

The Role of Interneuron Networks in Driving Human Motor Cortical Plasticity

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The after-effects of repetitive transcranial magnetic stimulation (rTMS) are highly variable between individuals. Because different populations of cortical neurons are stimulated more easily or are more excitable in different people at different times, the variability may not be due to differences between individuals in the plasticity of cortical synapses, but may instead be due to individual differences in the recruitment of cortical neurons. In this study, we examined the effects of rTMS in 56 healthy volunteers. The responses to excitatory and inhibitory theta burst stimulation (TBS) protocols were highly variable between individuals. Surprisingly, the TBS effect was highly correlated with the latency of motor-evoked potentials (MEPs) evoked by TMS pulses that induced an anterior–posterior (AP) directed current across the central sulcus. Finally, we devised a new plasticity protocol using closely timed pairs of oppositely directed TMS current pulses across the central sulcus. Again, the after-effects were related to the latency of MEPs evoked by AP current. Our results are consistent with the idea that variation in response to rTMS plasticity probing protocols is strongly influenced by which interneuron networks are recruited by the TMS pulse.

Keywords: LTD, LTP, Motor cortex, Transcranial magnetic stimulation

Introduction

Transcranial magnetic stimulation (TMS) is a non-invasive technique to stimulate the human brain. Initial studies used single-pulse stimulation, but more recently there has been much interest in repetitive TMS (rTMS). This is because rTMS appears to have after-effects on the excitability of the stimulated area that outlast the period of stimulation by minutes or even hours. Evidence suggests that at least some of these effects depend on activity in N-Methyl-D-Aspartate (NMDA) receptors and hence it is thought that they represent an analog of early stages of synaptic plasticity in the human brain (Ziemann et al. 2008).

A major issue with rTMS is that the responses are highly variable both within and between individuals. A number of factors contribute to this variability, including age, gender, time of the day, regular activity, attention, previous history of plasticity, neuromodulators, and genetics (see review by Ridding and Ziemann 2010). However, a major question underlies the interpretation of all these observations: is variability the result of differences in intrinsic mechanisms of synaptic plasticity (Ridding and Ziemann 2010) or is it due to the fact that different populations of cortical neurons are stimulated more easily or are more excitable in different people at different times (Day et al. 1989; Rothwell 1997)? In other words, is variability telling us something about

differences in synaptic plasticity or is it telling us about how easy it is to stimulate the axons of certain types of neurons?

To try to gain some insight into this, we examined two commonly used forms of rTMS, continuous and intermittent theta burst stimulation (cTBS and iTBS). cTBS tends to depress excitability of the primary motor cortex for 30 min whereas iTBS has the opposite effect (Huang et al. 2005). More importantly for the purpose of the current argument, it is known from spinal epidural recordings of the descending volleys evoked by single-pulse TMS before and after TBS that cTBS depresses early indirect waves (I-wave) which are thought to originate from a monosynaptic excitatory connection to pyramidal cells (Di Lazzaro et al. 2005). Furthermore, iTBS enhances late I-waves which are generated by more complex oligosynaptic circuits (Di Lazzaro et al. 2008). Our hypothesis was that some of the inter-subject variability in response to each protocol is due to differences in the population of neurones activated by each TMS pulse. We predicted that people in whom stimulation readily recruited early I-waves might have a good response to cTBS whereas those in whom late I-waves were recruited would have better responses to iTBS.

TBS is usually applied using a biphasic stimulus pulse. It preferentially activates the brain during the second depolarizing phase of the current in the brain, that is, with an anterior–posterior (AP) current (Maccabee et al. 1998; Di Lazzaro, Oliviero, Mazzone, et al. 2001). We therefore attempted to quantify in each individual the level of early/late I-wave recruitment by measuring onset latencies of motor-evoked potentials (MEPs) evoked in precontracted muscle at near-threshold intensities with a monophasic AP current (Day et al. 1989; Werhahn et al. 1994; Sakai et al. 1997; Di Lazzaro et al. 1998; Di Lazzaro, Oliviero, Saturno, et al. 2001). These were then compared with MEPs evoked by direct wave (D-wave) activation using latero-medial (LM) current stimulation in the same individuals. We reasoned that individuals in whom onset latencies were close to D-wave latency would readily recruit early I-waves whereas those with later onsets would tend to recruit later I-waves (Day et al. 1989; Rothwell 1997).

Subjects and Methods

We recruited 56 healthy participants (24 women; 18–52 years old; mean \pm SD, 30.3 \pm 7.4) who had no previous history of neurological, psychiatric, or other medical problems, and had no medical history that precluded them from TMS (Rossi et al. 2009). All participants gave written informed consent in accordance with the Declaration of Helsinki. The study was

approved by the Ethics Committee of the University College London.

Recordings

Participants were seated on a comfortable chair. The electromyogram (EMG) activity was recorded from the right first dorsal interosseous (FDI) muscle via Ag/AgCl cup electrodes in a belly-tendon montage. The raw signal was amplified and filtered with a bandpass filter of 20 Hz to 3 kHz (Digitimer, Welwyn Garden City, United Kingdom). Signals were digitized at 5 kHz (CED Power1401; Cambridge Electronic Design, Cambridge, United Kingdom) and stored on a computer for off-line analysis (Signal version 4.08, Cambridge Electronic Design, Cambridge, United Kingdom).

Transcranial Magnet Stimulation

TMS was performed using Magstim 200² stimulator (The Magstim Co. Ltd) connected to a figure-of-eight coil with an internal wing diameter of 7 cm. Previous studies have clearly shown that different descending volleys are elicited by single-pulse TMS depending on the current flow across hand area of the motor cortex. For example, posterior-anterior directed current (PA current) preferentially elicits early I-waves, whereas AP current recruits late I-waves and LM current at high stimulus intensity evokes direct-waves (D-wave; Day et al. 1989; Werhahn et al. 1994; Sakai et al. 1997; Di Lazzaro et al. 1998; Di Lazzaro, Oliviero, Saturno, et al. 2001). The following 3 different coil directions were used in the present study (Fig. 1). (1) PA-directed currents were produced by the figure-of-eight coil held posterolaterally at an angle of about 45° to the midline; (2) AP-directed currents were elicited by placing the coil 180° to the PA currents; and (3) the coil was placed with the handle pointing leftwards for LM-directed currents (90° from midsagittal line). By systematically moving the coil with PA currents at 0.5 cm intervals in the anterior-posterior and medio-lateral direction, the hotspot was identified as the position where the maximum and stable MEP response was achieved. This position was marked with a pen on the scalp for repositioning the coil. Another coil was connected to Magstim Super Rapid (The Magstim Co. Ltd) to

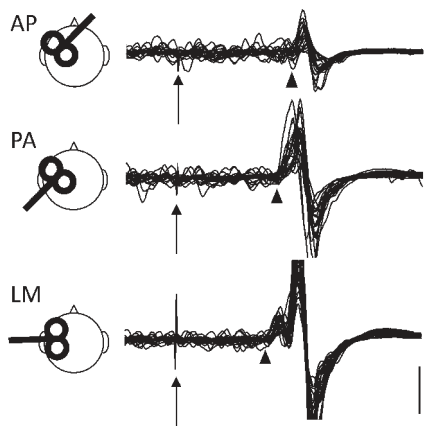


Figure 1. A schematic representation of the coil orientations and typical example of MEPs during contraction by each stimulus. Arrow indicates the timing of TMS and arrow head indicates the onset of MEPs. The PA-LM latency difference was 1.6 ms in this case, whereas AP-LM was 5.2 ms, compatible with known latency differences between D and I1-wave or I3-waves (Day et al. 1989). Calibration bars, 1 mV, 20 ms.

perform TBS. This coil was placed with usual coil orientation; that is, the handle pointing backwards at about 45° laterally. This orientation was to ensure that biphasic pulse elicited eddy currents in the brain PA-AP direction. The resting motor threshold (RMT) with PA currents (RMTpa) was defined as the minimum stimulation intensity over the motor hot-spot, required to evoke an MEP of no less than 50 μ V in 5 out of 10 trials (Rossi et al. 2009). The active motor threshold (AMT) was defined as the lowest intensity to evoke an MEP of 200 μ V in more than 5 of 10 consecutive trials while subjects maintained approximately 10% contraction of the target muscle. We measured AMT with PA, AP, and LM currents, defined as AMTpa, AMTAp, and AMTlm, respectively. In the experiment 1 (see below), AMT using the biphasic machine was also measured (AMTbi). All of these and following measurements (see below, experimental parameters) were performed at the hotspot determined by PA currents, since previous experiments have shown that the direction of the current does not significantly influence the position of the hotspot (Sakai et al. 1997; Arai et al. 2005).

