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Cerebellar brain inhibition in the target and surround muscles during voluntary tonic activation

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Abstract

Motor surround inhibition is the neural mechanism that selectively favors the contraction of target muscles and inhibits nearby muscles to prevent unwanted movements. This inhibition was previously reported at the onset of a movement, but not during a tonic contraction. Cerebellar brain inhibition (CBI) is reduced in active muscles during tonic activation; however, it has not been studied in the surround muscles. CBI was evaluated in the first dorsal interosseus (FDI) as the target muscle, and the abductor digiti minimi (ADM), flexor carpi radialis (FCR), and extensor carpi radialis (ECR) as surround muscles during rest and tonic activation of FDI in fourteen subjects. Cerebellar stimulation was performed under MRI-guided neuronavigation targeting lobule VIII of the cerebellar hemisphere. Stimulus intensities for cerebellar stimulation were based on the resting motor cortex threshold (RMT) and adjusted for the depth difference between the cerebellar and motor cortices. We used 90% to 120% of adjusted RMT as the conditioning stimulus intensity during rest. The intensity that generated the best CBI at rest in the FDI was selected for use during tonic activation. During selective tonic activation of FDI, CBI was significantly reduced only for FDI but not for the surround muscles. Unconditioned MEP sizes were increased in all muscles during FDI tonic activation compared to rest, despite background EMG activity increasing only for the FDI.

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Our study suggests that the cerebellum may play an important role in selective tonic finger movement by reducing its inhibition in the motor cortex only for the relevant agonist muscle.

Keywords

cerebellar brain inhibition (CBI); tonic activation; surround muscles; cerebellum; TMS

Introduction

Surround inhibition (SI) is the physiological mechanism where central signals are facilitated and eccentric signals are inhibited. It is well documented that in the sensory system it aids the spatiotemporal discrimination of the sensory inputs. In the motor system, surround inhibition (mSI) is the mechanism that supports the selective activation of target muscles to produce desired movements and inhibition of nearby muscles to prevent unwanted movements (Beck & Hallett 2011). This inhibition has been described in the adjacent muscles prior to onset and at onset of the movement (Beck et al. 2008, 2009, 2010; Kassavetis et al. 2011, 2014; Sohn & Hallett 2004a). It was demonstrated by suppression of MEPs of the surround muscles not involved in the task using transcranial magnetic stimulation (TMS) of the primary motor cortex at the onset of movement (Beck et al. 2008; Sohn & Hallett 2004a). The electrophysiologic findings in healthy subjects and focal hand dystonia patients suggested that the inhibitory networks in the motor cortex and the basal ganglia modulate the surround inhibition despite increased spinal excitability (Beck et al. 2008, 2009, 2010, 2011; Kassavetis et al. 2011, 2014; Mink 1996; Shin et al. 2009, Sohn & Hallett 2004a, 2004b; Stinear and Byblow 2004). However, no mSI was found during the tonic phase of contraction, despite isolated activation of the target hand muscle only (Beck et al. 2008). Thus, there might be other inhibitory mechanisms at the supraspinal level responsible for crafting the selective contraction of only one muscle during the tonic phase.

Cerebellar brain inhibition (CBI) is the parameter quantifying the inhibitory projection from the cerebellar cortex to the dentate and then influencing the motor cortex by reduction of facilitatory activity in the dentato-thalamo-cortical pathway (Ugawa et al. 1995). CBI has been demonstrated to be reduced in active muscles during tonic contraction (Pinto and Chen 2001). Interactions within the motor cortex and between the motor cortex and the basal ganglia have been speculated to be important in mSI (Beck & Hallett 2011). Cerebellar circuits are anatomically connected both to the basal ganglia and the motor cortex. A study in primates showed that the subthalamic nucleus and the striatum have anatomical connections to the cerebellar cortex and dentate nucleus (Bostan et al. 2010; Hoshi et al. 2005). Hence, it is possible that cerebellum might be able to modulate any interactions between the motor cortex and the basal ganglia, including mSI. Moreover, CBI can alter the inhibitory network at the motor cortex level where mSI occurs (Daskalakis et al. 2004). CBI modulation has not been studied in surround muscles during tonic activation phase. We conducted an exploratory study to probe this modulation. Our hypothesis was that CBI would be reduced in target muscles, but not in surround muscles during tonic activation.

