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Intensity-dependent effects of 1 Hz rTMS on human corticospinal excitability

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Abstract

Objectives: This study explored whether the effects of repetitive transcranial magnetic stimulation (rTMS) on corticospinal excitability are dependent on the stimulation intensity and examined the effect of rTMS on inhibitory function.

Methods: Nine normal volunteers received 15 min of 1 Hz rTMS at 85 and 115% of the resting motor threshold (RMT). Cortical excitability was measured before and after rTMS.

Results: rTMS at both intensities produced an increase in the RMT but only 115% stimulation reduced the size of motor evoked potentials (MEPs). rTMS had no effects on the cortical silent period or cortical inhibition measured with paired pulse TMS.

Conclusions: The effects of 1 Hz rTMS on motor cortex excitability are partially dependent on stimulus intensity and the effects of rTMS on motor thresholds and MEP size may differ.

Keywords: Repetitive transcranial magnetic stimulation; Motor excitability; Motor evoked potential; Cortical stimulation; Human

1. Introduction

Repetitive transcranial magnetic stimulation (rTMS) has recently gained considerable attention as a possible treatment for a number of neuropsychiatric disorders including major depression. However, the techniques by which rTMS alters brain activity and the optimal methods for maximising the response to stimulation remain to be elucidated. The effects of low frequency (0.9–1.0 Hz) stimulation is of particular interest as this form of stimulation has therapeutic potential in a variety of disease states including depression, schizophrenia, seizure disorders and writer's cramp (Klein et al., 1999; Siebner et al., 1999; Tergau et al., 1999; Hoffman et al., 2000). As low frequency stimulation can be safely applied through a range of stimulation settings, such as intensity, there is a considerable choice in establishing experimental protocols. There is limited information to guide the determination of these protocols.

Considerable knowledge of the brain's response to rTMS comes from studies of the effects of repetitive stimulation applied to the motor cortex. These studies have applied rTMS of varying duration and frequency to the motor cortex and measured its effect on a variety of electrophysiological variables, including single and paired pulse TMS (ppTMS). These studies have used both low and high frequency (5–20 Hz) stimulation. The main effect of low frequency rTMS is a reduction in motor evoked potential (MEP) size and an increase in motor threshold. These effects appear robustly with prolonged (15 min) supra-threshold stimulation (Chen et al., 1997; Tergau et al., 1997; Muellbacher et al., 2000) and appear less consistently with shorter trains of stimulation (Maeda et al., 2000). With very short stimulation trains at 1 Hz, stimulation does not appear to affect the cortical silent period (SP; Romeo et al., 2000). Another study found a trend towards increased cortical inhibition (CI) as measured with ppTMS but the train length was not specified (Tergau et al., 1997).

The aim of this study was to further investigate the response of the motor cortex to low frequency stimulation. The primary objective was to investigate whether TMS effects on cortical excitability are dependent on the intensity of stimulation when applied for a prolonged duration (15 min) to the motor cortex. A secondary objective was to investigate the effects of low frequency stimulation on the CSP, CI and CF as measured with ppTMS.

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2. Methods

2.1. Subjects

Nine normal subjects were studied (age 25–44, mean 32.9 ± 6.4, one female). The Human Subject and Ethics Committee of Southern Health, Dandenong Hospital approved the study and all subjects gave written informed consent.

2.2. Study procedure

All subjects were studied twice, at least 1 week apart with the order of the experiments randomly assigned. In one experiment rTMS was applied at 115% of the resting motor threshold (RMT) and in the other experiment at 85% of the RMT. In each experiment, several aspects of cortical excitability were measured before and immediately after, 15 min of rTMS applied to the motor cortex. Measures of excitability included the resting and active motor thresholds (AMTs), the MEP size evoked with supra-threshold stimulation, the CSP and CI and CF measured with ppTMS.

2.3. Electromyographic (EMG) recordings

EMG was recorded from the right abductor pollicis brevis (APB) muscle using self-adhesive electrodes (Medtronic). One electrode was placed over the muscle belly and another on the dorsal aspect of the interphalangeal joint of the thumb. An earth electrode was placed on the mid forearm. All EMG signals were amplified and filtered (low pass 2.4 kHz, high pass 10 Hz). They were sampled (rate 10 kHz) using a Digidata 1320A Data Acquisition board and pCLAMP 8.0 software (Axon Technologies) run on a PIII 450 MHz Hewlett Packard PC. EMG recordings were made at rest and during sustained contraction with visual display feedback.