Experimental Parameters

Onset Latency MEP Measurements During Muscle Contraction

We first measured onset latency of MEPs using PA, AP, and LM currents during mild contraction of the target muscle (~10% of the maximum voluntary contraction). Stimulus intensity was set at 110% AMTpa, 110% AMTAp, and 150% AMTlm (or 50% of maximum stimulator outputs (MSOs) in subjects whose 150% AMTlm did not reach 50% MSO). Relatively high stimulus intensities for LM currents were used in order to ensure that a D-wave was evoked. This was based on the previous study by Werhahn et al. (1994), which showed LM stimulation at intensities above 50% MSO induces MEPs with the same onset latency as those evoked by anodal electrical stimulation. Twenty responses for PA and AP currents and 10 for LM currents applied over the hotspot were obtained to confirm the reproducibility of the results. Every 10 measurements, the subjects were asked to relax their hand to avoid fatigue. These measurements were taken over 10–15 min. The shortest latency was measured from the superimposed waveforms by visual inspection (Rothwell et al. 1987; Day et al. 1989; Sakai et al. 1997; Di Lazzaro, Oliviero, Saturno, et al. 2001; Chen et al. 2008; Shirota et al. 2011). We also employed an automated method by which the onset was measured automatically to minimize observer bias using custom-made program. In each trial, the onset was defined as the time point where rectified EMG signals exceed an average plus two standard deviations of the pre-stimulus EMG level (–100–0 ms of TMS). For each subject and for coil orientation, the onset latencies were averaged and compared with those by manual measurements to test consistency between 2 methods (see below).

Previous studies have also found the onset latency of MEPs by PA currents to be 1–2 ms later than those by LM currents and AP currents 4–5.5 ms later (Day et al. 1989). This suggests that PA currents preferentially activate early I-waves, and AP currents mainly activate later (i.e. I3) waves (Day et al. 1989; Rothwell 1997; Sakai et al. 1997). Therefore in this study, we decided to use latency difference between LM and AP evoked MEP onsets as a measure of efficiency of late I-wave

Table 1Physiological measures (mean \pm SEM, range)

	cTBS	iTBS
RMTpa (%)	44.3 \pm 1.2 (26–63)	44.5 \pm 1.3 (27–63)
AMTpa (%)	33.4 \pm 0.9 (19–48)	33.4 \pm 0.9 (18–47)
AMTpa (%)	45.5 \pm 1.5 (25–75)	45.5 \pm 1.5 (21–70)
AMTlm (%)	38.4 \pm 0.9 (22–50)	38.5 \pm 0.9 (25–51)
AMTbi (%)	46.8 \pm 1.2 (28–62)	46.9 \pm 1.1 (29–63)
SI _{1mV} (%)	56.0 \pm 2.0 (36–90)	55.4 \pm 1.9 (32–90)
Baseline MEP sizes (mV)	1.02 \pm 0.03 (0.44–1.60)	1.12 \pm 0.04 (0.56–2.27)

Note: cTBS, continuous theta burst stimulation; iTBS, intermittent TBS; RMT, resting motor threshold; AMT, active motor threshold; pa, posterior-to-anterior; ap, anterior-to-posterior; lm, lateral-to-medial; bi, biphasic; SI_{1mV} stimulus intensity to elicit MEP sizes about 1 mV.

recruitment; the longer the latency difference, the more efficient is later I-wave recruitment. Although we investigated the correlation between TBS responses and threshold in this study (see Results), we acknowledge that threshold itself may be confounded by the fact that neurons stimulated by each coil orientation are likely to be different between subjects.

Motor Cortical Excitability

In each subject at baseline, 30 MEPs were recorded with the stimulus intensity set to evoke a stable MEP (of about 1 mV) (SI_{1mV}) every 4.5–5.5 s with the subject at rest (see Table 1 for mean baseline MEP amplitudes for each experiment). SI_{1mV} was kept constant throughout the experiment.

Interventions

Theta Burst Stimulation

TBS was applied to the hot-spot of FDI. Each burst consisted of three stimuli (80% AMTbi) given at 50 Hz. Continuous TBS (cTBS) was delivered as a sequence of 200 bursts given at a rate of 5 Hz (total duration of 40 s); intermittent TBS (iTBS) involved giving a 2 s train repeated every 10 s for 20 repetitions (190 s) (600 stimuli) (Huang et al. 2005).

Intracortical PAS

We developed a new method to examine the interaction of AP and PA currents in producing associative plasticity within the motor cortex: intracortical paired associative stimulation (icPAS). This involved placing 2 oppositely oriented figure-of-eight coils on top of each other (the coil for AP current was the bottom coil) (Fig. 6A). The AMT for PA current was re-measured (AMTpa+) prior to being used for intervention. This was because we noted AMTpa+ was significantly higher than AMTpa (AMTpa, 31.2 \pm 1.2%; AMTpa+, 65.4 \pm 2.4%; $t = -23.765$, $P < 10^{-15}$). The stimulus intensity was set at 100% AMTpa+ for PA currents and at 100% AMTpa for AP currents. A stimulus intensity of 100% AMT was chosen in order to activate relatively homogenous neurons with each pulse. PA current was preceded or followed by AP current (e.g. icPAS+5, AP-before-PA-after pairing at 5 ms; icPAS-5, PA-before-AP-after pairing at 5 ms). The intervals between the pulses were 5 ms; 60 pairs in total were applied at 0.2 Hz. In the icPAS protocol, it is important to note that even though the coils overlap, stimulation via one coil will not induce any current in the other coil since there is no current loop to flow around when the stimulator is “off” (i.e. the thyristor switch means that there is basically an open circuit). A similar approach has been employed successfully in a previous study (Ziemann et al. 1996).

Study Design

Experiment 1

Fifty two subjects participated in this crossover study, which consisted of 2 randomized ordered sessions, separated by at least 3 days (cTBS and iTBS). First, as described above, RMT, AMTs, and MEPs during contraction were measured. Because prior contraction of a target muscle influences the after-effects of TBS (Gentner et al. 2008; Iezzi et al. 2008) and because there is no effect of the prior contraction if the interval between TBS and prior contraction is longer than 10 min (Hsu et al. 2011), we waited for 15 min after measurements of AMTs and MEPs during contraction. Thereafter, SI_{1mV} was established and baseline motor cortical excitability measurements (30 MEPs with PA currents) were started. After the intervention, 30 MEPs were recorded every 5 min for 30 min (7 time points, T0, T5, ..., T30). Subjects were instructed to keep the FDI completely relaxed during and following the application of TBS, as activation during or following TBS has previously been shown to alter the after-effects (Huang et al. 2008). EMG signals were monitored throughout the experiments.

Experiment 2

We used the icPAS+5/–5 in 16 subjects of whom 4 had not participated in experiment 1 in a crossover study, separated by at least 3 days (icPAS+5 and icPAS–5). The order of physiological measures was same as those described in experiment 1: after the measurements of MTs and MEPs during contraction, SI_{1mV} was first established, followed by baseline measurements of motor cortical excitability (30 MEPs with PA currents). After the intervention, 30 MEPs were recorded every 5 min for 30 min. Of 16 subjects, 9 of them fulfilled the inclusion criteria, which specified that PA–LM latency difference must be 1–2 ms, and that AP latency must be at least 1 ms longer than PA latency. This inclusion criteria was established because we wished to avoid slight differences in stimulated neurons by PA currents (e.g. D-wave and I1-wave) and to ensure that neurons stimulated by AP currents were different from those by PA currents (e.g. I2 and I3-waves). The PA–LM latency difference was 1–2 ms (see Results), consistent with I1-wave recruitment. We reasoned that the outcome should depend on the difference in the character of neurons stimulated by AP currents. The AP–PA latency difference was chosen to correlate with icPAS effects because there was some variability in the PA–LM latency difference. Therefore we “normalized” the onset latency of AP currents to that of PA currents. In 5 subjects, we measured RMTpa and F-wave amplitudes (20 supra-maximal stimulation at wrist) before and after icPAS interventions. We also performed control experiments in 9 subjects using pairings of 2 stimuli with the same direction in order to show the importance of changing the orientation of 2 stimuli in the icPAS protocol. In brief, 2 stimuli at 5 ms intervals were applied at 0.2 Hz for 60 pairs (PA or AP pairing) (Fig. 6A). Stimulus intensity was 100% AMT for each orientation. The after-effects were measured as in icPAS intervention.

