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Age-related differences in short- and long-interval intracortical inhibition in a human hand muscle

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Abstract

Background: Effects of age on the assessment of intracortical inhibition with paired-pulse transcranial magnetic stimulation (TMS) have been variable, which may be due to between-study differences in test TMS intensity and test motor evoked potential (MEP) amplitude.

Objective: To investigate age-related differences in short- (SICI) and long-interval intracortical inhibition (LICI) across a range of test TMS intensities and test MEP amplitudes.

Methods: In 22 young and 18 older subjects, SICI and LICI were recorded at a range of test TMS intensities (110% - 150% of motor threshold) while the first dorsal interosseous (FDI) muscle was at rest, or producing a precision grip of the index finger and thumb. Data were subsequently compared according to the amplitude of the MEP produced by the test alone TMS.

Results: When pooled across all test TMS intensities, SICI in resting muscle and LICI in active muscle were similar in young and older adults, whereas SICI in active muscle and LICI in resting muscle were reduced in older adults. Regrouping data based on test MEP amplitude demonstrated similar effects of age for SICI and LICI in resting muscle, whereas more subtle differences between age groups were revealed for SICI and LICI in active muscle.

Conclusions: Advancing age influences GABA-mediated intracortical inhibition, but the outcome is dependent on the experimental conditions. Age-related differences in SICI and LICI were influenced by test TMS intensity and test MEP amplitude, suggesting that these are important considerations when assessing intracortical inhibition in older adults, particularly in an active muscle.

Introduction

The ageing process causes extensive changes to the structure and function of many brain areas, including the primary motor cortex (M1). For example, older adults show decreased thickness of M1 (1), degradation of corticospinal tract white matter (2), and demonstrate increased activation and reduced lateralisation of cortical activity during motor tasks (3, 4). Paired-pulse transcranial magnetic stimulation (TMS) is an increasingly utilised method to further investigate age-related changes in M1 function. When a subthreshold conditioning stimulus precedes a suprathreshold test stimulus by 1–5 ms, there is a reduction of the test motor evoked potential (MEP) amplitude that is likely to involve $GABA_A$-receptors (5), and is referred to as short-interval intracortical inhibition (SICI; 6). However, when both conditioning and test stimuli are suprathreshold and separated by 100–200 ms, there is a reduction of the test MEP amplitude that involves $GABA_B$-receptors (7), and is referred to as long-interval intracortical inhibition (LICI; 8). Several studies have shown that these GABAergic intracortical inhibitory circuits may be affected by advancing age (9-11), although other studies have shown no difference between young and older adults (12-14), even when using a range of conditioning TMS intensities (13, 15). The factors that contribute to these discrepancies between studies are currently unknown.

One important methodological consideration when performing paired-pulse TMS studies is the approach used to obtain the test MEP. In young subjects, the magnitude of inhibition recorded during SICI and LICI depends on test TMS intensity (16) and test MEP amplitude (17). Previous studies examining changes with advancing age have therefore matched one of these variables between age groups (14, 18). Furthermore, recent research suggests that the magnitude of intracortical inhibition in young subjects also depends on the proportion of the test alone MEP relative to the maximum muscle response (M_{max}; 18, 25). These findings suggest that comparisons of intracortical inhibition between young and older subjects, which

typically exhibit different M_{max} characteristics (20), may confound the estimate of SICI and LICI between subject groups. Furthermore, it is not clear whether changes in test TMS intensity or test MEP amplitude (absolute or normalised) have similar effects on the magnitude of SICI and LICI in young and older adults. The aims of the current study were therefore to compare the magnitude of SICI and LICI with increasing test TMS intensity between young and older subjects, and to assess the effects of absolute and normalised test MEP amplitude on age-related comparisons of SICI and LICI.