Material and Methods

Participants

21 healthy volunteers (HVs) (9 women and 12 men, ages 41–71 years, mean age $\pm$ standard deviation (SD) 57.3 ± 8.5) participated in this study. All subjects were right-handed by Edinburgh Handedness Inventory scale, with no history of neurological or psychiatric disorders and were not taking any medications. Informed consent was obtained before participation. The experiments conformed to the Declaration of Helsinki and were approved by the Institutional Review Board of the National Institutes of Health.

Recording

Electromyography (EMG) activities were recorded by using disposable surface Ag-AgCl electrodes. Surface electrodes (7×4 mm) were placed on the right first dorsal interosseus (FDI), abductor digiti minimi (ADM), flexor carpi radialis (FCR) and extensor carpi radialis (ECR) muscles using pairs of electrodes in a belly-tendon montage. The EMG signal was amplified using a conventional EMG machine (Viking Inc., Tokyo, Japan) with bandpass between 10 and 2000 Hz. The signal was digitized at a sampling rate of 5 kHz and fed into a computer that recorded with Signal software version 5.02 (Cambridge Electronic Design, Cambridge, UK) for offline analysis.

Motor task

All tests were performed at rest and during tonic activation of FDI. Subjects were seated in a comfortable chair, relaxed their right hand and forearm on the pillow, and the tip of their index finger was placed on a force transducer. During the movement task, they were instructed to flex their right index finger at the metacarpo-phalangeal joint constantly at 10% of their maximum voluntary force and keep other muscles relaxed. A relatively weak voluntary force minimizes overflow movements into nearby muscles. Continuous EMG of all muscles ensured that only the target muscle was activated. The individual force level was displayed on an oscilloscope placed in front of the subjects. For index finger flexion, FDI is a synergistic muscle rather than the primary muscle. However, a previous experiment with this FDI movement showed motor surround inhibition in ADM during the phasic phase (Sohn and Hallett 2004a). A practice session with this motor task, keeping the surround muscles relaxed, was required before the experiment. The subjects were asked to practice until they were able to consistently perform the task.

Transcranial Magnetic Stimulation (TMS)

CBI was obtained from paired-pulse TMS stimulation with conditioning stimulation (CS) delivered to the cerebellar cortex ipsilateral to the explored hand, followed 5 ms later by a test stimulus delivered to the contralateral motor cortex. The test stimulation was applied over the left M1 corresponding to contralateral FDI with a figure-of-eight coil with an external loop diameter of 70 mm, connected to the Bistim2 (Magstim, Whitland, Dyfed, UK). The optimal location and coil orientation for stimulation were based on the area for eliciting the best MEPs from the right FDI and optimal MEPs from the ADM, FCR and ECR. This location was recorded and maintained throughout the whole experiment using

MRI-based neuronavigation. At this location, the input-output curves (IO curves) parameters were obtained by delivering single TMS pulses at intensities in random order from 0 to 100% (increasing at 5% intervals) of the maximum stimulator output. Three pulses were delivered for each intensity. MEPs were recorded and their amplitudes were plotted against the corresponding stimulation intensity. The parameters of the resting motor threshold (RMT) and S50 were obtained from IO curves. S50 is the stimulation intensity that produces an MEP halfway between the EMG base and maximum MEP from IO plots using Boltzmann sigmoidal fitted. The RMT was determined using the adaptive threshold hunting method from Awiszus (Awiszus 2003). The intensity of the test stimulation (TS) was set at S50 both for all experimental conditions.