Data Analysis

Normality of all of the measurements values was tested with Kolmogorov–Smirnov tests. Log-transformed data were used whenever necessary. Levene’s test was used to test for

equality of variances between latency differences. Intraclass correlation coefficient (ICC) was calculated to test consistency of latency values between 2 sessions. ICC was also calculated between manual and automated methods of estimating onset latencies. One-way analysis of variance (ANOVA) was employed with factors "TIME" (baseline, T0–T30) using absolute MEP values in each experimental session. For each subject and time point, the single-trial peak-to-peak MEP amplitudes were averaged and normalized to the MEP amplitude measured at baseline. Two-way repeated measures ANOVA was conducted with within-subject-factors "TBS" (cTBS and iTBS) ("icPAS" (icPAS+5 and icPAS-5) or "pairing" (PA and AP pairing) for experiment 2) and "TIME" (T0–T30) using normalized values. The Greenhouse–Geisser correction was used if necessary to correct for non-sphericity. TBS, icPAS, or pairing effects were also assessed by the grand average of normalized MEP amplitudes measured at time points T0–T30 to evaluate the correlation between all of the measurements values and cTBS or iTBS effects. A regression analysis was computed with the TBS, icPAS, or pairing effects as the dependent and the neurophysiological measures as the independent variable. Regression analysis was also performed to investigate relationship between cTBS and iTBS. Pearson's correlation coefficient was calculated. In experiment 1, responder and non-responder was defined operationally according to the grand average of TBS responses below and above 1 for cTBS and vice versa for iTBS. Because our primary interest was how the efficiency of late I-wave recruitments was involved in TBS responses, we arbitrarily set 4 ms of AP–LM latency difference as an index of I3-waves according to Day et al. (1989), and then performed chi-square tests to evaluate the percentages of short (<4) and long (≥ 4) latency difference groups between responder and non-responders. We also performed unpaired *t*-test (two-tailed) to evaluate the dependency of gender or time of the day (morning vs. afternoon) on TBS effects (morning session, started from 10:00 am; afternoon session, started from 13:00 pm or later; Sale et al. 2007). For these analyses, multiple comparisons with Bonferroni's corrections were applied; significant level was set at 0.00125 since we compared 20 factors with 2 conditions (Fig. 4C, Table 2). Otherwise, $P < 0.05$ was considered significant. Data were analyzed using software (SPSS ver. 19.0 for Windows; SPSS Inc.). All data are given as mean \pm standard error of the mean (SEM).

Results

Baseline physiological measures are shown in Table 1. Three subjects reported slight discomfort during cTBS due to spread of stimulus sensation to face, but it did not persist after completion of cTBS. All participants completed 2 sessions in each experiment.

Latency Difference among Different Coil Orientations

As explained in the Methods, D-wave latency was estimated from the onset of large MEPs evoked by LM stimulation. Our estimate of the I-wave circuits activated during TBS was obtained by measuring the onset latency of near-threshold MEPs evoked by AP stimulation relative to D-wave activation. Finally, we also measured onset latencies of near-threshold MEPs evoked by more standard posterior–anterior (PA) stimulation.

Table 2

Correlation between physiological measure and TBS responses

Factors	cTBS grand average		iTBS grand average	
	Correlation coefficient	<i>p</i>	Correlation coefficient	<i>P</i>
Age	-0.306	0.027	0.070	0.623
RMTpa	0.018	0.901	-0.078	0.583
AMTpa	-0.098	0.492	-0.057	0.690
AMTpa	0.064	0.653	-0.328	0.018
AMTlm	0.021	0.855	-0.050	0.740
AMTbi	-0.327	0.018	0.099	0.483
Thd LM and PA	0.125	0.376	-0.023	0.872
Thd AP and LM ^a	-0.085	0.547	0.321	0.020
Thd AP and PA	0.223	0.113	-0.459	<10⁻³
LM lat	-0.141	0.318	0.057	0.690
PA lat	-0.348	0.011	0.405	0.0028
AP lat	-0.686	<10⁻⁷	0.672	<10⁻⁷
PA–LM LD ^a	-0.238	0.089	0.333	0.016
AP–LM LD	-0.712	<10⁻⁸	0.727	<10⁻⁸
SI _{mV} ^a	0.098	0.487	-0.047	0.740
Baseline MEP size	-0.152	0.282	-0.258	0.064

Note: LD, latency difference; Thd X and Y, threshold difference between two directions (X and Y), defined as following equation, $(AMT_x - AMT_y) / AMT_x \times 100$. Otherwise, abbreviations are the same as in Table 1. Boldface highlights significant after Bonferroni's multiple corrections.

^aThe measures with non-normal distribution (Kolmogorov–Smirnov test).

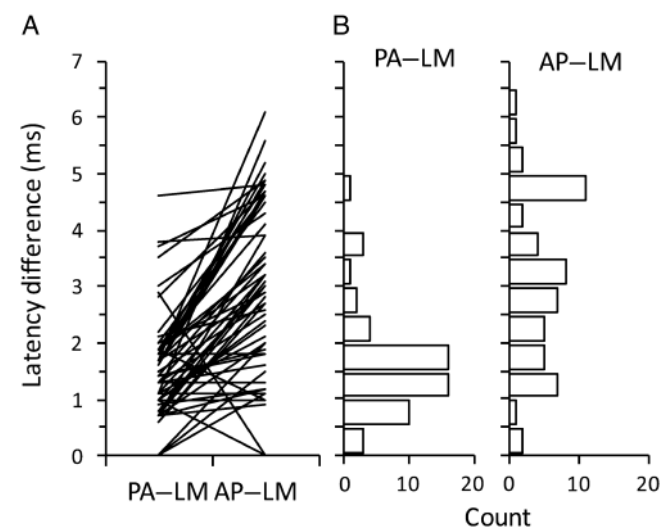


Figure 2. (A) PA–LM and AP–LM latency differences from 56 subjects. (B) The number of subjects at each latency difference value (0.5 ms step).

Figure 1 shows typical examples from one individual of MEPs evoked during contraction with different coil orientations. The PA–LM latency difference was 1.6 ms in this case, whereas AP–LM was 5.2 ms. These results are compatible with known latency differences between D and I1-wave or I3-waves, respectively (Day et al. 1989). The group results from 56 subjects in the first session (e.g. 52 from experiment 1 and another 4 from experiment 2) are plotted in Figure 2A. The PA–LM latency difference was relatively consistent, while AP–LM was not. Figure 2B replots the same data as a frequency distribution in which latency differences are expressed in 0.5 ms time bins. There was a much broader distribution of AP–LM latencies compared with PA–LM latencies (Leven's tests, $F = 16.786$, $P < 0.0001$). Since we measured these values twice in both experiments, ICC was calculated to test consistency between latency values measured in the same individual on different days (Fig. 3). ICCs were 0.874 (LM

