivered over the left hemisphere at an interstimulus interval (ISI) of 50, 100 and 150 ms reduced the amplitude of the test MEP evoked by a single TMS pulse delivered over the right hemisphere; and a conditioning rTMS train delivered over the left hemisphere increased the amplitude of the test MEP evoked by a single TMS pulse over the right hemisphere. A conditioning rTMS train delivered over the left hemisphere and paired magnetic shocks (test stimulus) at 3 and 13 ms ISIs over the right hemisphere reduced MEP inhibition at the 3-ms ISI but left the MEP facilitation at 13 ms unchanged. Using a control MEP size matched with that observed after a conditioning contralateral rTMS, we found that paired-pulse inhibition remained unchanged. Yet a single TMS conditioning pulse sufficiently strong to evoke a MEP in the contralateral FDI and biceps muscles simultaneously (as rTMS did) left paired-pulse inhibition unchanged. We conclude that the spread of EMG activity to non-target muscles depends on cortical mechanisms, mainly including changes in the excitability of the interneurones mediating intracortical inhibition.

Keywords Repetitive magnetic transcranial stimulation · Spread of cortical activation

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Introduction

Repetitive transcranial magnetic stimulation (rTMS) is a neurophysiological technique suitable for studying cortical motor area excitability in humans. It entails repetitive activation of cortical motor areas to elicit muscle evoked potentials (MEPs) in target muscles (Pascual-Leone et al. 1994; Jennum et al. 1995; Berardelli et al. 1998).

Some evidence indicates that rTMS elicits MEPs also in non-target muscles. In a study conducted several years ago in healthy subjects, Pascual-Leone et al. (1994) observed that trains of 10 stimuli delivered at various intensities (110%–220% of motor threshold) and frequencies (1, 3, 5, 10, 20 and 25 Hz) to the optimal scalp position for activating the contralateral abductor pollicis

brevis (target muscle) elicited MEPs also in the nearby muscles (non-target muscles). They attributed the emergence of MEPs in non-target (proximal) muscles during rTMS to changes in proprioceptive inflow caused by the repeated muscle twitches that alters segmental spinal motoneuron excitability or to lateral spread of excitation in the motor cortex. Whether electrical activity induced by rTMS spreads to non-target muscles through spinal or cortical mechanisms remains unclear. Changes of cortical excitability during and after the emergence of MEPs in non-target muscles (proximal) would support the hypothesis that cortical mechanisms play a role in the MEPs spread to non-target muscles.

A precise knowledge of the mechanisms underlying stimulus spread in healthy subjects is essential for studying the cortical spread of electrical activity in epilepsy, myoclonus and similar neurological conditions. It would also be useful in developing a neurophysiological method for investigating drug-induced effects on cortical motor excitability.

Prompted by the lack of further research to extend Pascual-Leone's findings (1994), we investigated in this study conducted in a group of healthy volunteers the physiological mechanisms underlying spread of excitation to non-target muscles. To minimize the potential risk of rTMS-induced, followed published safety guidelines (Pascual-Leone et al. 1993, 1994; Wassermann 1998). We therefore examined the effects of short-duration trains given at an intensity just above the motor threshold and at frequencies of 5, 10 and 20 Hz over the hand and arm motor area.

Methods

Subjects

We studied a total of 27 healthy subjects (18 women and 9 men) aged 25–45 years (mean 31.5±0.1 years). All of them gave their informed consent. They were asked to report adverse effects experienced during or after rTMS: abnormalities of consciousness, headache, paraesthesias, visual disturbances, vertigo, hearing loss, tinnitus and weakness. Stimulation was also immediately interrupted if afterdischarges were recorded at the end of the train.

The study was conducted by neurologists who were familiar with the rTMS technique and with the treatment of seizures and was approved by the Local Ethical Committee.