Materials and Methods

18 older (mean ± SD; 70.8 ± 5.0 years) healthy subjects were recruited to participate in the current study. These data were compared with the data from 22 young (mean ± SD; 22.3 ± 3.1 years) subjects, the results of which have been reported previously (21). Standard exclusion criteria were applied (22) and each subject provided written, informed consent prior to participation. All experimentation was approved by the University of Adelaide Human Research Ethics Committee and conducted in accordance with the declaration of Helsinki.

Experimental arrangement

Subjects were seated in a comfortable chair with their right arm and hand relaxed on a support placed next to them. Surface electromyography (EMG) was recorded from the first dorsal interosseous (FDI) muscle of the right hand using two Ag–Ag Cl surface electrodes in a belly-tendon montage. EMG was amplified (300 X), band-pass filtered (20 Hz-1 kHz) and digitized at 2 kHz (Cambridge Electronic Design data acquisition system, Cambridge, UK), before being recorded and stored offline for analysis.

Force and acceleration were recorded using a manipulandum that has been described previously (23) and is designed specifically for assessing performance during a grip-lift task

involving the thumb and index finger (24). Force signals were amplified (x1000-10,000) and filtered (100 Hz), while both force and acceleration signals were digitised at 400 Hz (Cambridge Electronic Design data acquisition system, Cambridge, UK) and stored offline for analysis.

Experimental Procedures

Maximal Voluntary Contraction. The maximal voluntary contraction (MVC) was assessed while subjects produced maximal precision grip force between the index finger and thumb for 3 s. Several contractions were performed, separated by 30 s rest, until the three greatest trials were within a 10% margin. The largest of these was chosen as the subjects MVC.

Transcranial magnetic stimulation. TMS was applied to left primary motor cortex using a figure-of-eight coil (external wing diameter 9 cms) with two Magstim 200 magnetic stimulators connected through a Bistim unit (Magstim, Dyfed, UK). The coil was held tangentially to the scalp at an angle of 45° to the sagittal plane, with the handle pointed backwards and laterally. The coil was positioned on the scalp over the location producing an optimum response in the relaxed FDI muscle. This location was marked on the scalp for reference and continually checked throughout the experiment. Resting motor threshold (RMT) was defined as the minimum TMS intensity producing a response amplitude $\geq$50 μV in three out of five trials in resting FDI muscle (25). Active motor threshold (AMT) was defined as the minimum TMS intensity producing a response amplitude $\geq$300 μV in three out of five trials (26) while FDI was active in performing a precision grip held at 5% MVC. TMS was delivered at 0.2 Hz for all conditions.

Intracortical inhibition. SICI and LICI were assessed while FDI was relaxed (rest state) or active in performing a precision grip at 5% MVC (active state). SICI used an 80% AMT conditioning stimulus and an interstimulus interval (ISI) of 3 ms (6), while LICI used a 120%

RMT conditioning stimulus and a 150 ms ISI (8). Both paradigms used the same test TMS intensities (110%-150% of motor threshold in 10% increments). At rest, these intensities were normalised to RMT, while during activation they were normalised to AMT. The order in which test TMS intensities were applied was pseudo-randomised between subjects. Both paired-pulse paradigms were applied in the same experimental block, allowing normalisation of conditioned responses to a common test alone state. Using this design, each experimental block contained 20 paired-pulse trials (10 SICI and 10 LICI) and 10 test-alone trials, with each subject receiving a total of 300 stimuli (5 test TMS intensities and 2 activity states).

Grip-lift task. Hand function was assessed during a grip-and-lift procedure, during which subjects held the manipulandum between index finger and thumb using a precision grip, lifted it to a height of approximately 10 cm and then set it down again. No practice was allowed and each subject completed 5 lifts for 3 loads of different mass (100 g, 200 g and 300 g).

Maximal compound muscle action potential (M_{max}) Electrical stimulation applied at the wrist was used to stimulate the ulnar nerve, generating maximal compound muscle action potentials within FDI. Stimuli were applied using a constant-current stimulator (DS7AH, Digitimer, UK) and bipolar surface electrodes with the cathode positioned distally. Each stimulus was a square wave pulse of 100 μs duration and intensity set at 120% of that required to produce a maximal response in FDI (i.e. 120% M_{max}). M_{max} was obtained by averaging the responses to 5 stimuli delivered at the end of the experiment.