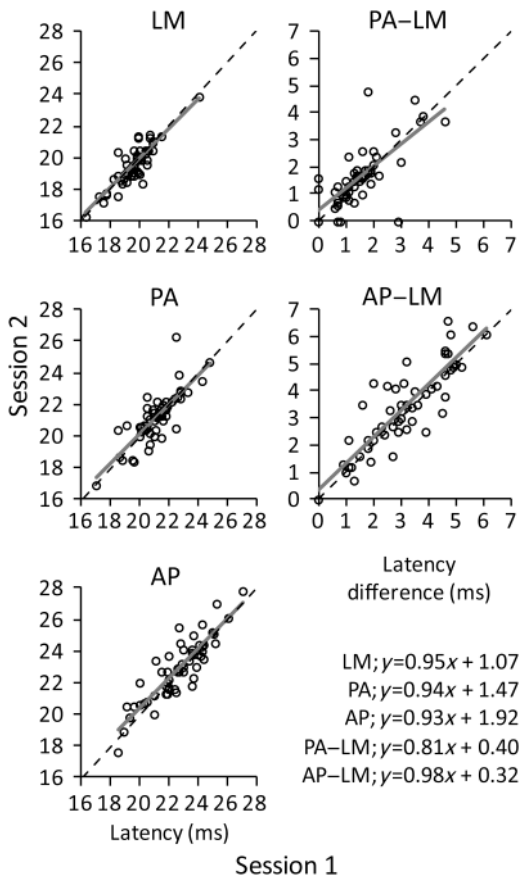


Figure 3. Test–retest reliability of each value. Equations by linear regression analysis were presented (gray line). X-axis, value in session 1; Y-axis, value in session 2. The dotted line, $y = x$.

latency, $P < 0.001$), 0.810 (PA latency, $P < 0.001$), 0.885 (AP latency, $P < 0.001$), 0.712 (PA–LM latency difference, $P < 0.001$), and 0.879 (AP–LM latency difference, $P < 0.001$). This confirms that the spread of data is due almost entirely to inter-individual differences in response to TMS. As a check on these manually measured onset latencies, we also employed an automated method (see Methods) in which the onset was measured as the time point where the rectified EMG signals exceeded an average plus 2 standard deviations of the pre-stimulus EMG level. The ICCs between the manual and automated methods were 0.879 (LM latency, $P < 0.001$), 0.886 (PA latency, $P < 0.001$), 0.888 (AP latency, $P < 0.001$), 0.868 (PA–LM latency difference, $P < 0.001$), and 0.912 (AP–LM latency difference, $P < 0.001$), indicating good consistency between them.

Finally, it could also be argued that the range of AP–LM latency values is related to differences between individuals in the size of MEP evoked by AP currents. However, there was no significant correlation between the AP–LM latency difference and MEP amplitude evoked by AP current ($r = -0.155$, $P = 0.174$).

Theta Burst Stimulation

Figure 4 shows time course of cTBS (Fig. 4A) and iTBS (Fig. 4B) in all 52 subjects. The cTBS and iTBS after-effects were highly variable. Indeed in this set of participants there was no overall effect of either form of stimulation: 1-way

ANOVA on cTBS and iTBS data separately revealed no significant main effect of TIME (cTBS, $F_{7,357} = 0.412$, $P = 0.895$; iTBS, $F_{7,357} = 0.329$, $P = 0.941$) and 2-way repeated measures ANOVA on combined cTBS/iTBS data showed no significant main effects of TBS ($F_{1,51} = 0.006$, $P = 0.939$), TIME ($F_{4,694,239.4} = 0.216$, $P = 0.949$), and TIME \times TBS interaction ($F_{4,827, 246.2} = 0.485$, $P = 0.781$). The same was true if the analysis was confined to the timing when maximum effects were reported previously (Huang et al. 2005). There was no mean effect of TBS after 5–10 min (cTBS, $t = -0.489$, $P = 0.627$; iTBS, $t = -1.094$, $P = 0.279$) and after 10–15 min (cTBS, $t = -1.193$, $P = 0.238$; iTBS, $t = -0.582$, $P = 0.563$).

We tested whether the TBS effects in each individual were correlated with any of the baseline physiological measures that we had collected (Table 2, Fig. 4C). Manhattan plots showed that the clearest correlations were with the AP latency and AP–LM latency difference for both iTBS and cTBS. There were borderline significant negative correlations of the response to iTBS with the threshold difference between PA and AP (Thd AP and PA). This seems to be due to a significant correlation between Thd AP and PA and AP–LM latency difference ($r = -0.516$, $P < 10^{-5}$). Finally, there were no significant correlations (after Bonferroni’s correction, significant level, $P = 0.00125$) with age (cTBS, $P = 0.027$; iTBS, $P = 0.623$), gender (cTBS, $t = -0.282$, $P = 0.779$; iTBS, $t = 0.068$, $P = 0.946$) or time of the day (cTBS, morning session, $n = 11$, $t = -0.552$, $P = 0.591$; iTBS, morning session, $n = 10$, $t = 2.729$, $P = 0.020$).

Figure 4D–F plots the relationships between effects of TBS and the AP–LM latency difference. In Figure 4D, TBS effects have been measured at a single time point of 5–10 min. The correlations here between MEP sizes and AP–LM latency difference (cTBS, T5, $r = -0.563$, $P < 10^{-4}$; iTBS, T5, $r = 0.542$, $P < 10^{-4}$) were slightly smaller than seen in the mean data from all time points which are plotted in Figure 4E and F. The latter show a clear negative correlation between the response to cTBS and AP–LM latency difference ($r = -0.712$, $P < 10^{-8}$; Fig. 4E, left) and a clear positive correlation for effects after iTBS ($r = 0.727$, $P < 10^{-8}$; Fig. 4F, left). The histograms to the right of these graphs plot the data in a different way. They show that participants who had the expected inhibition after cTBS (grand average effect < 1 ; Fig. 4E, right) or who had facilitation after iTBS (effect > 1 ; Fig. 4F, right) had a significantly greater AP–LM latency difference than non-responders (cTBS, $t = 5.083$, $P < 10^{-5}$; iTBS, $t = -5.091$, $P < 10^{-5}$). Conversely, people who had the opposite response to “expected” (i.e. facilitation to cTBS and suppression to iTBS) had a short AP–LM latency difference.

The proportions of participants who responded to TBS in the “expected” and “unexpected” (non-responders) ways are shown in Figure 5A and B. Subjects who had a AP–LM latency difference ≥ 4 ms were more likely to respond as expected to both forms of stimulation (cTBS, $\chi^2 = 14.359$, $df = 1$, $P = 0.00015$; iTBS, $\chi^2 = 18.876$, $df = 1$, $p = 0.000014$). Figure 5C shows that there was a significant negative correlation between the response to cTBS and iTBS ($r = -0.467$, $p = 0.00048$). Dividing individuals into groups with expected and unexpected responses (Fig. 5D) we found that expected responses (cTBS inhibitory and iTBS facilitatory) occurred in 13 subjects (25%), whereas 16 subjects (31%) showed completely opposite behavior. In addition, 9 subjects (17%) and 14 subjects (27%) showed either inhibitory or facilitatory effects irrespective of TBS protocol.