Stimulation

rTMS was delivered with a Magstim repetitive magnetic stimulator in subjects at rest. A figure-of-eight coil (8 cm outer diameter) was placed over the left hemisphere to determine the optimal position for activating the contralateral first dorsal interosseus (FDI) muscle (target muscle). Eight trains of 10 stimuli were delivered at frequencies of 5, 10 and 20 Hz. Stimulation intensity was set at 120% of the motor threshold. The motor threshold (Mth) was calculated at rest using the lowest intensity able to evoke a MEP of more than 50 μV in at least five of ten consecutive trials in FDI and biceps muscles. The inter-train interval was 2 min. Participants were asked to remain relaxed during the study.

Single stimulation was given with a Magstim 200 stimulator through a figure-of-eight coil, and paired magnetic stimuli were delivered through two magnetic stimulators connected to the figure-of-eight coil by a Bistim module.

Recording

EMG signals were recorded and filtered with a Digitimer D 360 (bandwidth 5 Hz–1 kHz) and a personal computer through a 1401 plus AD laboratory interface (Cambridge Electronic Design) and analyzed off-line. The electromyographic (EMG) activity was recorded through a pair of surface disk electrodes placed over the contralateral FDI and biceps muscles and over the FDI muscle ipsilateral to the motor area of stimulation.

Experimental paradigms

Trains of 10 stimuli were delivered to the FDI motor area of the left hemisphere at frequencies of 5, 10 and 20 Hz. An optimal scalp site was found for obtaining a response in the FDI (target muscle) alone, not in the biceps muscle (non-target muscle). EMG activity was simultaneously recorded from the contralateral FDI and biceps and from the FDI ipsilateral to the site of stimulation. This experiment was conducted in six subjects.

In five subjects, single TMS pulses were delivered at rest over the biceps motor area of the left hemisphere at 120% of biceps Mth intensity.

Conditioning–test experiments

In the first experiment, a conditioning stimulus was applied to the left hemisphere, and a test stimulus was applied to the right hemisphere ('transcallosal inhibition') (Ferbert et al. 1992; Gerloff et al. 1998; Di Lazzaro et al. 1999). Conditioning and test stimuli were given at the optimal scalp site for evoking responses in their respective contralateral FDI muscle. The intensity of stimulation for both the conditioning and test stimuli was 120% of Mth at rest. The conditioning–test interstimulus intervals were 50, 100 and 150 ms. The same conditioning–test experiment was also designed to investigate rTMS-induced changes in transcallosal inhibition.The conditioning stimulus consisted of a train of 10 magnetic rTMS pulses delivered over the left hemisphere at a frequency of 20 Hz, and the test stimulus was a single magnetic stimulus delivered with a Magstim 200 through a figure-of-eight coil placed over the FDI motor area of the right hemisphere. The intensity of stimulation for both the conditioning and test stimulus was 120% of the motor threshold at rest. The conditioning–test interstimulus interval (ISI) was 50 ms.

The second experiment was conducted to investigate the changes in intracortical inhibition (Kujirai et al. 1993) of the contralateral hemisphere after rTMS. The conditioning stimulus was a train of 10 magnetic stimuli delivered over the left hemisphere at a frequency of 20 Hz and at an intensity of stimulation of 120% of Mth at rest. The test stimulus consisted of paired magnetic pulses (Kujirai et al. 1993) delivered by two Magstim 200 magnetic stimulators connected through a Bistim module to a figure-of-eight coil positioned over the FDI motor area of the right hemisphere. Paired stimulation was given at rest with ISIs of 3 and 13 ms. The intensity of the conditioning stimulus was set at 80% of Mth and the test stimulus at 120% of Mth. The conditioning–test ISI was 50 ms. Paired-pulse inhibition was also evaluated by matching the size of the control MEP with that of the MEP after a conditioning train. In two of the four subjects who participated in this experiment, the conditioning stimulus was a train of 10 magnetic stimuli delivered at a frequency of 20 Hz and an intensity lower than that eliciting MEPs in non-target muscles.