Data Analysis

For each test TMS intensity, and in both activity states, SICI and LICI were quantified by expressing individual conditioned MEPs as a percentage of the average unconditioned test alone MEP. When expressing inhibition measurements based on test alone MEP amplitude, data were grouped into 1 mV bins for absolute amplitude and 10% M_{max} bins for normalised

amplitude. Although this alternative analysis resulted in an unequal number of responses from each subject in each bin (due to between-subject differences in test MEP and M_{max} amplitudes), we have previously shown that this sampling procedure produces a similar magnitude of SICI and LICI compared with when each subject contributes a sample to each bin (20).

For grip-lift data, the temporal phases of movement (preload, load, transition) were defined according to previously established criteria (24) using the first derivatives of the grip force (GF), lift force (LF) and acceleration (Acc) traces. The maximum GF (GF_{max} – expressed as a percentage of LF at the time of occurrence), LF (LF_{max}) and Acc (Acc_{max}) were recorded from raw data traces. Cross-correlations between the first derivatives of GF and LF were calculated and assessed via the maximum cross-correlation coefficient (ρ_{max}). The time shift of GF (relative to LF) required to achieve ρ_{max} (lag time) was also assessed.

Statistical Analysis

RMT, AMT and M_{max} amplitude were compared between groups using unpaired student's *t* tests. Individual mixed-model analyses were used to compare the effects of test TMS intensity (110%, 120%, 130%, 140% & 150% RMT/AMT), absolute test MEP amplitude (rest, 0–10, 10–20 and >20 mV; active, 0–10, 10–20, 20–30 and >30 mV) and normalised test MEP amplitude (rest, 0–10%, 10–20% and > 20% M_{max}; active, 0–10%, 10–20%, 20–30% and >30% M_{max}) on SICI and LICI between young and older subjects, in resting and active muscle. Subject was included as a random effect and significant main effects and interactions were further investigated using Bonferroni corrected custom contrasts. Repeated-measures analysis of variance ($ANOVA_{RM}$) was used to assess the impact of lift trial (1, 2, 3, 4, 5), weight (100g, 200g, 300g) and age (young, older) on grip-lift performance. Significant main effects and interactions were further investigated using unpaired student's *t* tests with Bonferroni correction. For all significant between-group interaction effects, the estimated

mean difference and corresponding 95% confidence interval (CI) was also calculated as an unstandardised indication of effect size. Linear regression of individual subject data was used to investigate interactions between measures of corticospinal excitability and grip-lift performance indices. Significance was set at $P < 0.05$ for all comparisons and data are shown as mean and 95% CI [lower limit, upper limit], unless otherwise stated.

Results

As the results of the young cohort have been previously reported (21), and the primary interest of the current study concerns age-related effects, only findings involving main effects or interactions of age will be described in detail. No differences were found between groups for RMT (young, 46.4 [43.3, 49.4] % MSO; older, 48.2 [43.2, 53.1] % MSO, $P = 0.5$) or AMT (young, 38.2 [35.2, 41.2] % MSO; older, 38.4 [34.3, 42.5] % MSO, $P = 0.9$), but M_{max} amplitude was 40% larger in young subjects (19.0 [17.4, 20.6] mV) than in older subjects (13.5 [12.0, 15.0] mV; $P < 0.0001$).