For the cerebellar CS, a double-cone coil (110 mm diameter) was positioned over the right cerebellum. The center of the coil was placed 3 cm lateral and 1 cm inferior to theinion to stimulate the cerebellar hand representations in lobule VIII of the cerebellum and the location was confirmed with MRI-based neuronavigator (Hardwick et al. 2014). This target is the same location used in the previous CBI experiments (Daskalakis et al. 2004; Ugawa et al. 1995). Stimulus intensity at the cerebellar cortex was set at 90–120% of the adjusted RMT (Popa et al. 2010). Adjusted RMT was calculated with the equation of Stokes et al (Stokes et al. 2005), used for correcting the effect of distances when TMS is targeting deeper structures: $Adjusted\ RMT = RMT + M \times (D_{cerebellum} - D_{M1})$, where RMT is the unadjusted RMT (in percentage of maximum stimulator output obtained from the adaptive threshold hunting methodology of Awiszus); $D_{cerebellum}$ is the shortest distance between the scalp surface and the target in the cerebellar cortex; D_{M1} is the shortest distance between the scalp surface and motor cortex (M1); and M is the spatial gradient relating RMT to distance. In cases in which the spatial gradient is unknown, an approximate derivation of distance-adjusted MT can be obtained by replacing with the average gradient that is $= 3$ (Stokes et al. 2005). The distance from the scalp to cerebellar cortex and motor cortex was measured for each individual with the MRI based neuronavigation system. (Figure 1) The coil current direction was downward to induce upward current in the cerebellar cortex. For the best cerebellar inhibitory effect, the interstimulus intervals between the test and condition stimulus were set at 5 ms (Daskalakis et al. 2004; Pinto & Chen 2001; Ugawa et al. 1995). Ten unconditioned pulses were delivered over M1 and 10 paired-pulses were delivered over the cerebellum and M1; the order was randomized in one block.

Experiment Design

There were five experimental blocks including four blocks of CBI at rest and one block of CBI under tonic contraction of FDI. The order of the block of CBI at rest with different CS intensities at 90% through 120% of adjusted RMT was randomized between subjects. The intensity that created the best CBI at rest in the FDI muscle was selected to be the intensity at tonic activation. CBI at tonic activation was performed in the separate block afterward.

Data analysis and statistics

MEP amplitudes of the right FDI, FCR, ECR and ADM were measured peak-to-peak in each trial in millivolts (mV). The mean amplitude of 10 trials of test alone and paired-pulse stimulation were calculated for each session. CBI was calculated by a ratio of mean

conditioned MEPs and unconditioned MEPs. CBI at tonic activation, best CBI at rest and CBI at rest with different cerebellar conditioning stimulus intensities were averaged in each individual muscle. EMG for 95 ms prior to the TMS stimulation was calculated by root mean square amplitude in each trial and individual muscle. CBI at rest with different conditioning stimulus intensities was compared to test the effect of stimulus intensity in the FDI muscle as the target muscle. Best CBI at rest and tonic phase was compared to test the effect of movement conditions (rest vs. tonic) and muscles. One-way ANOVA was performed using one within-subject factor of intensity (90%, 100%, 110%, 120%) to compare CBI with different CS intensities. Repeated measure ANOVA was performed with two within-subject factors; muscle (FDI, FCR, ECR and ADM) and condition (rest and tonic) to test CBI changes in different muscles during rest or tonic contraction of FDI. Background EMG and MEPs amplitude were compared between conditions (rest vs. tonic), different CS intensities and muscles by using a similar statistical analysis.

Statistical analysis method

A mixed model was used to model the repeated measures data with two within-subject factors. The experiments had a three-level structure: condition (or intensity), muscle and subject. The covariance structure was specified as a direct product of an unstructured (UN) matrix for the level of muscle with compound symmetry (CS) for the level of condition (or intensity) along with the subject random effect (UN@CS). Muscle, condition (or intensity) and the interaction between muscle and condition (or intensity) were fixed effects. For the experiments with one within-subject factor, compound symmetry covariance structure was used. Box-Cox procedure was applied to identify an appropriate exponent (Lambda) to transform data: inverse transformation ($\lambda = -1$) for EMG-background, S50 and RMT, natural logarithm ($\lambda = 0$) for MEP-size, and the others with no transformation. Shapiro-Wilk test was performed to evaluate the normality of residuals. Dunnett-Hsu adjustment was used in multiple comparisons with the FDI muscle (or the condition of test, or intensity of 90% adjusted RMT) as a control. The statistical analyses were performed using SAS version 9.2.

Results

A total of 21 participants completed the study of CBI at rest and tonic phase. The mean difference in depth between M1 and the cerebellar lobule VIII was 4.7 ± 3.8 mm (mean $D_{\text{cerebellum}} = 27.9 \pm 6.5$ mm, $D_{\text{M1}} = 23.4 \pm 4.0$ mm). The mean RMT at the motor cortex was 45.8 ± 12.1 % of MSO and the mean adjusted RMT was 59.4 ± 13.9 % of MSO.