In the third experiment, the conditioning stimulus was a single TMS pulse delivered over the optimal position for evoking responses in the right FDI muscle and sufficiently strong to evoke a simultaneous MEP in the right biceps muscle (stimulation intensity 150% of the motor threshold). The test stimulus consisted

of a single pulse, and paired TMS pulses delivered over the FDI motor area of the right hemisphere at ISIs of 3 ms. The conditioning–test ISI was 50 ms.

The EMG activity was recorded from the right and left FDI muscles and from the right biceps muscles. For all the experiments eight trials were collected and averaged. The various conditions of stimulation were randomly delivered. Each of these experiments was conducted in four subjects.

Measurements

The size of MEPs evoked by magnetic stimulation during rTMS was measured peak-to-peak and expressed as a percentage of the first MEP in the train. The presence of MEPs throughout the train of stimuli was evaluated by visual inspection as follows. First, we verified the optimal scalp position for evoking a MEP in the target muscle (FDI) alone and not in the non-target muscle, and a MEP facilitation over the course of the train. Then in the biceps muscle contralateral to the FDI motor area stimulated, we evaluated the presence of MEPs after the second or ensuing stimuli and the gradual increase in the amplitude of the MEPs over the course of the train. MEP latency was measured in the biceps muscle and FDI. In the biceps muscle, we also compared the latency of MEPs evoked by single TMS pulses delivered over the biceps motor area and by rTMS at 20 Hz.

In the conditioning–test paradigm the amplitude of the conditioned MEPs was measured from peak to peak (mV) and expressed as a percentage of the unconditioned MEPs.

Statistical analysis

For each frequency of stimulation, differences among MEP sizes and latencies were tested with a multivariate two-way ANOVA, with repeated measures for the factors frequency and the number of stimuli. Student's two-tailed *t*-test for paired samples was used to compare the intensity of Mth values at rest in the FDI (target muscle) and biceps muscles at the optimal stimulation site for the FDI muscle, and the latency of biceps MEPs elicited by single TMS pulses and by rTMS. In the conditioning–test paradigm, Student's two-tailed *t*-test was also used to compare the amplitude of the test MEP vs control MEP. All results were expressed as mean±SE; p values <0.05 were considered to indicate statistical significance.

Results

None of the rTMS variables used caused any of the participants to experience adverse effects (seizures, headache, paraesthesias, vertigo, weakness, visual disturbance or other bodily sensations). In three subjects, afterdischarges were recorded at the end of the train of stimuli delivered at the frequency of 20 Hz, and therefore repetitive stimulation was immediately interrupted.

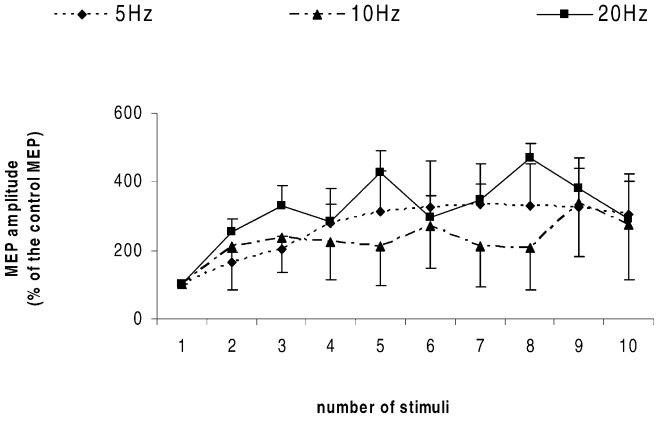

Fig. 1 Effects of repetitive transcranial magnetic stimulation (rTMS) at frequencies of 5, 10 and 20 Hz and 120% motor threshold (Mth) intensity on the amplitude of motor evoked potentials (MEPs) in the first dorsal interosseus (FDI) muscle. Data correspond to means±SE in %. Note the progressive increase in the amplitude of the MEP at all frequencies studied