Influence of test TMS intensity on the test alone MEP in young and old subjects

Increasing test TMS intensity resulted in larger absolute test MEP amplitudes in both resting ($P < 0.0001$, Fig. 1A) and active ($P < 0.0001$, Fig. 1C) muscle. At rest, the amplitude of the absolute test MEPs at each test TMS intensity were unaffected by age ($P = 0.1$) and there was no interaction between age and test TMS intensity ($P = 0.2$). With FDI active, absolute test MEP amplitude was reduced in older subjects ($P = 0.0001$) and there was an interaction between age and test TMS intensity ($P < 0.0001$). Post hoc testing showed that young subjects had larger absolute test MEP amplitudes than older subjects at 130% (estimated mean difference: 0.8 mV, 95% CI [0.3, 1.2], $P = 0.001$), 140% (estimated mean difference: 1.1 mV, 95% CI [0.7, 1.6], $P < 0.0001$) and 150% RMT (estimated mean difference: 1.7 mV, 95% CI [1.3, 2.2], $P < 0.0001$).

After normalising absolute amplitude test MEPs to individual subject M_{max}, increasing test TMS intensity produced larger amplitude normalised test alone MEPs in both resting (P < 0.0001, Fig. 1B) and active (P < 0.0001, Fig. 1D) muscle. At rest, no effect of age was found (P = 0.2), but there was an interaction between age and test TMS intensity (P < 0.0001). Between-group post hoc analysis showed larger normalised test MEP amplitudes in older subjects when applying the 150% RMT test intensity (estimated mean difference: 7.3 %M_{max}, 95% CI [1.3, 13.4], P = 0.02). In active muscle, normalised test MEP amplitude was not different between age-groups (P = 0.4) and there was no interaction between age and test TMS intensity (P = 0.4).

Influence of age on SICI and LICI

The main effects of age on SICI and LICI in resting and active muscle pooled across all test TMS intensities are shown in Fig. 2. In resting muscle, SICI was not different between groups (P = 0.3, Fig. 2A) whereas LICI was significantly reduced in older subjects (P = 0.007, Fig. 2C). With the muscle active, SICI was significantly reduced in older subjects (P = 0.02, Fig. 2B) but LICI was unaffected by age (P = 0.3, Fig. 2D).

Influence of test TMS intensity and test MEP amplitude on age-related changes in SICI

For SICI at rest, increasing test TMS intensity resulted in reduced inhibition (P < 0.0001, Fig. 3A), but there was no interaction between age and test TMS intensity (P = 0.5). In active muscle, greater test TMS intensity produced increased inhibition (P < 0.0001, Fig. 3D). However, interactions between age and test TMS intensity were not significant (P = 0.7).

When SICI data were regrouped according to absolute test MEP amplitude, resting SICI was reduced in response to larger amplitude test alone MEPs (P = 0.003, Fig. 3B) but this was not different between age groups (P = 0.2). However, SICI in active muscle was increased with larger absolute amplitude test alone MEPs (P < 0.0001, Fig. 3E) and there was a significant

interaction between age and absolute test MEP amplitude ($P < 0.0001$). Age-related comparisons within each bin showed that older subjects had increased SICI for test MEP amplitudes of 0–1 mV (estimated mean difference: 17.9%, 95% CI [4.9, 30.8], $P = 0.007$) and 2–3 mV (estimated mean difference: 10.9%, 95% CI [1.2, 20.6], $P = 0.03$), but reduced SICI for test MEP amplitudes >4 mV (estimated mean difference: 16.4%, 95% CI [11.1, 21.8], $P < 0.0001$).

When SICI data were grouped according to normalised test MEP amplitude, measurements in resting muscle were unaffected by changes in amplitude ($P = 0.09$, Fig. 3C), but a significant interaction between age and normalised amplitude was found ($P = 0.001$). However, age-related comparisons within individual bins were not significant. In active muscle, a main effect of normalised MEP amplitude was found ($P < 0.0001$) and there was an interaction between age and normalised test MEP amplitude ($P < 0.0001$, Fig. 3F). Age-related comparisons within each bin showed that older subjects had increased SICI for normalised MEP amplitudes of 0–10% M_{max} (estimated mean difference: 13.7%, 95% CI [4.0, 23.3], $P = 0.005$), but reduced SICI for normalised MEP of 10–20% (estimated mean difference: 8.8%, 95% CI [1.6, 16.0], $P = 0.02$) and >30% M_{max} (estimated mean difference: 18.6%, 95% CI [13.0, 24.2], $P < 0.0001$).