2.4. Transcranial magnetic stimulation

Subjects were seated in a reclining chair with a headrest for stabilisation of the head. Single and paired pulse stimulation was administered with a figure-of-8 coil (70 mm diameter, peak magnetic field 2.2 T) using two Magstim 200 magnetic stimulators (Magstim, UK) linked with a Bistim module (Magstim, UK). The stimulators were triggered through the pCLAMP software and Digidata 1320A Data Acquisition board. The optimal site for stimulation of the APB muscle was first established. The coil was placed on the scalp at the estimated position of the left motor cortex (5 cm lateral to and 2 cm anterior to the vertex). The coil was moved until the position that produced the largest MEP response at supra-threshold intensity was located. This position was marked on the scalp and used throughout the experiment. The coils were held tangential to the scalp with the handle pointing back and away from the midline at 45°. The induced current flow was posterior to anterior in the cortex perpendicular to the central sulcus. RTMS was applied with a Magstim Super Rapid stimulator (Magstim, UK) and a 70 mm figure-of-8 coil. The coil orientation was identical to that used for single and paired pulse stimulation. As the RMT produced with the Magstim Super Rapid and Magstim 200 vary, the RMT was remeasured with the Magstim Super Rapid prior to the rTMS and the stimulus intensity used for the rTMS was based on this measurement.

2.5. Measurement procedure

The following measures of cortical excitability were assessed before and after rTMS in the same order. For all subjects, testing of the measures of corticospinal excitability was completed within 15 min of the completion of the rTMS train.

2.6. Resting and active motor thresholds

The RMT was the minimum stimulator intensity that evoked a peak-to-peak MEP of >50 μV in at least 5 out of 10 consecutive trials. AMT was measured during a sustained low intensity contraction (5–10% of maximum). The AMT was the lowest intensity producing at least one MEP of 100 μV in 5 trials.

2.7. MEP size and CSP

MEP size was measured at rest and during a sustained tonic contraction. For measurement at rest, 10 sweeps of data were collected during stimulation at 125% of the RMT. Both MEP size and the CSP duration were measured during sustained contraction of 5% of maximum. Ten sweeps were recorded with stimulation at 125% AMT. The peak-to-peak MEP amplitude and the CSP duration were measured on individual sweeps off-line and the results were averaged. CSP duration was calculated from the time of stimulation to the return of spontaneous EMG activity. An investigator blinded to the status (85 or 115% rTMS stimulation intensity) of the subjects made all off-line measurements. If the RMT or AMT changed after rTMS, the stimulation intensity used for measuring the resting MEP size or CSP after rTMS was adjusted so that it remained 125% of the threshold intensity. As the MEP size is dependent on the level of stimulation intensity above motor threshold (Devanne et al., 1997), an increase in RMT alone could result in a decrease in MEP size. Adjustment of the stimulus intensity used for the measurement of MEP size controls for this potential confound.

2.8. Cortical inhibition and facilitation

The procedure for measuring CI and CF followed that described in the literature (Kujirai et al., 1993; Ziemann et al., 1996a,b). All measurements were conducted at rest with continuous EMG monitoring. Occasional sweeps with EMG activity (less than 2% of the total) were discarded. The initial or conditioning stimulus was set at 5% below the AMT. The second stimulus (test stimulus) was adjusted to produce

Table 1
Mean values (±SD) for each dependent measure before and after stimulation for the supra- and sub-threshold groups[a]

	Supra-threshold rTMS			Sub-threshold rTMS		
	Before stimulation	After stimulation	*P*	Before stimulation	After stimulation	*P*
RMT (%)	41.9 ± 5.5	43.9 ± 5.3	0.015	40.9 ± 6.3	42.9 ± 6.1	0.005
AMT (%)	33.0 ± 4.9	32.4 ± 5.2		32.5 ± 5.5	32.0 ± 6.1	
MEP size (rest) (μV)	989.8 ± 393.3	1220.5 ± 961.4		1238.0 ± 864.5	976.7 ± 707.6	
MEP size (active) (μV)	1408.4 ± 681.5	905.8 ± 568.9	0.008	1477.4 ± 543.5	1261.3 ± 605.5	0.30
SP duration (ms)	130.6 ± 28.2	126.3 ± 41.8		130.3 ± 31.9	127.2 ± 34.2	
Cortical inhibition (%)	55.8 ± 29.4	47.1 ± 27.5		57.6 ± 25.5	49.6 ± 29.7	
Cortical facilitation (%)	148.0 ± 79.5	180.6 ± 77.0		164.9 ± 73.1	208.7 ± 170.3	

[a] Post hoc *t* test results are presented for the variables in which a significant effect of stimulation was seen with ANOVA models.