CBI at rest with different cerebellar conditioning stimulus intensities

CBI was calculated as a ratio of mean conditioned MEPs and unconditioned MEPs. Increasing CBI ratio was defined as a decreasing inhibitory effect from the cerebellum to the motor cortex or reduction of CBI. 14 participants completed the study of CBI at rest with different intensities. However, the CS intensities at 100% of adjusted RMT of 3 participants reached 80% of maximum stimulus output (MSO) that was set to be the maximum tolerated intensity for cerebellar stimulation in this study (Ugawa, 2009). Thus, these 3 participants did not complete experiment of CBI at rest at CS intensities at 110–120% of adjusted RMT

because the intensities were over 80% of MSO. 1 subject and 3 subjects were unable to tolerate CS intensity at 110% and 120% of adjusted RMT, respectively. Nevertheless, no serious adverse effect was reported after cerebellar stimulation.

There were significant effects for intensity (90%, 100%, 110%, 120% of adjusted RMT, $F(3, 29) = 5.38, p < 0.01$) for CBI at rest in the FDI muscle. *Post hoc* pairwise comparison using CS intensity at 90% of the adjusted RMT as a baseline showed a significant increment of CBI at 110% and 120% compared to 90% of adjusted RMT ($p < 0.01, p < 0.01$). (Figure 2) The mean cerebellar CS intensities of 90%, 100%, 110% and 120% of the adjusted RMT were $54.8 \pm 14.7\%$, $59.9 \pm 16.8\%$, $61 \pm 14.8\%$ and $61 \pm 16.6\%$ of MSO, respectively. MEP sizes with different CS intensities did not differ significantly. ($F(3, 25.1) = 1.32, p = 0.29$)

CBI at pyramidal threshold

There have been some concerns about choosing the suitable intensity for cerebellar stimulation to select just the cerebello-thalamo-cortical pathway and avoid antidromic corticospinal tract activation (Fisher et al. 2009; Ugawa 2009). To determine whether any CS intensities at 90–120% of the adjusted RMT might directly activate the corticospinal tract, pyramidal threshold hunting was performed. We further recorded pyramidal threshold and CBI based on the CS intensity below pyramidal threshold in 12 subjects. CS intensity was set at the closest 5% of the stimulator output below the active pyramidal threshold. The active motor threshold for the pyramidal tract was determined with the double cone coil centered at theinion while subjects made a phasic movement of the FDI. The stimulus intensity was increased in 5% increments and the active motor threshold was determined as the minimum intensity required to elicit MEPs of more than 50uV above the background in 50% of the cases (Daskalakis et al. 2004; Pinto & Chen 2001; Ugawa et al. 1995). The mean pyramidal threshold was $67 \pm 10.3\%$ of MSO (the pyramidal threshold could not be identified in 2 of 12 subjects, 17%). There was no difference between CBI at rest at 110% of adjusted RMT and CBI based on pyramidal threshold in the FDI muscle (unpaired t-test, $p = 0.82$). (Figure 2)

CBI at rest and tonic activation

A further 7 subjects underwent CBI at rest at the CS intensity of 110% of adjusted RMT that seemed to be the optimal intensity to create CBI, and unnecessary repeated cerebellar stimulations could be avoided. Overall, a total of 21 participants completed the study of CBI at rest and tonic phase. The mean CS intensity that created the best CBI at rest was $63 \pm 13.1\%$ of MSO. The baseline CBI at rest was significantly different from 1 in all muscles (FDI; $p < 0.01$, FCR; $p = 0.04$, ECR; $p = 0.01$, ADM; $p < 0.01$). Although CBI at rest was strongest in the FDI muscle, it was not significantly different from other muscles ($F(3, 63) = 0.13, p = 0.94$).

