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Repetitive TMS over the human oculomotor cortex: Comparison of 1-Hz and theta burst stimulation

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Abstract

The aim of the study was to compare the effect duration of two different protocols of repetitive transcranial magnetic stimulation (rTMS) on saccade triggering. In four experiments, two regions (right frontal eye field (FEF) and vertex) were stimulated using a 1-Hz and a theta burst protocol (three 30 Hz pulses repeated at intervals of 100 ms). The same number of TMS pulses (600 pulses) was applied with stimulation strength of 80% of the resting motor threshold for hand muscles. Following stimulation the subjects repeatedly performed an oculomotor task using a modified overlap paradigm, and saccade latencies were measured over a period of 60 min. The results show that both 1-Hz and theta burst stimulation had inhibitory effects on saccade triggering when applied over the FEF, but not over the vertex. One-hertz rTMS significantly increased saccade latencies over a period of about 8 min. After theta burst rTMS, this effect lasted up to 30 min. Furthermore, the decay of rTMS effects was protocol-specific: After 1-Hz stimulation, saccade latencies returned to a baseline level much faster than after theta burst stimulation. We speculate that these time course differences represent distinct physiological mechanisms of how TMS interacts with brain function.

Keywords: Human; rTMS; Saccades; Frontal eye field; Stimulation protocol

Repetitive transcranial magnetic stimulation (rTMS) is a widely used technique in human brain research. A train of repetitive pulses can induce a modulation of cortical excitability and allows investigating cortical functioning. Depending on the frequency of stimulation, this effect may range from inhibition to excitation. In the motor cortex, low frequency rTMS, i.e. trains at frequencies of 1-Hz or lower, result in a decrease of cortical excitability whereas high frequency rTMS, i.e. trains with a stimulation frequency of 20 Hz or more lead to an increase of cortical excitability (for a review see [6]). In such studies, the size of motor evoked potentials is used as a measure of cortical excitability, since other parameters such as strength and speed of contraction have shown to be unsuitable for this purpose [8]. The observed changes of motor evoked potentials after low or high frequency rTMS can extend beyond the stimulation period but are usually short term. Only recently, Huang et al. [5] have shown that the lifetime of electrophysiological changes can be substantially extended with a TMS protocol, which was modified from theta burst stimulation protocols used in animal preparations to induce long-term potentiation (LTP) [1]. They found that the motor evoked potential was reduced in its amplitude for up to 1 h.

We have recently shown [11] that low frequency rTMS has an inhibitory behavioural effect on saccade triggering that extended beyond stimulation. Six hundred pulses applied with 1-Hz over the right frontal eye field (FEF) delayed saccade triggering for about 10 min. The aim of the present study was (1) to evaluate the effect of theta burst stimulation of the FEF on saccade triggering and (2) to compare the effect duration of theta burst rTMS with that of the known inhibitory effect of 1-Hz rTMS in the same subjects. Using the oculomotor system to study rTMS induced modulation of cortical functioning has the advantage of producing easily and precisely quantifiable parameters. For

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instance, stimulation of the frontal eye field, which is crucially implied in saccade triggering [14], can serve as model to evaluate rTMS effects by assessing the latencies of horizontal saccades in response to a visual target (for a review, see [10]).

Three right-handed, male subjects participated in the study (mean age: 38 years, range: 29–48 years). All had normal or corrected to normal vision. The study was approved by the ethical committee of the State of Bern and is consistent with the Declaration of Helsinki. All subjects gave written consent prior to participation.

The same oculomotor paradigm was used as previously described [11]. Each trial started with a central fixation point. A lateral target with unpredictable amplitude at 4, 8, 12 or 16° left or right from the central fixation point was shown for 80 ms after a pseudo-randomised duration between 2000 and 2900 ms. Subjects were instructed to make a saccade to the position where the target was shown, and then to fixate again the central fixation point. The experiment was conducted in total darkness and the subject's head was fixed on a chin rest to avoid head movements. Eye movements were measured with an infrared corneal reflection device (Iris Skalar, Delft, The Netherlands) with a spatial resolution of 0.1° and a sampling rate of 1000 Hz. The digitized signals were stored on the computer for off-line analysis.