Influence of test TMS intensity and test MEP amplitude on age-related changes in LICI

Increasing test TMS intensity reduced the magnitude of LICI in resting muscle ($P < 0.0001$, Fig. 4A) and there was a significant interaction between age and test TMS intensity ($P = 0.004$). Age-related comparisons within each test intensity showed that older subjects had less inhibition than young subjects at 110% RMT (estimated mean difference: 30.0%, 95% CI [6.6, 53.4], $P = 0.01$), 120% RMT (estimated mean difference: 28.7%, 95% CI [5.3, 52.2], $P = 0.02$), 130% RMT (estimated mean difference: 39.6%, 95% CI [16.2, 63.1], $P = 0.001$) and

140% RMT (estimated mean difference: 40.4%, 95% CI [17.0, 63.9], $P = 0.01$). In active muscle, increasing test TMS intensity produced increased LICI ($P < 0.0001$, Fig. 4D) and a significant interaction between age and test TMS intensity was found ($P < 0.0001$), with age-related comparisons within individual test intensities showing that older subjects had more LICI than young subjects at 110% RMT (estimated mean difference: 46.2%, 95% CI [5.9, 86.5], $P = 0.03$).

When LICI data were regrouped according to absolute test MEP amplitude, measurements in resting muscle were reduced in response to larger absolute amplitudes ($P < 0.0001$) and there was an interaction between age and absolute test MEP amplitude ($P = 0.02$, Fig. 4B). Age-related comparisons within each bin showed that older subjects had less inhibition than young subjects for absolute amplitudes that were 0–1 mV (estimated mean difference: 43.0%, 95% CI [17.5, 68.6], $P = 0.001$) and 1–2 mV (estimated mean difference: 34.1%, 95% CI [8.1, 60.1], $P = 0.01$). In active muscle, LICI was increased when assessed using larger absolute test MEP amplitudes ($P < 0.0001$) and there was an interaction between age and absolute test MEP amplitude ($P < 0.0001$, Fig. 4E). Post hoc comparisons showed that older subjects had significantly more LICI than young subjects for absolute test MEP amplitudes of 0–1 mV (estimated mean difference: 52.4%, 95% CI [10.9, 94.0], $P = 0.01$) and 1–2 mV (estimated mean difference: 41.7%, 95% CI [1.1, 82.4], $P = 0.04$).

When LICI data were regrouped according to normalised test MEP amplitude, measurements in resting muscle were reduced in response to larger normalised test MEPs ($P < 0.0001$) but there was no interaction between factors ($P = 0.4$). Age-related comparisons within individual bins showed that LICI was significantly reduced in older subjects for test MEP amplitudes that were 0–10% (estimated mean difference: 29.8%, 95% CI [4.6, 55.0], $P = 0.02$) and 10–20% M_{max} (estimated mean difference: 35.3%, 95% CI [8.6, 61.9], $P = 0.01$). In active muscle, LICI was increased in response to larger normalised test MEP amplitudes ($P <$

0.0001) and there was a significant interaction between age and normalised amplitude ($P <$ 0.0001, Fig. 4F). Between-group comparisons showed a trend towards increased inhibition in older subjects for test MEP amplitudes that were 0–10% M_{max} (estimated mean difference: 40.1%, 95% CI [0.3, 80.6], $P = 0.05$).

Grip-lift performance and linear regression

Grip-lift data were obtained from 10/22 young subjects (mean age ± SD; 22.1 ± 1.2 years) and all 18 older subjects. Age-related comparisons of performance parameters are shown in Table 1. Linear regression analysis found significant associations between M_{max} amplitude and the duration of both preload ($r^2 = 0.46$, $P = 0.0001$) and transition ($r^2 = 0.18$, $P = 0.03$) phases and significant association were also found between the magnitude of LICI in resting muscle and the duration of the transition phase ($r^2 = 0.04$, $P = 0.02$). All other comparisons were not significant.