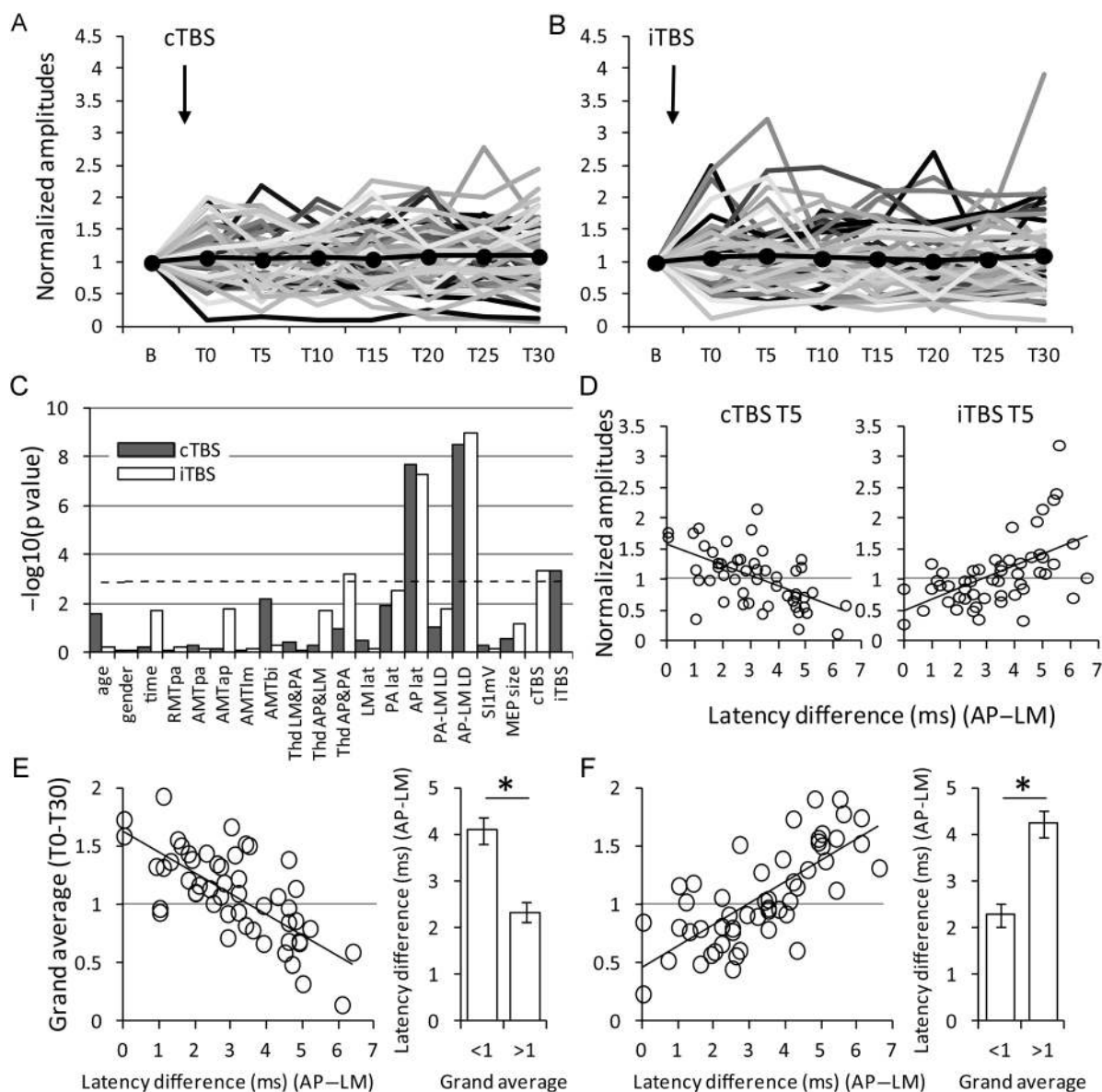


Figure 4. TBS effects and its relationship to latency difference. (A) and (B) Time course of cTBS and iTBS. X-axis shows time points and Y-axis shows normalized amplitude of MEPs to baseline (B). Thick black line and dot indicate mean. (C) Manhattan plot ($-\log_{10}(P\text{-value})$) of measured variables. Dotted line indicates significant level after multiple comparisons with Bonferroni's corrections. Time indicates the factor time of the day. Otherwise, abbreviations are same as those in Tables 1 and 2. (D) Correlations between AP-LM latency difference and TBS effects at 5 min. A significant correlation was found at each time point. (E) and (F) Left, correlation between grand average of cTBS (E) or iTBS (F) effects (T0-T30) and AP-LM latency difference. Right, average AP-LM latency differences between responder and non-responder.

Intracortical PAS

The analysis above assumes that the AP-LM latency difference is a surrogate measure of the relative ease of recruiting early and late I-wave input to corticospinal neurons. In order to provide some further evidence for this interpretation, we devised a novel form of "spike timing dependent", or associative, plasticity in motor cortex. We assumed that PA stimulation would recruit early I-waves whereas AP stimulation would tend to recruit late I-waves. For further analysis, we chose people in whom the PA-LM latency difference lay between 1 and 2 ms and the AP-PA latency difference was equal to or greater than 2 ms (mean of approximately 4 ms). This was because, in people who did not fulfill the criteria (see Methods, and Fig. 6D, cross), the short AP-PA latency difference may indicate that the same neuronal elements are

stimulated by AP and PA currents. Indeed, in some subjects the PA-LM latency difference was shorter than 1 ms or longer than 2 ms implying that a D-wave or later I-waves may have been stimulated by the PA stimulus. Since the number of subjects in each group is too small to draw firm conclusions, we do not discuss the data from these subjects.

The protocol involved giving 60 pairs of PA and AP stimuli separated by an interstimulus interval of 5 ms. We hypothesized that synaptic inputs to corticospinal neurons activated by the first stimulus of the pair would be strengthened if the second stimulus sometimes provoked a corticospinal discharge. If PA and AP stimuli activate inputs that arrive at different times relative to the TMS pulse, as implied in the I-wave hypothesis, then the effects of paired stimulation should differ depending on whether the first stimulus of the

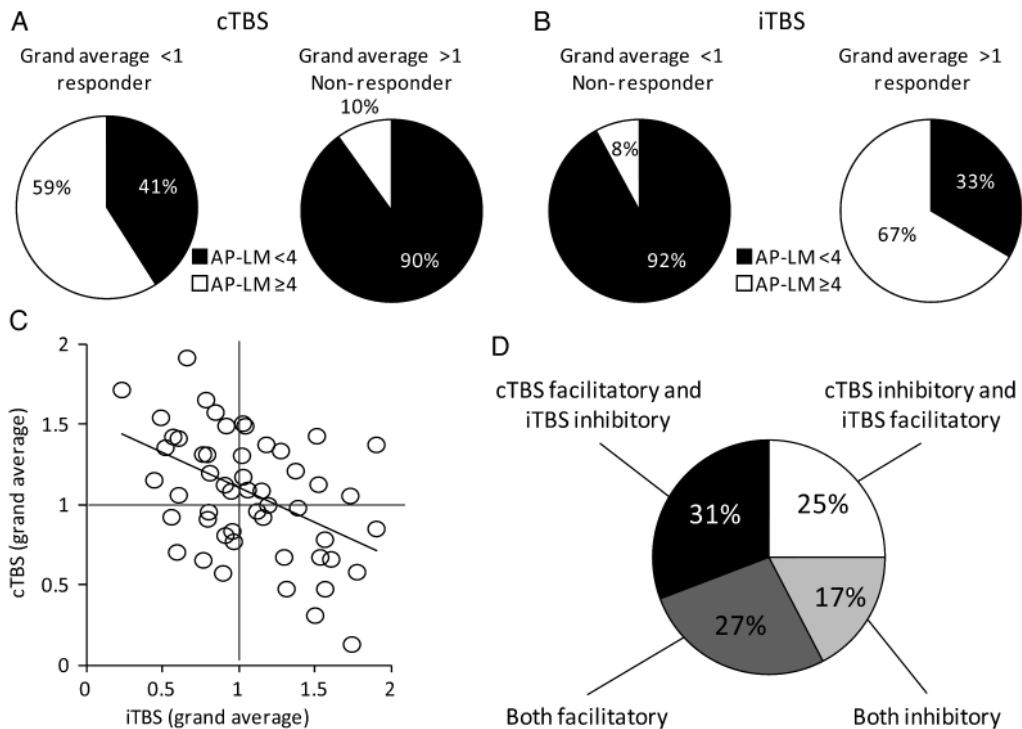


Figure 5. (A) and (B) The percentage of responder and non-responder in each TBS session. Subjects with long AP-LM latency difference (AP-LM ≥ 4) were significantly frequent in responder group. (C) and (D) Correlation between cTBS effects and iTBS effects. (C) Significant negative correlation between cTBS and iTBS effects was found. (D) The number of subjects among four conditions.

pair is an AP or a PA pulse. Thus, in the case of icPAS-5 (PA-before-AP-after pairing), the interval between the arrival of the respective inputs at the corticospinal neuron would be quite long. For example, if the PA pulse activated early I-wave inputs whilst the AP pulse activated later inputs that arrived 4 ms later, then the total interval between the inputs would be 9 ms (i.e. $4 + (\text{ISI})_5$). In contrast, if the order of stimuli was reversed (icPAS + 5: AP-before-PA-after pairing), then the interval between the arrival of the respective inputs would be quite short (1 ms in this example). We might therefore predict that icPAS + 5 would have a larger effect than icPAS-5.