Repetitive biphasic magnetic pulses were generated with a TMS stimulator (MagPro, Medtronic Functional Diagnostics, Skovlunde, Denmark). The magnetic pulses were delivered by a figure-eight-coil (Magnetic Coil Transducer MC-B70, Medtronic) with an outer radius of 50 mm. For all experiments, the stimulation strength was set at 80% of the subject's resting motor threshold of the small hand muscles.

With each subject, four TMS experiments were performed, with a time-interval between experimental sessions by at least 1 week. In each experiment, 600 TMS pulses were applied. In the first experiment, the right FEF was stimulated using a 1-Hz protocol as previously described [11]. In the second experiment, which served as control condition, 1-Hz rTMS was applied over the vertex. In the third experiment, the right FEF was stimulated using a theta burst protocol of 200 bursts. Each burst consisted of three pulses at 30 Hz, repeated at intervals of 100 ms. Thus, this stimulation protocol is more than 10 times shorter than the 1-Hz protocol. Finally, in the fourth experiment, theta burst rTMS was applied over the vertex as a control condition.

The FEF was localised according to previously described procedures [12,9]. In brief, the right motor cortex was stimulated with single pulses to determine the individual motor threshold by corresponding muscle twitching of the subject's relaxed small hand muscles. The coil was then moved anterior to the hand area by 2 cm. The handle of the coil pointed backwards in a 45° angle to the sagittal line. For vertex stimulation, the centre of the coil was held over the vertex and the handle pointed backwards. The subjects were asked to keep their eyes closed throughout the stimulation procedure.

In the first and second experiment (1-Hz stimulation), the recording device was calibrated immediately after rTMS application, and the oculomotor paradigm was performed in seven blocks, each lasting on average 40 s. In each block, seven saccades to the left and seven saccades to the right side were performed in total, the direction of the saccades was pseudo randomized. Between two blocks a pause of 80 s was included. In the non-TMS condition, the same oculomotor paradigm was also performed without stimulation to assess baseline values.

In the third and fourth experiment (theta burst stimulation), the device was calibrated after stimulation, and saccades were performed in five blocks each containing 42 saccades with the direction of the saccades being pseudo randomized to obtain the same number of leftward and rightward saccades. The blocks started immediately after rTMS application, i.e. after 0, 10, 20, 30, and 60 min. This different task schedule with longer measurement intervals for the theta burst protocol was chosen since a pilot study of our laboratory has suggested that theta burst rTMS over the FEF has much longer effects on saccade latencies.

Finally, mean latencies for leftward and rightward saccades of the non-TMS condition were calculated for each subject.

Latencies with stimulation were then standardized to the non-TMS (baseline) values, i.e. for a given individual of a given experiment, the percentage of increase in saccade latency was calculated for each saccade with the following formula: $100\% \times [(\text{latency TMS}/\text{mean latency without TMS}) - 1]$. Thus, a value of 0% means no TMS effect.

The statistical analysis of the four experiments was based on a repeated measures ANOVA with dependent variable "block" (increase in latency) and one categorical factor "side" (leftward, rightward saccades). Bonferroni corrected post-hoc comparisons between stimulation and no stimulation were performed with least squares means for blocks (Statistica 6.0, StatSoft, Inc., Tulsa, Oklahoma, USA).

Saccade latencies without stimulation and with different stimulation conditions are presented in Table 1. Both 1-Hz ($F(7, 217) = 21.31$, $p < 0.0001$) and theta burst rTMS ($F(5, 420) = 30.71$, $p < 0.0001$) over the FEF resulted in a significant increase of saccade latency, whereas stimulation over the vertex had no significant effect (Fig. 1). In all experiments, no significant effect for side was found, therefore we pooled left- and rightward saccades for the analysis and presentation of the results.