There were significant effects for the interaction between muscle and condition ($F(3, 24.9) = 5.05, p < 0.01$) and condition ($F(1, 17.9) = 9.62, p < 0.01$) but not for the main effect of muscle ($F(3, 25.2) = 0.64, p = 0.59$). *Post hoc* pairwise comparison demonstrated significant CBI reduction in the tonic phase only for the FDI muscle ($p < 0.01$). There were no

significant CBI reductions in the ADM ($p=0.13$), FCR ($p=0.92$), and ECR muscles ($p=0.33$). (Figure 3)

Background EMG at rest and tonic activation

For background EMG, there were significant main effects of condition ($F(1, 21.3) = 20.75$, $p < 0.01$), muscle ($F(3, 25.5) = 50.14$, $p < 0.01$), and interaction between condition and muscle ($F(3, 26.1) = 25.52$, $p < 0.01$). Background EMG in the tonic phase was significantly higher compared to rest only in the FDI as the active muscle ($p < 0.01$). (Figure 4A)

MEP size at rest and tonic activation

MEP amplitudes at test stimulation alone were compared between at rest and tonic activation. The main effects for condition ($F(1, 27.8) = 39.99$, $p < 0.01$) and muscle ($F(3, 28.1) = 27.51$, $p < 0.01$) were significantly different whereas it was not significant for the interaction between condition and muscle ($F(3, 26) = 2$, $p = 0.14$). MEPs sizes were increased in all muscles during tonic activation compared to rest. (Figure 4B) MEP sizes were smaller in proximal muscles of the forearm than the hand muscle.

CBI at S50 matching at tonic phase

Unconditioned MEP sizes at tonic phase were higher than those at rest in all muscles. Since CBI may be affected by MEP sizes (Pinto and Chen 2001), we adjusted the TS intensity using S50 at tonic phase and determined any changes in the level of CBI. We further recorded the CBI at tonic phase with using S50 from IO curve of the FDI muscle during tonic activation of FDI instead of higher S50 from IO curve at rest as the TS intensity in 6 subjects. S50 was selected rather than matched absolute MEP sizes because it is the most sensitive point of changes from IO curve and has less variability between subjects (Pitcher et al. 2015). CS intensity at 110% of the adjusted RMT was selected because it was the lowest intensity that created good CBI. The mean S50 during tonic contraction ($S50_{\text{tonic}}$) was $60 \pm 5.4\%$ of MSO which was decreased from $66 \pm 2.7\%$ of MSO at rest ($S50_{\text{rest}}$). Because the sample size for this additional experiment was small, a non-parametric method was selected for statistical analysis. Although the $S50_{\text{tonic}}$ was lower than $S50_{\text{rest}}$ but unconditioned MEPs sizes did not differ significantly between using $S50_{\text{tonic}}$ and $S50_{\text{rest}}$ as TS intensity (Wilcoxon signed rank test, $p = 0.12$). Moreover, there was no significant difference in CBI in FDI between $S50_{\text{tonic}}$ and $S50_{\text{rest}}$ conditions ($p = 0.06$). CBIs were reduced at both TS intensities compared to rest. (table 1)

S50 and RMT in 4 muscles with stimulation at the FDI hotspot

Since the FDI muscle was selected to be the hotspot for the motor cortex stimulation, S50 and RMT of individual muscle from these stimulations were compared between muscles. S50 and RMT of the FCR, ECR, FDI and ADM were not significantly different ($F(3, 54.2) = 2.45$, $p = 0.07$, $F(3, 54.1) = 2.69$, $p = 0.05$, respectively). (figure 5)

Discussion

Our results demonstrate that CBI was significantly reduced only when the FDI was the target muscle but not for the surround muscles in the hand (ADM) and the forearm (FCR

and ECR) during tonic activation phase compared to rest. This result fits our hypothesis that cerebellar inhibition changes differently in the target and the surround muscles during the tonic phase.

MEP sizes were increased in both target and surround muscles in the tonic phase similar to previous reports (Beck et al. 2008). This indicates that excitability increases during a tonic contraction for the representations of all adjacent muscles within the motor cortex. However, mSI was thought to be absent during the tonic phase because there was no MEP suppression in the surround muscles and spinal excitability was decreased. Moreover, short intracortical inhibition (SICI) was not altered during the tonic phase compared to rest (Beck et al. 2008). It is unknown which modulatory networks play roles in mSI during tonic activation phase. Our data show that the contraction of the FDI was highly selective, based on continuous background EMG monitoring. Hence, there might be some other modulatory networks acting upon the motor cortex to maintain selective finger movement and suppress background EMG in the surround muscles.