Figure 6B shows that there was a tendency for this to be the case. icPAS + 5 tended to produce a facilitatory after-effect on the response to single-pulse TMS, whereas there was no effect of icPAS-5. However, statistical analysis showed that in fact there were no significant effects overall because of variability of the data. Separate 1-way ANOVAs on the response to icPAS + 5 and icPAS-5 showed no effect of TIME (icPAS + 5, $F_{7,56} = 0.695$, $p = 0.676$; icPAS-5, $F_{7,56} = 0.498$, $p = 0.832$). Similarly, a 2-way repeated measures ANOVA showed no significant effects of icPAS ($F_{1,8} = 0.835$, $p = 0.388$) and no TIME \times icPAS interaction ($F_{2,445, 19,56} = 1.118$, $p = 0.357$). There was no difference in baseline excitability between sessions (baseline MEP size; icPAS + 5, 1.19 ± 0.16 mV; icPAS-5, 1.28 ± 0.18 mV).

Given the data in the first set of experiments we next tested whether the variability in the data was related to the AP-PA latency difference in individual participants. Figure 6D shows that for icPAS+5/-5, there was a significant negative/positive correlation, respectively, to the AP-PA latency difference (icPAS + 5, $r = -0.717$, $p = 0.030$; icPAS-5, $r = 0.877$, $p = 0.002$). In the 5 subjects where we investigated RMTpa before and

after icPAS protocol, it remained constant (icPAS + 5, before, $39.8 \pm 3.3\%$, after, $40.2 \pm 3.2\%$; icPAS-5, before, $40.2 \pm 3.1\%$, after, $40.8 \pm 2.9\%$, not significant). We also confirmed that F-wave amplitudes did not change after intervention (icPAS + 5, before, 191.7 ± 41.7 μV , after, 219.6 ± 33.2 μV ; icPAS-5, before, 223.8 ± 19.8 μV , after, 214.2 ± 21.8 μV , not significant).

To confirm the importance of the oppositely direct current pulses in the icPAS intervention, we performed control experiments using pairs of stimuli with the same direction (i.e. PA/PA or AP/AP; Fig. 6A). We did not observe any effects on MEP excitability after either protocol (Fig. 6C) (1-way ANOVA, AP pairing, $F_{7,56} = 0.702$, $p = 0.670$; PA pairing, $F_{7,56} = 0.669$, $p = 0.697$). Likewise, a 2-way repeated measures ANOVA showed no significant effects of pairing ($F_{1,8} = 0.449$, $p = 0.522$) and no TIME \times pairing interaction ($F_{6,48} = 0.505$, $p = 0.801$). There was no difference in baseline excitability between sessions (baseline MEP size; AP pairing, 0.98 ± 0.06 mV; PA pairing, 1.05 ± 0.08 mV). Figure 6D shows that there was no significant correlation with the AP-PA latency difference for either PA or AP pairings (AP pairing, $r = -0.322$, $p = 0.399$; PA pairing, $r = 0.162$, $p = 0.676$).

Discussion

The present results show that the response to TBS protocols is highly variable between individuals, similar to other plasticity protocols such as 1 Hz rTMS (Maeda et al. 2000) or paired associative stimulation (PAS) (Stinear and Hornby 2005; Bagnato et al. 2006; Fratello et al. 2006; Muller-Dahlhaus et al. 2008). The new finding is that about 50% of this variation was predicted by our postulated marker for the efficiency of late I-wave recruitment: participants in whom

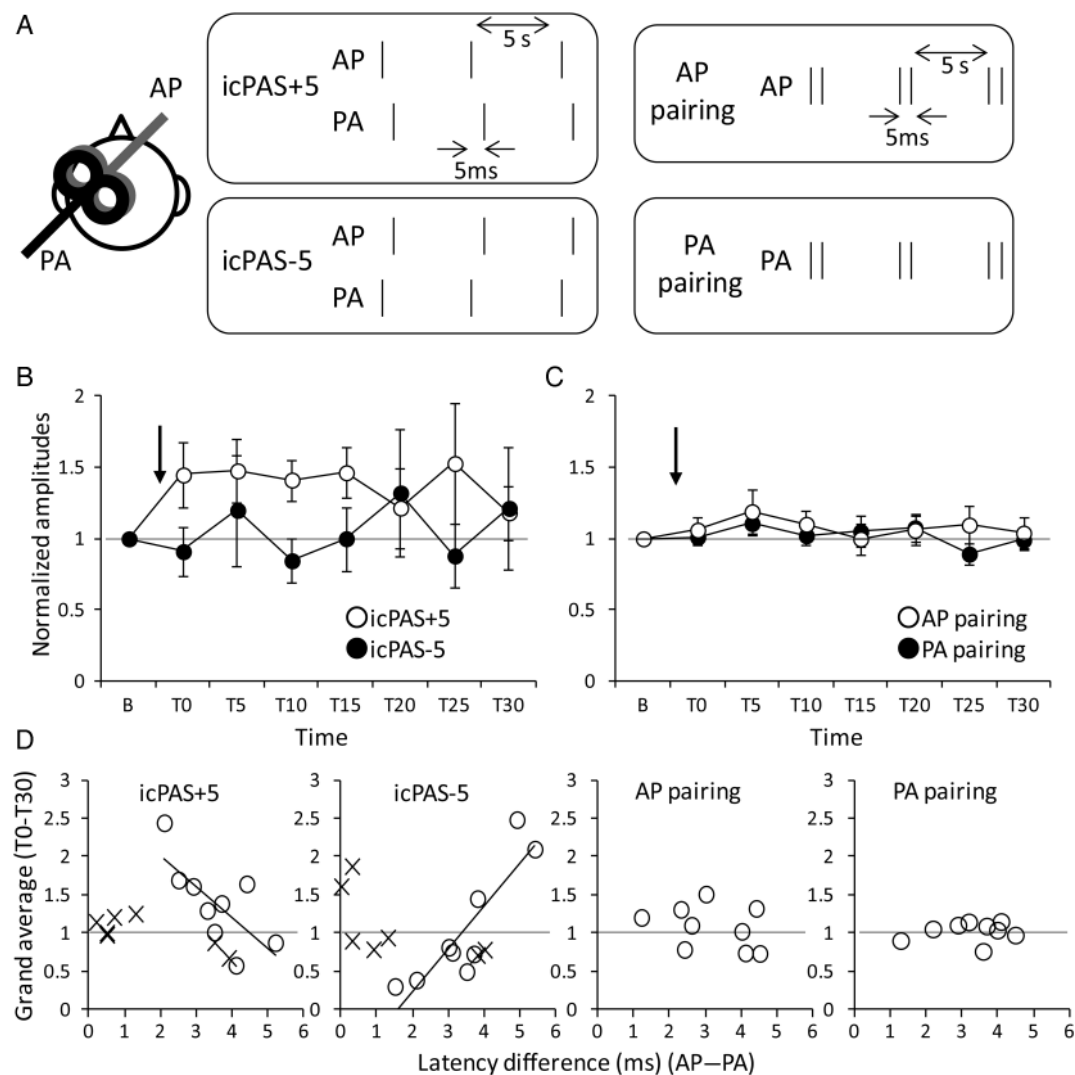


Figure 6. (A) icPAS and pairing protocols. (B) Time courses of icPAS + 5 and icPAS-5. (C) Time courses of AP and PA pairings. Arrow indicates the time of intervention. (D) Correlation between grand average of icPAS + 5, icPAS-5, AP pairing, or PA pairing and AP-PA latency difference. The cross indicates icPAS responses of people who did not fulfill the criteria (see Methods).

there was a large latency difference between MEPs evoked by LM and AP stimulation showed the “expected” inhibition after cTBS and facilitation after iTBS, whereas the opposite was true for people in whom the latency of LM and AP responses was closer. A second set of experiments showed that differential recruitment of I-waves between individuals also influenced the after-effects of a new intracortical spike timing-dependent plasticity protocol. We conclude that much of the inter-individual variation in response to TBS plasticity protocols is due to differences in the population of neurons activated by each TMS pulse rather than differences in the intrinsic amount of synaptic plasticity in cortical neurons.