MEPs of 0.5–1.0 mV. Ten trials were recorded for each of 3 conditions in a pseudo-random order; a control single test stimulus and 2 and 15 ms interstimulus intervals (ISIs). Stimuli were delivered 5 s apart. For each sweep, the peak-to-peak MEP size was measured and the average MEP size was calculated for each ISI and the control condition. CI and CF were then expressed as percentages of the mean control condition.

2.9. Statistical analysis

Several repeated measures two-way analysis of variance

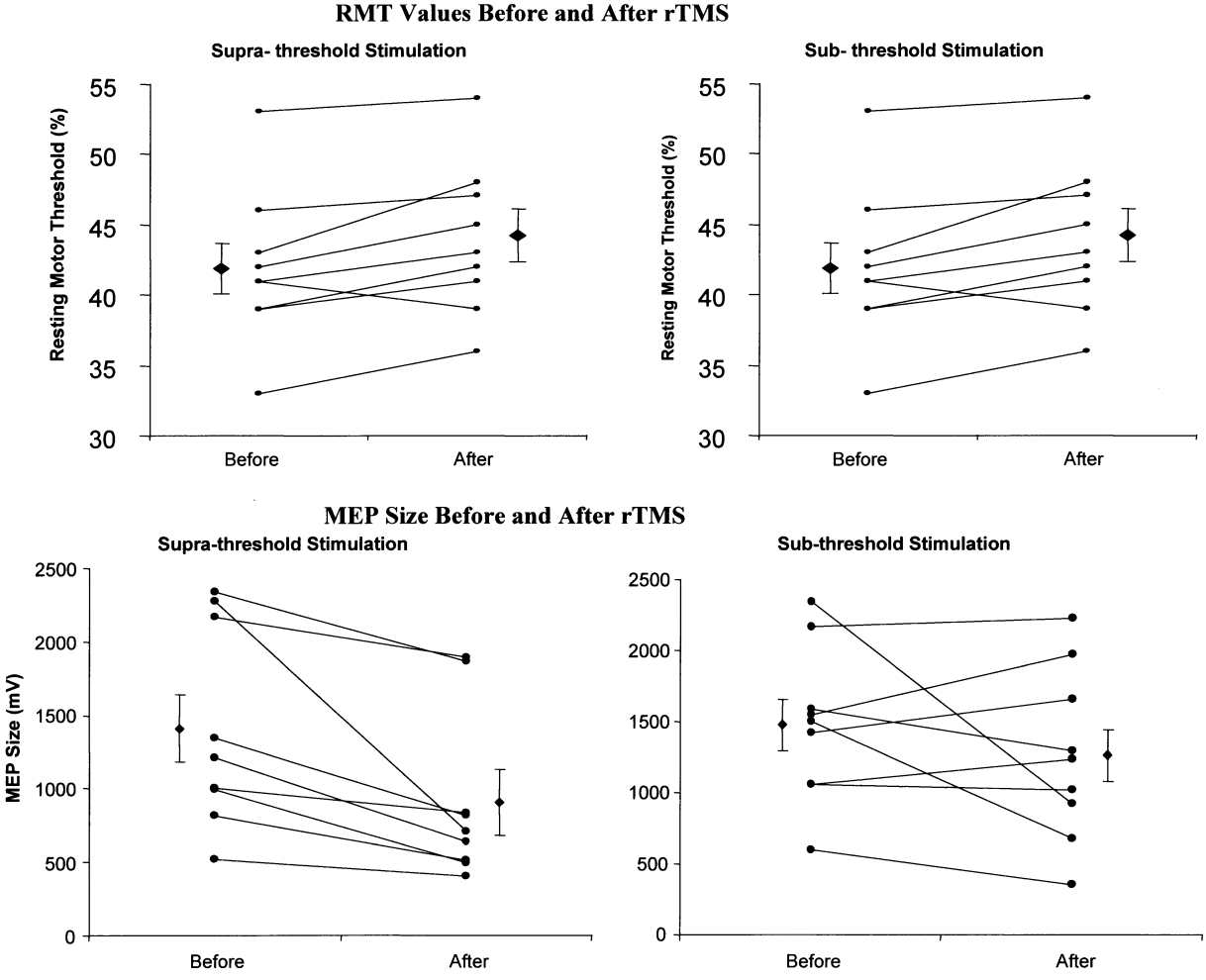

Fig. 1. Measures of RMT and MEP size (tonic contraction) before and after sub- and supra-threshold rTMS. Results before and after sub- and supra-threshold stimulation are presented for each subject with the means (±SEM) displayed on the sides of each figure. There was a significant increase in RMT at both intensity levels but the reduction in MEP size (tonic) was only significant with supra-threshold stimulation.