We observed CBI at rest in the FDI as the target muscle as well as in the surround muscles. Even though CBI was strongest in the FDI at rest, it was not significantly different from other muscles. Thus, we were able to compare how CBI changed between muscles. We demonstrated that CBI changed differently between the target and surround muscles. Cerebellar and motor cortex interactions might have a role in modulating the surround muscles in the tonic phase. Previous imaging studies suggest that the cerebellum acts like an internal feed-forward system in motor control to predict and detect the error of voluntary actions from sensory feedback in order to optimize movements (Doya 2000; Manto et al. 2012; Wolpert & Flanagan 2001). Visual and proprioceptive feedbacks are important to maintain constant tonic activation. The cerebellum also plays a role in visuo-kinesthetic perception of hand movement by interacting with other parts of the brain (Manto et al. 2012). This might support the idea that the cerebellum might be involved in motor coordination and maintenance of selective finger movement by differently modulating the active and surround muscles. For the active muscle, the cerebellum may reduce inhibition within the motor cortex through the cerebello-thalamo-cortical pathway rendering the motor cortex more excitable and thus less prone to be inhibited by other circuits. On the other hand, this interaction between the cerebellum and motor cortex seems to be different in the surround muscles.

The cerebellum does not seem to be involved in mSI during phasic movement (Kassavetis et al. 2011; Sadnicka et al. 2013), when CBI has been demonstrated to be reduced in both the target and surround muscles (Kassavetis et al. 2011). It was hypothesized that cerebellum might increase motor cortex excitability at the onset of movement by withdrawing inhibition in a non-topographic fashion in both muscles. During movement initiation, the basal ganglia and other cortical areas might have the main role in creating the surrounding inhibition within the motor cortex (Beck et al. 2008, 2009, 2010, 2011; Kassavetis et al. 2011, 2014; Mink 1996; Shin et al. 2009; Sohn & Hallett 2004a, 2004b). During the maintenance phase of movement, our results show that the cerebellum might contribute to the precise cropping of motor cortex excitability. It may do so in a feed-forward manner after integrating multimodal inputs from other parts of the brain (Manto et al. 2012). This fits the hypothesis

from a previous study with transcranial direct current stimulation (tDCS) of the cerebellum that the cerebellum might not only modulate the excitability of the motor cortex but also tune the shape of corticomotor maps with “a focus effect” (Oulad Ben Taib & Manto 2013).

We further performed the CBI experiment with adjusted TS intensity with S50 from the IO curve during tonic activation of the FDI muscle to determine the direction of CBI changes. S50 was selected for TS intensity adjustment rather than matched MEP size because the intensity at S50 from the IO curve is believed to activate neurons at a similar excitability state and produce less variability between subjects than matched absolute MEP (Pitcher et al. 2015). Even though S50 in the tonic phase was slightly lower than S50 at rest in the target muscle, the difference was not statistically different and did not lead to significant changes in unconditioned MEP sizes. However, small changes might not have been detected due to the small sample size in the additional experiment. We adjusted TS intensity only at the FDI muscle because FDI was the active muscle that increased excitability and CBI at the FDI changed differently from other muscles. CBI with adjusted TS intensity at matched S50 confirmed CBI reduction at the FDI muscle.

CBI at rest increased with increasing cerebellar CS intensity. This finding supported results from previous studies (Ugawa et al. 1995; Hardwick et al. 2014). However, we used a different method from the original CBI experiment (Ugawa 1995; Pinto & Chen 2001). CS intensity was based on motor cortex threshold and adjusted for a deeper structure to compensate for the magnetic field attenuation with distance from the coil (Popa et al. 2010; Stokes et al. 2005; Deng et al. 2014). This method is more convenient than the method using CS intensity based on pyramidal threshold for several reasons. Pyramidal threshold is sometimes controversial to define and is often overestimated (Ugawa 2009; Fisher et al. 2009), and may not be observed in all subjects. In our study, we could not observe pyramidal threshold in 2 of 12 subjects (17%). In addition, the pyramidal threshold hunting method by stimulation applied at the inion can sometimes be painful due to facial and neck muscle contraction. We can avoid the process of pyramidal threshold hunting by using CS intensity based on the adjusted RMT of the motor cortex. CS intensity at 90% of adjusted RMT is not enough to create CBI in all subjects as noted in a previous study (Popa et al. 2010). Our study results show that increasing CS intensity to 110% of adjusted RMT seems optimal for CBI. This might be a different method to measure the distance from the cerebellar cortex to the scalp. Although there could be concerns that CS intensity higher than 100% of adjusted RMT might directly stimulate the corticospinal tract, the CS intensity we used, even at 120% of the adjusted RMT, remained below the average intensity used in previous studies based on pyramidal threshold (Pinto & Chen 2001; Fisher et al. 2009). Thus, we are confident that the intensity at 110% of adjusted RMT is strong enough to stimulate the Purkinje cells sufficiently to reduce activity in the dentato-thalamo-cortical pathway without directly stimulating the corticospinal tract. This confirmed the 110% adjusted RMT may be used as the CS intensity for CBI instead of searching for a pyramidal threshold.