Table 1
Mean saccade latencies (ms) after 1-Hz and theta burst rTMS

	1 Hz stimulation		Theta burst stimulation	
	FEF	Vertex	FEF	Vertex
	Mean (S.E.M.)		Mean (S.E.M.)	
No stimulation	213 (4)	213 (4)	213 (4)	213 (4)
Time after stimulation				
0 min	292 (12)	226 (8)	267 (9)	194 (4)
2 min	319 (16)	215 (7)		
4 min	310 (18)	224 (9)		
6 min	293 (12)	226 (8)		
8 min	247 (11)	218 (9)		
10 min	238 (9)	213 (7)	258 (8)	201 (4)
12 min	227 (9)	226 (9)		
20 min			247 (7)	204 (5)
30 min			229 (7)	209 (4)
60 min			211 (5)	198 (4)

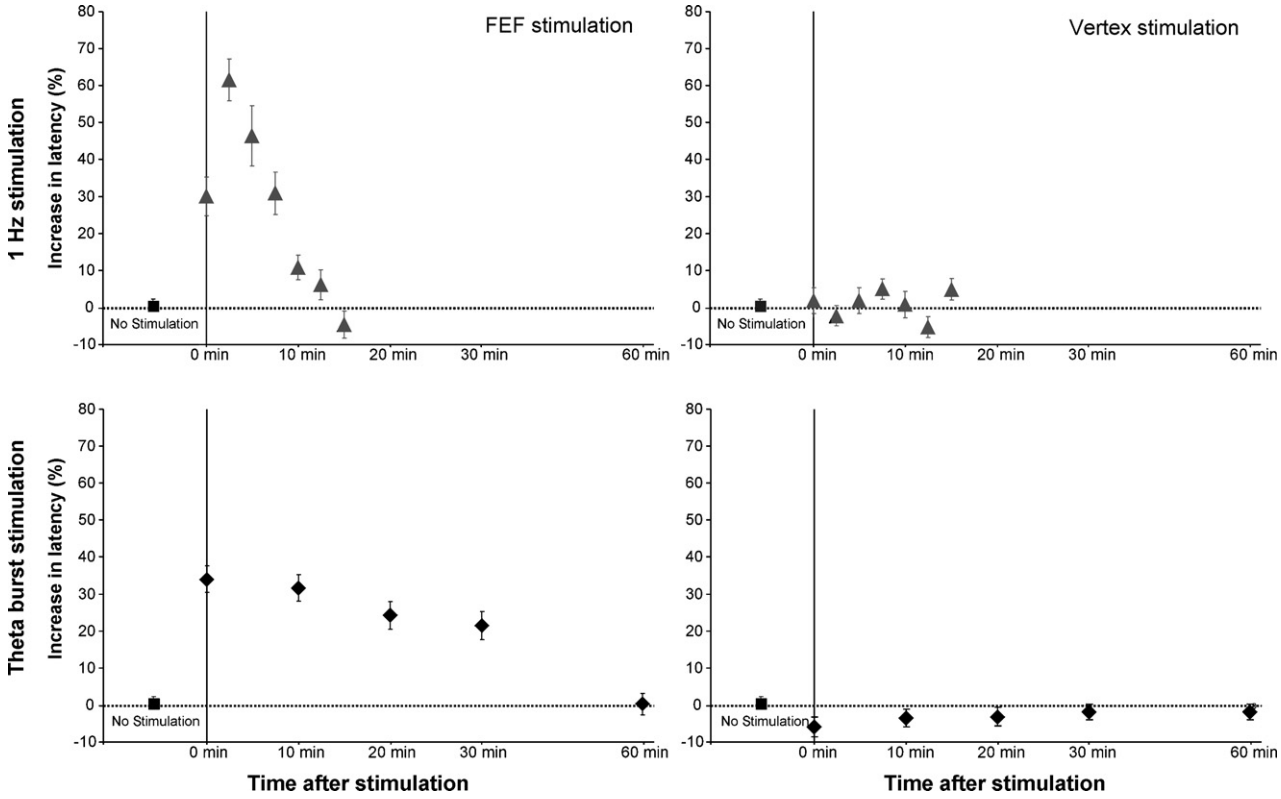

Fig. 1. Effect of 1-Hz and theta burst rTMS over the FEF and vertex on saccade latency (mean increase in saccade latency (in percent) and S.E.M. is shown). Both rTMS protocols (with the same number of TMS pulses) increased saccade latency when applied over the FEF but not when applied over the vertex. The duration of the effect lasted longer if the theta burst TMS protocol was applied. One-hertz stimulation significantly increased saccade latency up to 8 min, whereas the effect of theta burst TMS significantly lasted until 30 min.

Bonferroni corrected post-hoc comparisons showed a significant increase of saccade latencies after 1-Hz rTMS for about 8 min (from block 1–4: $p < 0.0001$). Latency increased from block 1 (mean increase 30%) to block 2 (mean increase 62%). Then, from block 3 to block 7, latency decreased again (block 3: mean increase 46%; block 4: mean increase 31%; block 5: mean increase 11%; block 6: mean increase 6% and block 7: mean decrease 4%).

Bonferroni corrected post-hoc comparisons showed a significant increase of saccade latencies after theta burst rTMS for about 30 min (for the first four measurements: $p < 0.001$). The latency increased by 34% after stimulation and remained at 32% after 10 min, at 24% after 20 min, at 22% after 30 min until it reached again 0.5% after 1 h.

The present study shows that both theta burst and 1-Hz rTMS over the right FEF are able to induce a robust delay in saccade triggering. This effect was specific since stimulation over the vertex had no significant effect. In both rTMS protocols, saccade latency significantly increased: 1-Hz stimulation induced an inhibitory effect for a few minutes as already described [11]. Theta burst rTMS, however, induced a much longer lasting effect of up to 30 min.

Theta burst rTMS protocols have so far been used in motor or premotor cortex physiology [5,7,4]. In two studies, motor evoked potentials were found to be reduced suggesting an inhibitory effect [5,4]. Mochizuki et al. found a behavioural effect in a hand reaction task [7] with increased choice reaction times of both hands for a period of 5–10 min.