MEPs in the FDI and biceps muscles

The resting Mth for the FDI and biceps muscles at the optimal stimulation site for the FDI muscle was significantly lower in the FDI than in the biceps muscle (FDI muscle 51.6%±2.8%; biceps muscle 68.2%±2.7%; $p<0.0001$, by Student's two-tailed *t*-test). The resting Mth for the biceps muscle at the optimal stimulation site for the biceps muscle was 68.0%±2.2%. The latency of the MEPs in the FDI and biceps muscles remained unchanged during the course of the train (FDI muscle, F for factor number of stimuli =0.29, $p=0.95$; F for factor frequency =2.76, $p=0.26$; biceps muscle, F for factor number of stimuli =3.2, $p=0.2$; F for factor frequency =0.6, $p=0.5$) (Table 1). In the biceps muscle, the latencies of MEPs evoked by single TMS pulses were shorter than those evoked by rTMS at the optimal stimulation site for the FDI muscle (13.2±0.8 ms vs 14.7±0.6 ms; $p=0.04$, by Student's two-tailed *t*-test). The latency of biceps muscle MEPs remained unchanged during the course of the train (onset latency of the last MEP vs first MEP: 14.7±0.6 ms vs 14.6±0.5 ms; $p=0.12$).

Table 1 Latency of the first and the last motor evoked potentials (MEPs) in the first dorsal interosseus and biceps muscles during spread of excitation produced by repetitive transcranial stimulation at all frequencies studied. All data correspond to means±SE

Muscles	Frequencies					
	5 Hz		10 Hz		20 Hz	
	First MEP	Last MEP	First MEP	Last MEP	First MEP	Last MEP
	(ms)	(ms)	(ms)	(ms)	(ms)	(ms)
First dorsal interosseus	21.1±0.56	21.5±1.16	21.8±0.73	21.6±0.58	21.7±0.41	22±0.22
Biceps	11.16±1.3	11.2±2.2	11.6±0	10.19±1	11.4±2.3	12.0±0.4

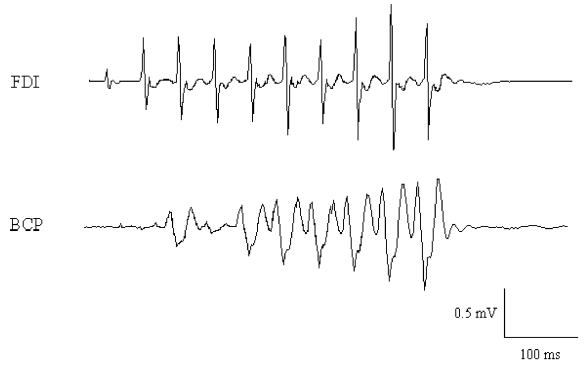

Fig. 2 Effects of 20 Hz rTMS on the amplitude of the motor evoked potential (MEP) in the FDI and biceps muscles in a representative subject. Each trace is the average of eight single electromyographic trials. Horizontal calibration is 100 ms, and vertical calibration is 0.5 mV. Note the facilitation of the MEP amplitude during the train of stimuli in the FDI. In the biceps muscle the MEP appeared only after the second stimulus in the train and progressively increased in amplitude during the course of the train

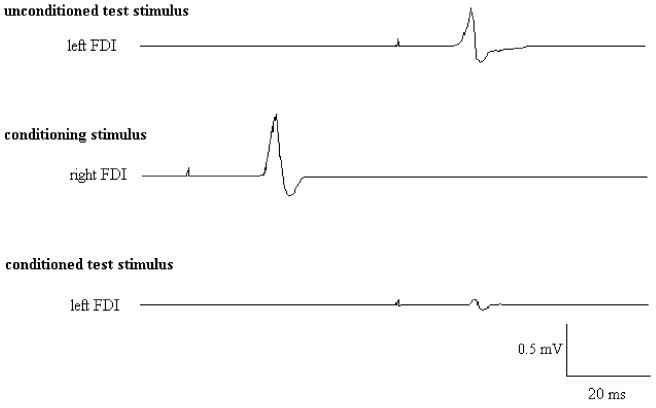

Fig. 3 Effect of a single TMS conditioning pulse delivered over the FDI motor area of the left hemisphere on the amplitude of the test MEP elicited by a single pulse delivered over the right hemisphere at 50 ms interstimulus interval in a representative subject. Each trace is the average of eight single electromyographic trials. Horizontal calibration is 20 ms, and vertical calibration is 0.5 mV. Note the significant reduction in amplitude of the test MEP