Variability of TBS Responses

As noted in the Introduction, there is a very large inter-individual variation in the response to TMS protocols that test early forms of synaptic plasticity in the human motor cortex (Ridding and Ziemann 2010). The TBS methods we applied here were no exception; indeed in this group of 52 people,

there was no overall response to either intervention. Approximately one quarter of participants had the “expected” response of inhibition after cTBS and excitation after iTBS, and just under half responded as expected to one but not the other. A number of studies have noted that the effects of TBS protocols can occasionally differ from those originally reported (Martin et al. 2006; Cheeran et al. 2008; McAllister et al. 2011; Goldsworthy et al. 2012; Hasan et al. 2012) and that individual responses can be considerably variable (Gentner et al. 2008; Zafar et al. 2008; Swayne et al. 2009; Todd et al. 2009; Di Lazzaro, Dileone et al. 2011; Mori et al. 2011). However, none of these studies systematically investigated variability of both cTBS and iTBS responses in as large a group of healthy volunteers as we describe here.

The variation in response between individuals was not related significantly to age, gender, time of day, or any initial difference in thresholds, stimulus intensity, and baseline MEP sizes. Furthermore, we can probably discount any influence of prior (Gentner et al. 2008; Iezzi et al. 2008), ongoing or subsequent (Huang et al. 2008) neuronal activity related to

levels of muscle contraction since this was carefully monitored during experiments. The variability presumably reflects some intrinsic difference between people in their response to TBS. Because spinal epidural recordings have shown that cTBS reduces the amplitude of early I-waves whereas iTBS affects late I-waves (Di Lazzaro et al. 2005; Di Lazzaro et al. 2008) we had initially hypothesized that the variability in the response to TBS depends on the relative recruitment of late versus early I-waves: we predicted that people in whom TMS readily recruited early I-waves might have a good response to cTBS whereas those in whom late I-waves were recruited would have better responses to iTBS. The present experiments show that this was not accurate; iTBS and cTBS produce long-lasting facilitation and inhibition, respectively (Huang et al. 2005) in individuals in whom late I-waves are recruited easily. People with poor late I-wave recruitment have opposite responses to the same paradigms.

Postulated Marker of I-Wave Recruitment

Before discussing the physiological relationship between I-wave recruitment and response to TBS, it is important to evaluate the evidence in support of a link between MEP latency and I-wave recruitment which is critical to our argument. First, the reasoning is only valid for MEP latencies measured during background muscle contraction since latencies in relaxed muscle are contaminated by the time taken for multiple corticospinal excitatory postsynaptic potentials (EPSPs) to raise the resting membrane potential of spinal motoneurons above firing threshold (Day et al. 1987; Rothwell et al. 1987). Although larger MEP sizes will have shorter peripheral conduction times because of axon size, there was no significant correlation between the AP–LM latency difference and MEPs evoked by AP currents. It is therefore unlikely that the latency difference is due to the relative sizes of the potentials. Given this proviso, many authors in the past have postulated that the variation of MEP latency in an individual is caused by differential recruitment of I-wave inputs to corticospinal neurons (Day et al. 1989; Werhahn et al. 1994; Sakai et al. 1997; Di Lazzaro et al. 1998; Di Lazzaro, Oliviero, Saturno, et al. 2001; Hanajima et al. 2002; Kujirai et al. 2006; Ni et al. 2011). In particular, Sakai et al. (1997) showed that changes in MEP latency with different directions of induced current corresponded to changes in the time of recruitment of individual single motor units, which previously had been associated with I-wave recruitment by Day et al. (1989) and Di Lazzaro et al. (2001a, 2001b). We think therefore that a substantial body of evidence supports our hypothesis. It could be argued that estimation of onset of individual MEPs, particularly during a background contraction, might be insecure and open to observer bias. However, the consistency across blinded observers using the manual measurements has already been demonstrated in the study by Shirota et al. (2011). In addition, the present results showed a high ICC value between manual and automatic methods, ruling out any potential subjective bias in the observer. Thus, the only way to improve on our measures would be to perform motor unit or epidural recordings in all subjects.

In detail, we suggest that long latency MEPs reflect activation of late I-waves whereas earlier MEPs are the result of early I-waves. The variability of the AP–LM latency difference (Fig. 2) is consistent with this idea since previous work has

shown that the order of I-wave recruitment by AP currents is much more variable than those recruited by PA currents (Di Lazzaro, Oliviero, Saturno, et al. 2001). Interestingly, Sakai et al. (1997) found a more consistent AP–LM latency difference (about 4–5 ms) in all seven subjects they investigated. Although it seems likely from the much larger number of individuals studied here that this was a chance observation, we cannot discount the possibility that there may be racial differences between Japanese and (predominantly, in the present study) European participants.

Finally, we located the TMS coil at the same spot using both AP and PA currents as well as biphasic (when applying TBS) stimulation. Previous work has shown that the “hot-spots” for AP and PA orientations are coincident (Sakai et al. 1997; Arai et al. 2005). In addition, all previous TBS studies in human primary motor cortex employ the hotspot determined by PA currents. The biphasic TMS pulses used during TBS preferentially activates cortical neurons on the reverse phase of the stimulating current (Maccabee et al. 1998; Di Lazzaro, Oliviero, Mazzone, et al. 2001), and therefore MEPs evoked by a monophasic AP pulse are likely to represent activation of the same elements.

Physiological Mechanisms of TBS Effects: the Role of Interneuron Networks

The results illustrated in Figure 4E and F show that the response to TBS protocols is strongly related to which I-waves are likely to be recruited by the TMS pulses. People in whom late I-waves are recruited have the “expected” responses to cTBS and iTBS, whereas those in whom early I-waves are recruited demonstrate the “opposite” effect. There is uncertainty about the mechanism and cellular location of I-wave inputs to corticospinal neurones (CSNs) and there are no data on the rules for synaptic plasticity at each site. However, according to the canonical model of cortical circuitry (Di Lazzaro, Profice, et al. 2011), early I-waves are likely to reflect monosynaptic input to CSNs from layer II and III interneurons whereas more complex oligosynaptic circuits including inhibitory neurons are involved in late I-wave generation. Amassian et al. (1987) additionally suggested that early I-wave inputs are located on proximal parts of the CSNs whereas later I-waves target more distal dendrites. Our results would therefore be compatible with the idea that the oligosynaptic inputs evoked on distal dendrites behave differently to the monosynaptic proximal inputs (Golding et al. 2002; Branco and Häusser 2011). It could be, for example, that the effects of dendritic inputs are governed by local generation of dendritic potentials whereas the effect of proximal inputs rely on somatic potentials (Williams and Stuart 2002). However, we cannot dismiss the possibility that modulation of late I-waves is the result of changes in synaptic connections to neurons other than CSNs (Amassian et al. 1987).

It is also important to consider the contribution of inhibitory inputs to the overall response to TBS protocols. Recent studies in rat cortex have shown that iTBS acutely increases activity of fast-spiking (FS) inhibitory neurons which have terminals located mainly around the soma, whereas cTBS modulates non-FS neurons that predominantly regulate dendritic activity (Markram et al. 2004; Benali et al. 2011; Funke and Benali 2011). If late I-wave inputs are located on dendrites, then activation of non-FS inhibition during cTBS might

favour synaptic suppression by increasing ongoing levels of GABAergic inhibition (Steele and Mauk 1999), which tends to reduce LTP-like effects. Activation of FS interneurons by iTBS might suppress excitability at the soma, isolating dendritic integration of late I-wave inputs. In addition, iTBS induced GABAergic activation onto the soma might favour LTD-like effects in individuals where TMS activates predominantly early I-wave inputs. A location-specific role of inhibitory interneurons in practice dependent plasticity has also been postulated in experiments in humans (Teo et al. 2009).

This type of arrangement can be used to explain the experimental data showing that cTBS reduces both early and late I-waves (Di Lazzaro et al. 2005) even though our model implies that only late I-waves are important contributors of the response to cTBS (and iTBS). Although long AP–LM latencies indicate that later I-waves are easily recruited, there is a spread of data in any one individual and on some occasions, early I-waves may be recruited by the same stimulus. This might then equate to intermittent stimulation of early I-wave inputs, which could lead to inhibition of early I-waves (cf. people who recruit early I waves have an inhibitory response to iTBS). Another interesting possibility based on our model is that the effects of TBS could be clearer if tested with MEPs by AP currents. We did not test this possibility in the present study because we tried to avoid any possible influence on TBS after-effects: indeed, high stimulus intensities are often needed to evoke MEP sizes around 1 mV with AP currents, and this could potentially change any after-effects of TBS.