(ANOVA) models were used. In each model, one dependent variable (i.e. RMT, AMT, MEP size (resting and active), CSP duration and CI and CF measured with ppTMS) and sub- or supra-threshold stimulation were entered as the within-subjects repeated measures. Where significant effects were seen for a dependent variable, paired t tests were used to measure differences in the dependent variables before and after rTMS with the sub- and supra-threshold groups studied separately. All of the dependent variables were normally distributed except for the results for CF which were corrected with a log transformation prior to analysis. Significance was set at $P < 0.05$. Statistical analysis was conducted in SPSS 10.0 (SPSS for Windows 10.0, Chicago: SPSS; 2000) and Stata (Stata for Windows, Texas: Stata Corp; 2001).

3. Results

Mean values for each dependent variable before and after rTMS are presented in Table 1. Significant effects of stimulation in the ANOVA models were seen with an increase in RMT size ($F = 25.0$, d.f. $= 1$, $P < 0.01$) and a decrease in MEP (active) size ($F = 8.47$, d.f. $= 1$, $P < 0.05$). There was no stimulation versus intensity (sub and supra) interaction for either model (RMT: $F = 0.0$, d.f. $= 1$, $P > 0.05$, MEP active: $F = 1.35$, d.f. $= 1$, $P > 0.05$). There was no effect of stimulation on the magnitude of the AMT, MEP size (rest), CSP duration, CI and CF. Results for the paired t tests for the sub- and supra-threshold stimulation groups for RMT and MEP (active) are presented in Table 1 and the change in absolute values for each measure is presented in Fig. 1. In both groups, rTMS significantly increased RMT. A decrease in MEP size (active) was seen in the supra-threshold group but not in the sub-threshold group.

4. Discussion

This study confirms that 1 Hz rTMS applied to the motor cortex alters cortical excitability as indicated by a change in the RMT and the size of the MEPs produced during tonic contraction. The changes in RMT are seen with both sub- and supra-threshold stimulation intensities although the decrease in MEP size occurred only with supra-threshold rTMS. Moreover, low frequency rTMS at either sub- and supra-threshold stimulation intensities had no effect on the CSP, CI and CF as measured with ppTMS.

Several studies have explored the motor effects of trains of 1 Hz stimulation with effects on cortical excitability seen with both sub- and supra-threshold intensities of stimulation. For example, Maeda et al. (2000) reported reduced MEP size with stimulation for 26.7 min at 90% of RMT and several studies have reported changes either in MEP size or RMT with supra-threshold stimulation (Chen et al., 1997; Muellbacher et al., 2000). However, previous studies did not systematically compare the response to rTMS at various stimulation intensities in the same individuals. It is of considerable interest that whilst the effect on RMT was the same at the two stimulation intensities, there was a differential response in MEP size (active). This is not necessarily surprising as there is evidence that RMT and MEP responsiveness reflect differing aspects of cortical excitability (Hallett et al., 1999) and a differential effect of 1 Hz rTMS was seen over time in the study of Muellbacher et al. (2000). In this study, supra-threshold rTMS at 1 Hz produced changes in both RMT and MEP size with the change in MEP size but not RMT persisting for 30 min post stimulation.

The most significant difference seen between the results of our study and previous reports has been our failure to detect a reduction in resting MEP size which has been reported by several groups (Chen et al., 1997; Maeda et al., 2000; Muellbacher et al., 2000; Touge et al., 2001). There are several potential reasons why we did not detect this difference. Firstly, we only evoked 10 MEPs in the measurement of this variable. It is well established that there is considerable variability in MEP amplitudes, especially at lower intensity levels (van der Kamp et al., 1996). More sensitive means of assessing MEP size, such as measuring 'response' or 'recruitment' curves (Wassermann et al., 1998) would be more appropriate to the assessment of subtle changes in MEP size. It is also possible that an effect was obscured by differences between subjects in the time course of the effects. All of the post-TMS measures were obtained in the same order in all subjects and took a similar duration of time. However, we have not assessed the time course of the presence of motor cortical activity changes and subtle differences in the time course of effects or the timing of measurements could also affect our ability to distinguish relevant changes.

A third possibility relates to our adjustment of the post-rTMS stimulation intensity when applied in measurement of the MEP size. In most studies in which MEP size has been measured before and after rTMS, the stimulation intensity in the post-rTMS testing has not been adjusted to take into account alterations in motor threshold that have occurred with rTMS (for example Tergau et al., 1997). This experimental methodology allows the assessment of 'absolute' MEP size. However, as the MEP size is dependent on the level of stimulation intensity above motor threshold (Devanne et al., 1997), it is possible that decreases found in MEP size may simply reflect an increase in the RMT. Therefore, we chose to assess the MEP 'relative' to RMT and adjusted the stimulation intensity to do this. Whether changes in MEP size can still be seen with adjustment in stimulation intensity will require further assessment using sensitive techniques such as assessment of response curves pre- and post-rTMS.