rTMS stimulation delivered over the FDI motor area

rTMS delivered at frequencies of 5, 10 and 20 Hz to the FDI motor area of the left hemisphere produced a MEP in the contralateral FDI muscle that gradually increased in amplitude over the course of the train (last MEP vs first MEP: 304%±11.7% at 5 Hz, 275%±16.3% at 10 Hz and 287%±11.2% at 20 Hz) (Fig. 1). ANOVA showed a main effect for the factor number of stimuli and not for the factor frequency (F for factor number of stimuli =4.1; p=0.0006; F for factor frequency =0.24, p=0.78).

The first stimulus in the rTMS train delivered to the FDI motor area of the left hemisphere did not produce a MEP in the biceps muscle. With rTMS delivered at a frequency of 5 Hz, the second stimulus of the rTMS train elicited a biceps muscle MEP in five subjects studied, and the third stimulus elicited a MEP in one subject. At 10 Hz frequency, the second stimulus elicited a MEP in three subjects, and the third stimulus elicited a MEP in three subjects. At 20 Hz frequency, the second stimulus elicited a MEP in four of the six subjects and the third stimulus, in two subjects. MEPs recorded in the biceps increased significantly in amplitude over the course of the train (last MEP vs first MEP: 217%±60% at 5 Hz, 173%±62% at 10 Hz and 534%±272% at 20 Hz). ANOVA showed a main effect for the factor number of stimuli and frequency (F for factor number of stimuli =4.23, p=0.00029; F for factor frequency =4.6; p=0.037); the interaction between number of stimuli and frequency was not significant (p=0.07) (Fig. 2).

A simultaneous EMG recording from the contralateral FDI and biceps muscles and from the ipsilateral FDI muscle showed that rTMS elicited no MEPs in the FDI ipsilateral to the side of stimulation.

Conditioning–test experiments

In the first experiment, a single TMS conditioning pulse significantly reduced the amplitude of the test MEP elicited by a single TMS pulse delivered over the right hemisphere at 50, 100 and 150 ms ISIs (test MEP vs control MEP: 50.8%±7.22% at the 50 ms ISI; 59%±10.5% at 100 ms; and 42.1%±10.4% at 150 ms (F=14.4; p=0.01). At all ISIs studied, the conditioning stimulus induced similar inhibitory effects on the test MEP (F=0.76; p=0.49) (Fig. 3). rTMS delivered to the FDI motor area of the left hemisphere significantly increased the size of the contralateral MEP in the FDI. After the second or third stimulus in the train, it also elicited MEPs in the biceps muscle. These responses gradually increased in amplitude over the course of the train (last MEP 419.16%±91.5% in the FDI muscle and 1236.7%±375.7% in biceps muscle, p=0.001). Conditioning rTMS significantly increased the size of the test MEP evoked in the FDI muscle by stimulating the right hemisphere (test MEP vs control MEP: 548.5%±276%; p=0.04, by Student's two-tailed t-test).

In the second experiment, when the paired shocks were not conditioned by rTMS, the test MEPs were significantly inhibited at the 3 ms ISIs (p=0.001 Student's two-tailed t-test) and facilitated at 13 ms ISIs (p=0.04). In contrast, when paired shocks were conditioned by an rTMS train, there was less inhibition at 3 ms ISIs (conditioned–test MEP vs unconditioned–test MEP 82%±51% vs 18%±11.2%; p=0.0026 by Student's two-tailed t-test) and a similar facilitation at 13 ms ISIs (conditioned–test MEP vs unconditioned–test MEP 148.5%±56% vs 153%±47.4%; p=0.7 by Student's two-tailed t-test) (Fig. 4). When the size of the control MEP matched that observed after a conditioning train, the test MEP at the 3 ms ISI was significantly inhibited (19%±10.8%; p=0.01 by Student's two-tailed t-test). In