Another concern is the selection of the FDI hot spot instead of the ADM or other surround muscles. Since we wanted to select a hot spot able to simultaneously elicit MEPs in all hand and forearm muscles to demonstrate CBI in each muscle, FDI proved to be the easiest spot to find over the homunculus in the motor cortex. The representations of hand muscles (APB

and ADM) largely overlap, even though the ADM is slightly more medially located (Wilson et al. 1993). The representations of hand and arm muscles are also partially overlapped (Metman et al. 1993), allowing us to evoke motor potentials in ECR and FCR with magnetic pulses delivered over the FDI hot spot. Plotting the input-output curves from these stimulations revealed that S50 and RMT did not differ between the muscles. All this allowed us to infer that the FDI hot spot could be the spot-of-choice for our CBI experiment, even if it might remain more representative for the hand muscles rather than the forearm muscles. Despite the MEP amplitudes for FCR and ECR being smaller than those for FDI and ADM, the fact that S50 was the same for all muscles and that cerebellar stimulation is nonspecific for any given muscle makes it reasonable to consider that CBI would have been equally prone to change.

Motor surround inhibition is an important neural mechanism in motor control for selection and maintenance of selective voluntary movement. The interactions between cortical and subcortical structures including the basal ganglia and the cerebellum contribute to this mechanism. At the onset of movement, the inhibitory networks within the cortex and the basal ganglia play important roles for mSI to select a particular movement. During maintenance of the movement, mSI requires other inputs from other parts of the brain such as the cerebellum. Our data might bring some novel insights into the roles of the cerebellar circuits in the pathogenesis of any disorders with impaired motor surround inhibition such as dystonia.

Conclusions

Our data suggest that cerebellum may play a role in maintaining selective tonic movement by modulating CBI differently in the target and the surround muscles. The ability to maintain selective tonic movement might be impaired if inhibitory networks involving cerebellum are disordered.

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Abbreviations

ADM	abductor digiti minimi
CBI	Cerebellar brain inhibition
CS	conditioning stimulation
EMG	Electromyography
ECR	extensor carpi radialis
FDI	first dorsal interosseus
FCR	flexor carpi radialis

MEP	motor evoked potential
MSO	maximum stimulus output
RMT	resting motor cortex threshold
RMS	root mean square
TMS	transcranial magnetic stimulation
TS	test stimulation

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Figure 1. Location of cerebellar lobule VIII

The distance from the scalp to the cerebellar cortex and motor cortex was measured with an MRI based neuronavigation system.

Figure 2. CBI at rest with different CS intensities of the FDI muscles showed a significant increase of CBI at 110% and 120% compared to 90% of adjusted RMT ($p < 0.01$, $p < 0.01$).

Figure 3. CBI at rest and during tonic activation of the FDI muscle of 4 muscles including the FCR, ECR, FDI and ADM

This demonstrates significant CBI reduction in the tonic phase only of the FDI muscle.

($p < 0.01$)

Figure 4. A: Background EMG before TMS stimulation at rest and tonic phase of 4 muscles Background EMG in the tonic phase was increased compared to rest only for the FDI as the active muscle ($p < 0.01$). **B: MEPs amplitudes in rest and tonic phases.** MEP size increased in all muscles.

Figure 5.
S50 and RMT in 4 muscles with stimulation at the FDI hotspot at rest.

Table 1Result of additional experiment with TS intensity at S50_{tonic} and S50_{rest}

	S50 _{rest}	S50 _{tonic}	P value *
CBI at tonic phase	0.96 ± 0.08	0.85 ± 0.17	0.06
TS intensity	66 ± 2.68	60 ± 5.44	0.12
MEP sizes	6.43 ± 3.34	4.8 ± 2.05	0.12

*Wilcoxon signed rank test