The reduction in active MEP size following low frequency rTMS found in our study has not been previously reported. Romeo et al. (2000) found no change in MEP size during tonic activity with 1 Hz stimulation but only a total

of 80 stimuli were administered in 4 trains. Touge et al. (2001) used a much longer period of stimulation (1500 stimuli) and found no reduction in MEP size. The authors argued that the effect of voluntary contraction would overcome changes in the excitability of the corticospinal projection. However, the stimulus intensity used in this study was 95% of the RMT, consistent with the lack of an effect with sub-threshold stimulation in our study and it is possible that prolonged supra-threshold stimulation may be necessary to change MEP size during tonic activity. This raises the possibility that prolonged supra-threshold stimulation may produce effects in synaptic efficacy (the efficacy of the synaptic inputs onto corticospinal neurones) whereas the effects of sub-threshold stimulation may occur only through changes in neuronal excitability as was suggested in the study of Touge et al. (2001).

We found no effect of rTMS on the CSP, CI and CF measured with ppTMS. This is consistent with the study of Romeo et al. (2000), which found changes in CSP duration with stimulation only at frequencies greater than 1 Hz although the effect did not outlast the duration of the train. We have similar findings with a considerably longer duration of stimulation. Our data is also consistent with the lack of a significant effect of 1 Hz stimulation on CI in the study of Tergau et al. (1997). Several lines of evidence suggest that the CSP and CI are produced through activation of different cortical GABAergic circuits (Sanger et al., 2001). The lack of an effect on CSP occurred simultaneously with the changes recorded in MEP size (active) suggesting that the effects of 1 Hz stimulation on RMT and MEP size are not mediated through an up regulation of local inhibitory activity. The lack of an effect on CF also indicated that changes in excitability were not associated with altered excitatory input to the motor neurone. These findings are consistent with previous research that has suggested that changes in MEP size and RMT strength relate directly to changes in neuronal excitability (Touge et al., 2001).

The mechanism through which rTMS at 1 Hz results in reduced corticospinal excitability is unclear. Parallels have been drawn between this type of response and the induction of synaptic long-term depression (LTD). LTD can be produced in homosynaptic cellular preparations from tissue areas that include the cortex (Kirkwood et al., 1993; Hess and Donoghue, 1996) and TMS has been seen to produce LTD like changes in animal cortical tissue (Wang et al., 1999). It seems possible that the changes seen with rTMS in humans are produced through LTD but this has not been definitively established. Separate mechanisms appear to underlie the changes seen in MEP size and RMT, since RMT and MEP reflect different aspects of cortical excitability (Hallett et al., 1999), respond differentially to sub-threshold stimulation and differing stimulation train duration (Muellbacher et al., 2000). The mechanisms involved remain unclear although it is suggested that MEP size reflects global corticospinal pathway excitability and RMT is determined in part by membrane related aspects of cellular excitability (Ziemann et al., 1996a,b; Devanne et al., 1997). The intensity dependent nature of the effect of rTMS on MEP size also suggests another possibility: that changes in MEP size, but not RMT, may result from the stimulation of the pre-motor cortex rather than the primary motor cortex itself. Pre-motor cortex stimulation at very low intensities reduces MEP size, although the same intensity was insufficient to produce changes when applied directly to the motor cortex (Gerschlager et al., 2001). Supra-threshold primary motor cortical stimulation may produce an effect on MEP size via the 'overflow' of stimulation to the pre-motor area, rather than any effect on the primary motor cortex. Since changes in RMT size were observed with sub-threshold stimulation, these are more likely to be related to primary motor cortex stimulation, as the degree of 'overflow' to adjacent regions would seem likely to vary with intensity.

Both sub- and supra-threshold rTMS applied at 1 Hz produced changes in corticospinal excitability but only supra-threshold stimulation produced reliable changes in the size of MEPs. 1 Hz stimulation had no persisting effect on the CSP, CI and CF. The relevance of these findings to the application of rTMS in therapeutic settings is unclear. Although the findings suggest that more robust clinical affects may be seen with supra-threshold stimulation levels, as we have little knowledge of the relationship between these types of changes and therapeutic response rates, it may be premature to abandon the investigation of stimulation regimes that use sub-threshold intensities.

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