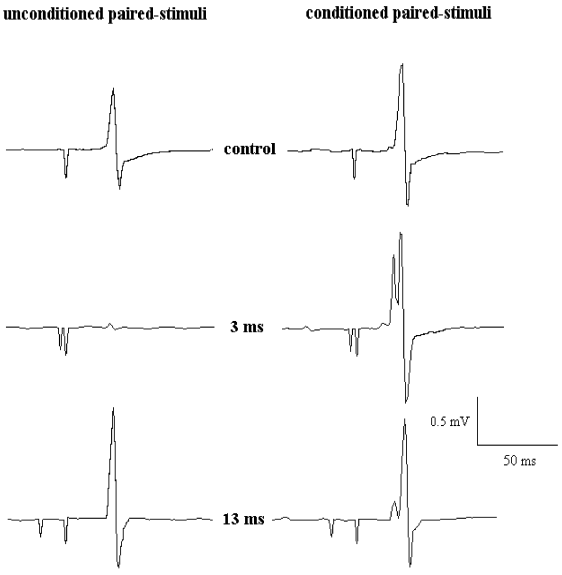

Fig. 4 Effect of 20 Hz rTMS delivered over the FDI motor area of the left hemisphere on intracortical inhibition (paired stimulation at the 3 ms ISI) and facilitation (paired stimulation at the 13 ms ISI) in a representative subject. Each trace is the average of eight single electromyographic trials. Horizontal calibration is 50 ms, and vertical calibration is 0.5 mV. When the paired shocks were not conditioned by rTMS, the test MEPs were significantly inhibited at the 3 ms ISI and facilitated at 13 ms, whereas when paired shocks were conditioned by an rTMS train, there was less inhibition at 3 ms ISIs and a similar facilitation at the 13 ms ISI

the two subjects studied with rTMS at a lower intensity than that needed to obtain the facilitation and spread but able to evoke responses in the target muscle (last MEP vs first MEP: 71%±0.05% in the first subject; 76%±0.09% in the second), conditioning rTMS left paired-pulse inhibition in the contralateral cortical FDI area unchanged (at the 3 ms ISI, conditioned–test MEP vs unconditioned–test MEP 41.9%±18% vs 35%± 11%; p=0.54 by Student's two-tailed t-test, in the first subject; and 17.4%±6.7% vs 25%±4.7%; p=0.22, in the second; and at the 13 ms ISI, conditioned–test MEP vs unconditioned–test MEP 158%±77% vs 159%±105%; p=0.77 in the first subject; and 133%±45% vs 130%±24%; p=0.87 in the second).

In the third experiment, the single conditioning TMS pulse was sufficiently strong to evoke MEPs simultaneously in the FDI and biceps muscles. The test MEPs evoked by a single TMS pulse over the right hemisphere at 50 ms ISIs were significantly reduced in size (35.6%±17.7%; p=0.01 by Student's two-tailed t-test). The test MEPs evoked by paired-pulse delivered over the right hemisphere at 3 ms ISIs were significantly inhibited (9.1%±0.6%; p=0.01 by Student's two-tailed t-test).

Discussion

In the healthy volunteers we studied, rTMS delivered to the FDI motor area of the left hemisphere in short trains at safe frequencies and at an intensity just above Mth facilitated MEPs in the target muscle (FDI); caused spread of excitation to the non-target muscle (biceps); and elicited marked changes in transcallosal inhibition and in intracortical excitability of the contralateral hemisphere. These findings suggest that cortical excitation spreads to non-target muscles through cortical mechanisms, probably by altering the excitability of the intracortical interneurons.