The present study now shows that lasting inhibitory behavioral effects can also be observed after theta burst stimulation of the oculomotor cortex. By comparing its effect duration with that of 1-Hz rTMS, the results further show that the theta burst protocol seems to induce more stable plastic changes in the human brain. How rTMS interferes with physiological mechanisms in the human brain is far from clear: The effect of low frequency rTMS has been related to long-term depression (LTD) of cortical synapses [3,16]. Another study could not find evidence for direct involvement of LTD, but suggested a reduction of the excitability of cortical neurones themselves [15]. The effect of the theta burst rTMS protocol has been discussed within the context of LTD and LTP [5]. These authors suggested that theta burst TMS may produce a mixture of facilitatory and depressing effects on synaptic transmissions.

The present results suggest that 1-Hz and theta burst rTMS induce different physiological mechanisms. Although the identical number of pulses with the same stimulation strength was applied, the increased saccade latency following 1-Hz rTMS normalized with a sharp drop within a few minutes, whereas following theta burst rTMS a gradual return to baseline values over a period of an hour was observed. Whether the extension of saccade latencies originates in a change of excitability or in synaptic modifications remains speculative. The time course of

the 1-Hz rTMS effect in the present study is comparable to that observed after post-tetanic modification of synaptic transmission [17,13]. The finding that the effect of theta burst rTMS lasts over 30 min, however, suggests that more stable, perhaps morphological alterations take place. Indeed, in animal preparations, LTP induced by theta burst stimulation that lasts for 1 h depends on gene transcriptions, protein synthesis, and morphological changes [2].

In conclusion, the results show that both theta burst rTMS and 1-Hz rTMS are able to delay saccade triggering. The induced lifetime of saccade delaying, however, is about three times longer after theta burst rTMS. These findings suggest that theta burst rTMS induces more stable plastic changes in the human brain, perhaps morphological alterations, similar to that observed in animal studies.

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