Discussion

The current study examined age-related differences in SICI and LICI in resting and active muscle with increasing test TMS intensity. This approach produced a broad range of MEP responses, allowing an investigation of age-related differences in inhibition at different absolute and normalised (relative to M_{max}) test MEP amplitudes. When data were pooled across test TMS intensities, SICI in active muscle and LICI in resting muscle were reduced in older compared with young subjects, but there were no age-related differences in SICI at rest and LICI in active muscle. However, these effects varied depending on the approach used to compare the test response (test TMS intensity/test MEP amplitude) between groups, suggesting that this is an important consideration when assessing age-related differences in SICI and LICI.

Advancing age influences SICI in active but not resting muscle

Within the current study, SICI in resting muscle was not affected by age, suggesting maintenance of resting $GABA_A$-mediated intracortical inhibition in old adults (5). This finding supports the results of several previous studies (12-15, 20, 27-29) but is in contrast to others (9-11, 18, 30). Although the reasons for these discrepancies are unclear, they are commonly attributed to methodological differences between studies. For example, previous studies have matched either test TMS intensity or test MEP amplitude between groups, as both factors are thought to influence estimates of SICI in resting muscle (16, 17). We investigated whether estimates of SICI at rest, assessed using several different approaches to compare the test alone MEP, were differentially effected by age. We found that resting SICI did not differ between age groups when data were matched for test TMS intensity or test MEP amplitude (absolute or normalized), suggesting that these factors are unlikely to contribute to previous inconsistencies between studies. Furthermore, as conflicting effects of age on SICI have been reported from studies that have used the same conditioning intensity and ISI (13, 18, 31), it seems unlikely that variations in stimulus parameters alone can account for these inconsistencies. We therefore suggest that factors other than TMS parameters may contribute to inter-study variations in effects of age on resting SICI, such as target muscle, or subject characteristics, such as health status, physical activity levels, or habitual hand function.

In contrast with resting muscle, SICI in active FDI was significantly reduced in older subjects, but the effect varied depending on the approach used to compare the test MEP between groups. When data were pooled over all test TMS intensities, SICI in active muscle was less in older adults. However, regrouping data relative to test MEP amplitude (absolute or normalised) showed that older adults had increased SICI for small amplitude test MEPs (0-1 mV/0-10% M_{max}) but decreased SICI for large amplitude test MEPs (>4 mV/>30% M_{max}).

Only one previous study has examined age-related changes in SICI during muscle activation (although age-effects during movement preparation have been investigated; 10, 28, 29). Using a 2 mV test MEP and low-intensity (15% MVC) contraction of the flexor carpi radialis muscle (FCR), McGinley and colleagues failed to observe any effect of age on SICI in active muscle (11). The results of the current study support this, as there was no difference in active SICI between groups when matching absolute test MEP amplitude at 1-2 mV (Fig. 2E). However, differences were observed when smaller or larger test MEPs were used, suggesting that test MEP amplitude has important implications for the comparison of active SICI between young and older adults. Furthermore, the effects of age on active SICI were variable and dependent on the specific test MEP amplitude used, suggesting that more than one test MEP amplitude should be used to adequately characterise age-related differences in SICI in active muscle.

Advancing age influences LICI in resting and active muscle

The most striking difference between young and older adults in the present study was a reduction in resting LICI in older adults. This effect was observed irrespective of how the data were grouped, suggesting strong age-related alterations in resting $GABA_B$ mediated intracortical inhibition (7). This effect was most pronounced at low-moderate test TMS intensities (<140% RMT) and test MEP amplitudes (<2 mV/<20% M_{max}). In contrast to our findings, the only previous study to examine age-related differences in resting LICI showed an *increase* in LICI in older adults (11). We suspect that the use of different ISI's within each study may have contributed to these divergent findings. Our study used an ISI of 150 ms rather than the 100 ms interval used previously (11), because recent research suggests that inhibition observed using the shorter ISI (100 ms) may be influenced by changes in spinal excitability (32). These possible timing-dependent effects of age on LICI may be consistent with recent suggestions that measurements of LICI using a 100 or 150 ms ISI do not

represent activation of the same cortical process (33-35). Nonetheless, for low-moderate test TMS intensities and test MEP amplitudes that are commonly used experimentally, our data show a relatively consistent reduction in resting LICI in older adults.