Overall, our findings along with current knowledge exclude stimulus spread to non-target muscles through spinal mechanisms. First, rTMS left the latency of EMG responses in the FDI and biceps muscles unchanged during the trains of stimuli. If expectation had spread through spinal mechanisms, muscle twitches in the target muscle after rTMS would have altered the segmental proprioceptive inflow, thereby altering spinal motoneuron excitability (Calancie et al. 1987). It would therefore have elicited EMG responses at a shorter latency during rTMS (Benecke et al. 1988; Rothwell et al. 1987; Siggelkow et al. 1999). In addition, biceps responses evoked by rTMS delivered over the FDI motor area of the left hemisphere had a longer latency than biceps responses evoked by TMS delivered at rest over the biceps motor area of the left hemisphere. The longer latency probably depends on intracortical delay rather than spinal mechanisms (Pascual-Leone et al. 1994). Moreover, because the proprioceptive afferent fibres do not cross contralaterally (Kossev et al. 2001), altered proprioceptive inflow would not explain why rTMS facilitated the size of the MEP elicited by a single TMS pulse delivered to the contralateral hemisphere.

Our findings raise the question of how excitation spreads through the cortex to non-target muscles. One explanation is that rTMS at high frequencies increases cortical excitability through non-synaptic mechanisms of electrical fields (Nitsche and Paulus 2001). Yet this local mechanism would not explain why rTMS increased the size of the contralateral MEP.

A further hypothesis is that rTMS could in theory increase cortical excitability, resulting in facilitation of the I-waves induced by the ensuing stimuli (Jankowska et al. 1975; Pascual-Leone et al. 1994). Using epidural recordings and the electrical paired-pulse technique, Inghilleri et al. (1989) observed no I-wave facilitation at ISIs longer than 3.5 ms. Neither would an eventual I-wave facilitation explain why rTMS increased the size of the contralateral MEP.

Previous investigators (Ferbert et al. 1992; Gerloff et al. 1998; Di Lazzaro et al. 1999) found that a single conditioning magnetic stimulus over the motor cortex of one hemisphere reduces the size of the MEP in a distal hand muscle induced by a magnetic test stimulus applied over the opposite hemisphere at various ISIs (from 6 to 50 ms). Current knowledge suggests that the inhibition of a response conditioned by a contralateral shock is mediated through a cortico-cortical pathway through the corpus callosum. Accordingly, Meyer et al. (1995) showed that this inhibition is absent in patients with focal lesions of the corpus callosum. Others then demon-

strated reduced transcallosal inhibition in patients with cortical myoclonus (Brown et al. 1991, 1996; Hanajima et al. 2001).

In our conditioning–test experiments testing transcallosal inhibition, rTMS facilitated the size of the MEP elicited by a single magnetic stimulus delivered to the contralateral hemisphere. In contrast, as previously reported (Ferbert et al. 1992; Gerloff et al. 1998; Di Lazzaro et al. 1999), a single conditioning TMS pulse delivered over the left motor cortex significantly inhibited the response to a test stimulus given over the right motor cortex at all ISIs studied (50, 100 and 150 ms). Hence, unlike a single TMS pulse, rTMS could activate a larger cortical area, thus altering transcallosal input to the contralateral cortex. Yet it clearly did not do so because a single TMS conditioning stimulus sufficiently strong to evoke a MEP in the contralateral FDI and biceps muscles simultaneously (as rTMS did) elicited transcallosal inhibition rather than facilitation. In our experiment the facilitation of the MEP produced by the contralateral rTMS therefore suggests that when MEPs spread to non-target muscles, the cortical motor area excitability increases.

The second conditioning–test experiments strongly suggest that rTMS elicits MEPs in non-target muscles by activating the intracortical interneurons. It does so either by increasing the activity of excitatory interneurons or by reducing the activity of inhibitory interneurones. The paired stimulation technique at 3 ms and 13 ms ISIs investigates the excitability of intracortical inhibitory and excitatory interneurones (Kujirai et al. 1993; Ziemann et al. 1996). In all the healthy subjects we tested, rTMS delivered to the left motor cortex significantly reduced the inhibition at the 3 ms ISI but left the facilitation at the 13 ms ISI on the opposite hemisphere unchanged. A conditioning rTMS train reduced intracortical inhibition, leaving the intracortical facilitation unaltered. This selective change suggests that as MEPs spread to non-target muscles, the inhibitory action of intracortical interneurones diminishes.