Our hypothesis tries to reconcile previous and present results of TBS with knowledge obtained from animal experiments. More complex scenarios could be configured within the framework of metaplasticity theory (Gentner et al. 2008; Hamada et al. 2008; Hamada et al. 2009), gating hypothesis (Gamboa et al. 2010; Siebner 2010), the theoretical model of TBS (Huang et al. 2011), or differential generation of later I-waves (Di Lazzaro, Profice, et al. 2011; Ni et al. 2011). Currently, none of these assumptions can provide a simple explanation of the linear correlation of TBS effects with AP–LM latency difference. Further work is needed to understand how and where on the CSNs, excitatory and inhibitory inputs mutually interact during TBS.

Further Evidence to Support Different Effects on Synaptic Plasticity of Early and Late I-Wave Inputs

The final set of experiments was devised to try to confirm that the rules for synaptic plasticity are different for early and late I-wave circuits. The icPAS protocol involved pairing a standard PA stimulation with AP stimulation at 5 ms interval. Repeated pairing led to long-lasting changes in excitability that were again related to the AP–PA latency difference. Since there were no changes in RMT or F-waves, it seems likely that the effects of icPAS are cortical in origin and reflect short-term changes in synaptic effectiveness similar to those observed after applying peripheral nerve/TMS PAS protocols (Stefan et al. 2000; Wolters et al. 2003). If so, then the results are consistent with the idea that the neural populations recruited by TMS are critical for determining the net direction (suppressive or facilitatory) of long-term effects on corticospinal excitability.

We have no direct information on the mechanism of the icPAS effect. Our hypothesis was that it recruits spike timing dependent plasticity (STDP) according to a “pre-post” rule in which synapses are strengthened if a presynaptic input is regularly followed by a postsynaptic discharge (Caporale and Dan 2008). By definition, AP and PA stimuli at threshold can produce substantial depolarization (and perhaps on occasion an action potential) in at least some CSNs. If the likelihood of discharge increases when one input is preceded by the other, then AP-before-PA-after pairing at ISI at 5 ms (icPAS + 5) would mean that late I-wave inputs (from AP stimulation) would be followed by a postsynaptic discharge (from the later PA input). According to the pre-post rule, the AP input would be strengthened. Figure 6D shows that maximum facilitation occurred when the interval between MEPs evoked by PA and AP stimulation was 2 ms. Given an ISI of 5 ms, this corresponds to a “pre-post” interval of 3 ms that is within the range of STDP window (Dan and Poo 2006). Rapid decline of facilitation as the MEP latency difference increases can be accounted for by the assumption that maximum STDP occurs at 3 ms in human motor cortex since previous STDP-like plasticity by PAS has relatively narrow time window compared with animal experiments (Wolters et al. 2003).

This simple “pre-post” model, however, fails to account for the data from icPAS-5 (i.e. PA-before-AP-after pairing), where maximum facilitation occurs when the MEP latency difference is 5 ms. In this case, the “pre-post” interval is 10 ms (corresponding to early I-waves evoked by PA followed by very late I-waves from AP). Although this timing fits with the relatively wide STDP window in animals (Dan and Poo 2006), we would expect this to be ineffective under normal conditions if we assume optimal timing for STDP in humans is 3 ms as described above. Furthermore, excitatory inputs separated by this interval are unlikely to summate and produce an action potential after the AP input. An alternative possibility is that very late I-wave inputs are located on distal dendrites, where the rules for STDP are reversed (Letzkus et al. 2006). In this case, it would be possible that even though the late I-waves occur after PA inputs, they may be strengthened by any preceding cell discharge which is occasionally produced by the PA pulse.

We did not observe any plasticity using pairs of stimuli with the same direction of current pulses (Fig. 6C). Taken together with the findings of icPAS, it is likely that inputs have to be in separate synapses for the icPAS effect to occur. Our findings are consistent with animal studies of STDP showing that repetitive bursts of EPSPs alone did not induce any plasticity (Markram et al. 1997). In addition, this might also explain why long-lasting facilitation is not observed after repeated high intensity single-pulse TMS in which both early and later I-waves are recruited. Repetitive single-pulse TMS might be expected to induce plasticity in early I-waves since each stimulus leads to an action potential in the CSNs, and this should reinforce at least some of the synapses that were active beforehand. However, we presume that this occurs in a very random way. If STDP rule is applied to this synaptic modulation, some late I-wave activation would be suitable, in terms of timing, to induce synaptic plasticity (such as LTP), but others would be unsuitable for inducing LTP and would instead be appropriate for LTD, for example. As a result, we may not observe any plasticity overall.

In summary, although the precise mechanisms of icPAS are still uncertain, we have found that the latency of I-wave inputs recruited by TMS pulses substantially affects synaptic plasticity within motor cortex. These results therefore support our hypothesis that synaptic inputs recruited by AP and PA stimulation differ between different subjects and that this determines the direction of change in presumed synaptic plasticity.

General Implications

The main implication from this study is that the relative recruitment of early and late I-waves has an important influence on the after-effects of TBS protocols. The variation in MEP latencies in the present experiments was highly repeatable from day to day and therefore this could be one major factor in inter-individual variation. However, this is not the only source of variability. The day-to-day variation within an individual to TBS protocols has not been examined systematically but by analogy with the effects of rTMS (Maeda et al. 2000; Fratello et al. 2006) is likely to be high. Other factors must account for this temporal variation. Despite the presence of these other drivers of variance, only 25% of the subjects showed “expected” responses to TBS, while 31% of the subjects showed “opposite” responses. It is generally assumed that TBS induces LTP or LTD-like plasticity depending of the pattern of TBS (Huang et al. 2005). The strength of the association between MEP latencies and TBS response means that it is not possible to assume that the amplitude of the response to TBS is necessarily a measure of the intrinsic level of synaptic plasticity in the cortex. In other words, it is likely that the different forms of TBS do not specifically induce LTP or LTD-like plasticity, but have different effects on circuits activated by early and late I-waves. This is in line with recent results showing that the same form of cTBS can induce LTP or LTD depending on the current state of the cortex (Gentner et al. 2008). The notion further suggests that the lack of LTP or LTD-like plasticity after TBS in Parkinson’s disease may not be due to the altered synaptic plasticity in the cortex (Suppa et al. 2011; Kishore et al. 2012), but may simply reflect the variability of TBS we observed in the present study. Furthermore, efficiency of late I-wave recruitment may also be relevant to the recent conflicting results for TBS responses in Parkinson’s disease (Bologna et al. 2012; Zamir et al. 2012) and even for genetic effects on stimulation-induced plasticity (Cheeran et al. 2008; Antal et al. 2010; Nakamura et al. 2011). Measurement of MEP onset latency is easy and rapid, offering practical benefits to predict TBS responses beforehand in each subject. This may be of particular benefit for optimization of TBS paradigms in therapeutic settings. To improve TBS responses further, it might be useful to explore more fully the optimal position for late I-wave recruitment.

Conclusions

We found that efficiency of late I-wave recruitment as well as the after-effects of iTBS and cTBS protocols is highly variable among subjects. Furthermore, the after-effects of TBS and icPAS correlated with the ease of late I-wave recruitment. The present results provide evidence that variation in response to rTMS plasticity probing protocols is strongly influenced by which neuronal networks are recruited by each TMS pulse.

Our findings further suggest that the rules for synaptic plasticity are different for early and late I-wave circuits.

Funding

This work was supported by EU FP7 Collaborative Project (223524: PLASTICISE). Masashi Hamada is supported by the Japan Society for the Promotion of Science Postdoctoral Fellowships for Research Abroad. Alkomiet Hasan is supported by the Deutsche Forschungsgemeinschaft (DFG grant HA 6091/1-1).

Notes

We are grateful to Dr Binith Cheeran and Dr James T. H. Teo for constructive comments and support for data analysis. We would also like to thank Ms Louise Marshall for English editing. *Conflict of Interest:* The authors declare no potential conflicts of interest relating to the subject of this report.

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