Our assessment of LICI in active FDI muscle did not find any main effect of age. However, separating data based on test TMS intensity or test MEP amplitude revealed increased LICI in older subjects at low test TMS intensities (110% AMT) and absolute test MEP amplitudes (< 2 mV), with no difference between age groups for normalised test MEP amplitude. In the only other study to examine age-related changes in LICI in an active muscle, no effect of age was found when data in FCR muscle was matched between groups using a 2 mV test MEP amplitude (11). As suggested above, it is possible that the use of different ISI's may have contributed to these contradictory findings. However, the contraction intensity also varied between studies (15% MVC by the previous study, 5% MVC in the current study) and, as increasing contraction intensity has been shown to have non-linear effects on the magnitude of active LICI (32), age-related variations in this effect may also have contributed to the contrasting results.

Interestingly, when low test TMS intensities and test MEP amplitudes were used to compare LICI between groups, young subjects demonstrated MEP facilitation, whereas older subjects displayed MEP inhibition. A previous study in young subjects assessing LICI in active muscle also reported a tendency for MEP facilitation when using low intensity test stimuli (110% RMT) and ISIs of 150–160 ms (36). Furthermore, two recent studies have observed a period of cortical disinhibition at long intervals (>165 ms) after application of suprathreshold TMS (37, 38), which was suggested to relate to the previously observed MEP facilitation (37). Although our differential effect of test TMS intensity on LICI in active muscle may therefore suggest an age-related reduction in this cortical disinhibition, the lack of any strong

correlation of LICI with grip-lift performance suggests that the functional implications, at least during this task, are relatively minor.

In the present study, it was not possible to directly compare the magnitude of intracortical inhibition in resting and active states because the test TMS intensities were not the same under both conditions, due to normalisation to either the resting (in the rest state) or the active motor threshold (in the active state). Despite this caveat, the magnitude of LICI in older adults over a range of test TMS intensities was less than young adults at rest, but the effect was removed (or even reversed at some TMS intensities) in the active muscle. These results suggest that the activity-related modulation of LICI may be reduced in older adults, which supports recently reported observations for SICI (10, 28). The activity-related disinhibition of SICI has been suggested to stem from an increased contribution of I_1 waves to the MEP (39, 40), as well as a reduced inhibition of I_3 waves from SICI circuits (40). Given that SICI and LICI both modulate the amplitude of late I waves (41, 42), age-related differences in the activity-dependent modulation of LICI may therefore reflect differences in the ability to modulate these intracortical inhibitory circuits, or age-related differences in the way these inhibitory circuits influence the descending volley during muscle activation. Nonetheless, the mechanisms contributing to the disinhibition of LICI during muscle activation, along with the functional implications of age-related changes in these mechanisms, remain to be explored.

In conclusion, we found age-dependent differences in the magnitude of SICI and LICI, suggesting alterations to $GABA_A$- and $GABA_B$-mediated intracortical inhibition. However, the nature of these effects depended on the activity state of the target muscle, the technique used to compare the test response between groups (test TMS intensity/test MEP amplitude) and the specific characteristics of the test response. Our findings suggest that future studies investigating age-related changes in SICI and LICI during muscle activation should consider

the use of multiple test TMS intensities or test MEP amplitudes when quantifying the magnitude of intracortical inhibition.

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Conflict of Interest

The authors declare no conflict of interest.