A further possibility is that the MEP facilitation (1st conditioning–test experiment) or reduction of inhibition (2nd conditioning–test experiment) could be generated in the contralateral right hemisphere by one of the stimuli preceding the 10th stimulus of the conditioning train (the 8th or 9th stimulus). At 20 Hz, these stimuli precede the contralateral test stimulus by 150 and 100 ms. In the first conditioning–test paradigm (single test stimulus/single conditioning stimulus), a single conditioning TMS pulse given 50, 100 and 150 ms before the test stimulus inhibited the test MEP. These results also suggest that rTMS facilitates the MEP by exciting cortical motor areas.

Evidence that intracortical inhibition and muscle spread change simultaneously does not of course necessarily imply that the two are causally related. Because larger test responses are generally associated with less inhibition, the diminished inhibitory intracortical effect could depend on an increased size of the conditioned control MEP. Using a MEP size matched with that observed after a conditioning contralateral rTMS, however, we found that the paired-pulse inhibition remained unchanged. Yet a single TMS conditioning pulse sufficiently strong to evoke a MEP in the contralateral FDI and biceps muscles simultaneously (as rTMS did) left the paired-pulse inhibition unchanged. These results imply that the spread of excitation and changes in the excitability of the contralateral hemisphere are related.

The emergence of MEPs in non-target muscles always coincided with changes in intracortical inhibition. In the two subjects in whom rTMS elicited no MEP facilitation in the target muscle and no MEPs in non-target muscles, the intracortical inhibition remained unchanged. Hence, rTMS presumably activates intracortical excitatory interneurons by concurrently facilitating MEPs in the target muscle and inducing spread of excitation to non-target muscles.

The muscle spread we observed could represent the recruitment of higher threshold outputs due to an overlapping mosaic in the cortical spatial distribution of single muscle/single column in the muscle target. Microstimulation studies in animals (Asanuma and Rosén 1972; Asanuma 1975) showed that the hand motor cortex is organized as a mosaic of overlapping motor columns, activating only a single or a few distal forelimb muscles. The overlapping of columns is especially valid for distal limb muscles (Asanuma and Rosén 1972; Asanuma 1975). In humans, studies using TMS to map the cortical representation of muscles have shown a somatotopic progression on the scalp of proximal to distal muscles along a posteromedial to anterolateral axis (Cohen et al. 1991; Wassermann et al. 1992). Although the areas for the various limb muscles overlapped, the areas for distal muscles tended to be more lateral than those for proximal muscles, and the maps of distal and proximal muscles differed significantly in area, threshold and volume. Because the zones projecting to proximal muscles were located more rostrally than those projecting to distal muscles, we consider overlapping mosaics less valid for the biceps than for the hand muscles. Hence 'muscle spread' seems unlikely to represent recruitment of higher threshold outputs alone. Our findings also indicate that rather than simply producing a physical spread of the electric field to more distantly located motor cortical areas, rTMS caused a spread by activating the cortico-cortical axons (Amassian et al. 1991). Finally, the experiments testing the transcallosal inhibition demonstrated a contralateral facilitation that cannot be explained by an overlapping mosaic at the cortical level.

In conclusion, rTMS delivered in short-lasting trains at an intensity just above threshold and at a frequency ranging from 5 to 20 Hz recruits MEPs in non-target muscles of the same limb. The spread of EMG activity possibly depends on a cortical mechanism that involves changes in the excitability of the interneurones mediating cortical inhibitory mechanisms in the arm cortical motor areas.

The interest of our findings lies also in the potential use of MEP recordings in target and non-target muscles of the upper limb as a neurophysiological method for studying the spread of electrical activity at the cortical level in neurological conditions such as epilepsy and myoclonus. MEP recordings may also be useful for testing the effects of drugs acting on the excitability of cortical motor areas.

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