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Figure 1. Changes in the amplitude of the test alone MEP with increasing test TMS intensity. Data show the average amplitude of the absolute (mV; A, C) and normalised (% M_{max}; B, D) test MEP amplitude recorded at each test TMS intensity for young (black circles) and older (white circles) subjects in resting (top panels) and active (bottom panels) FDI muscle. Positively directed error bars show the upper limit of the 95% CI, negatively directed error bars show the lower limit of the 95% CI. *P < 0.05. Abbreviations: mV, millivolts; RMT, resting motor threshold; AMT, active motor threshold; M_{max}, maximum compound muscle action potential.

Figure 2. Main effects of age on SICI (A, B) and LICI (C, D) in resting (left panels) and active (right panels) muscle when data are pooled across individual test TMS intensities. Black columns show the response of young subjects, white columns show the response of old subjects. The dotted horizontal line represents no inhibition, with values below 100% representing inhibition of the test MEP. Error bars show the upper limit of the 95% CI. *P < 0.05. Abbreviations: MEP, motor evoked potential

Figure 3. Effects of test TMS intensity (A, D), absolute test MEP amplitude (B, E) and normalised test MEP amplitude (C, F) on SICI in young (black circles) and old (white circles) adults at rest (top panels) and during activation (bottom panels) of FDI. The dotted horizontal line represents no inhibition, with values below 100% representing inhibition of the test MEP. Positively directed error bars show the upper limit of the 95% CI, negatively directed error bars show the lower limit of the 95% CI. *P < 0.05. Abbreviations: MEP, motor evoked

potential; RMT, resting motor threshold; AMT, active motor threshold; M_{max}, maximum compound muscle action potential.

Figure 4. Effects of test TMS intensity (A, D), absolute test MEP amplitude (B, E) and normalised test MEP amplitude (C, F) on LICI in young (black circles) and old (white circles) adults at rest (top panels) and during activation (bottom panels) of FDI. The dotted horizontal line represents no inhibition, with values below 100% representing inhibition of the test MEP. Positively directed error bars show the upper limit of the 95% CI, negatively directed error bars show the lower limit of the 95% CI. *P < 0.05. Abbreviations: MEP, motor evoked potential; RMT, resting motor threshold; AMT, active motor threshold; M_{max}, maximum compound muscle action potential.

Table 1. **Grip-lift performance parameters for young and old subjects**

	Young			**Old**		
	100	200	300	100	200	300
Finger force / coordination						
GF_{max} (% LF)	268.7 (194.4, 343.0)	228.1 (184.9, 271.3)	229.4 (179.6, 279.2)	363.2 (302.5, 423.8)	296.9 (257.8, 335.9)	279.6 (255.6, 303.7)
LF_{max} (N)	2.6 (2.5, 2.7)	3.8 (3.7, 3.9)	5.0 (4.8, 5.2)	2.6 (2.5, 2.7)	3.7 (3.6, 3.8)	4.8 (4.7, 4.9)
Acc_{max} (g)	1.8 (1.4, 2.2)	1.7 (1.3, 2.1)	1.5 (1.1, 2.0)	1.5 (1.1, 2.0)	1.5 (1.1, 1.8)	1.3 (1.0, 1.6)
ρ_{max}	0.84 (0.81, 0.86)	0.86 (0.85, 0.88)	0.87 (0.84, 0.89)	0.79 (0.76, 0.82)	0.83 (0.81, 0.85)	0.83 (0.80, 0.86)
Phase Duration (ms)						
Preload	49 (38, 60)	52 (39, 64)	52 (85, 135)	135 (99, 172)*	115 (86, 144)*	110 (85, 135)*
Load	145 (111, 179)	168 (124, 211)	198 (139, 256)	226 (166, 287)	287 (202, 372)	305 (218, 393)
Transition	1107 (937, 1277)	1097 (935, 1258)	1285 (1058, 1511)	1680 (1400, 1961)*	1699 (1441, 1956)*	1668 (1383, 1952)

Values are shown as mean (95% CI; lower limit, upper limit). * = $P < 0.05$ when compared to the same weight in young subjects. Abbreviations: ms, milliseconds