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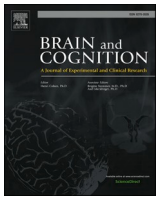

Corticospinal suppression in response to pics with implied hand actions: A follow up TMS study

Riccardo Dalla Volta [a,b,*], Francesco Scarfone [c], Dario Brambilla [a], Roberto Esposti [a], Paolo Cavallari [a,b]

[a] Sezione di Fisiologia, Dipartimento di Fisiopatologia Medico-Chirurgica e dei Trapianti, Università degli Studi di Milano, Via Mangiagalli 32, 20133 Milano, Italy
[b] Laboratorio Sperimentale di Fisiopatologia Neuromotoria, IRCCS Istituto Auxologico Italiano, Meda, Italy
[c] Dipartimento di Scienze Mediche e Chirurgiche, Università Magna Graecia di Catanzaro, Viale Europa, Loc. Germaneto, 88100 Catanzaro, Italy

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ABSTRACT

Presentation of bodily actions is known to affect motor system activity in perceivers' brain. A previous study (Gianelli, Kuehne, Lo Presti, Mencaraglia & Dalla Volta, 2020) employing hand-tool interaction with apparent motion showed early suppression of corticospinal excitability in hand muscles. To control for the role of apparent motion and to investigate the suppression duration, in the present follow up study participants observed pics displaying hand-tool actions, with no apparent motion but only implied motion. Single pulse TMS was delivered on the hand sector of the left motor cortex at 1 s after fixation cross (baseline), at 150, 350, 500 and 700 ms from stimulus onset, while motor evoked potentials (MEPs) were recorded from the contralateral first dorsal interosseus muscle. Results showed a difference in MEP amplitude between hand action-related and control pics where hand action observation suppressed corticospinal excitability, suggesting early and enduring motor inhibition. In addition, MEP amplitude decreased over time. These findings rule out a necessary role of apparent motion, indicating that the simple presentation of hand actions with implied motion effectively induced motor inhibition. Corticospinal suppression may act to prevent the motor system from automatically transforming observed actions into overt movements whenever an action is observed.

1. Introduction

The ability of the brain motor system to react to perceived movements in the absence of varied electromyographic (EMG) activity is known as motor resonance. The neural underpinnings of this remarkable phenomenon are considered the mirror neurons in monkeys and the mirror neuron system (MNS) in humans (Rizzolatti & Craighero, 2004). Mirror neurons are a class of visuomotor neurons originally recorded in the ventral premotor cortex that are modulated both when a bodily action is performed and when the same action is just observed in another individual. Evidence for a system equivalent to monkey mirror neurons has accumulated also in humans by means of non-invasive techniques such as metabolic imaging, transcranial magnetic stimulation (TMS) and EEG/MEG (for review see Rizzolatti, Cattaneo, Fabbri-Destro, & Rozzi, 2014). From a functional perspective, motor resonance, by mapping perceived actions onto one's own motor repertoire, is considered to support cognitive functions like action understanding, prediction of motor intentions and imitative behavior (for review see Keysers, Paracampo, & Gazzola, 2018).

TMS has proved to be a fundamental tool for investigating motor resonance. Alone (e.g. (Fadiga, Fogassi, Pavesi, & Rizzolatti, 1995; Strafella & Paus, 2000) or in combination with other techniques such as H-Reflex testing (e.g. Baldissera, Cavallari, Craighero, & Fadiga, 2001), it can probe corticospinal excitability changes in response to action presentation. For instance, TMS-evoked motor potentials (MEPs) recorded from muscles involved in the execution of the observed actions show a modulation in amplitude. In addition, TMS is endowed with a high temporal resolution that allows researchers to reveal the chronometry of excitability changes (e.g. Gangitano, Mottaghy, & Pascual-Leone, 2001; 2004).

The majority of TMS studies reports that observing bodily actions facilitates the motor system of the observer, as indicated by an increase of MEP amplitude with respect to baseline or control conditions (e.g. Fadiga et al., 1995; for review see Naish, Houston-Price, Bremner & Holmes, 2014). Among the stimuli generally showing facilitatory effects are live actions performed in front of the observer, videoclips showing

* Corresponding author.
E-mail address: riccardo.dallavolta@unimi.it (R. Dalla Volta).

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the unfolding in time of actions or even still images in which the unfolding of an action is only implied. However, there are also reports of a modulation in the opposite direction. Indeed, decreases of MEPs amplitude with respect to a baseline (e.g. Gianelli, Kuehne, Lo Presti, Mencaraglia, & Dalla Volta, 2020; Sartori, Xompero, Bucchioni, & Castiello, 2012) or a control condition (Hardwick, McAllister, Holmes, & Edwards, 2012; Lago, & Fernandez-del-Olmo, 2011; Villiger, Chandrasekharan, & Welsh 2011) and even no net modulation (e.g. Hannah, Rocchi, & Rothwell, 2018) were found. Consequently, it has to be concluded that the motor resonance phenomenon is not a straightforward increase in motor excitability when an action is viewed. At first sight, the coexistence of corticospinal facilitation and inhibition in response to action observation may appear difficult to reconcile and challenges the comprehension of motor resonance. Generally, it has been suggested that motor inhibition acts to prevent the execution of movements during action observation (Keysers & Gazzola, 2010; van Leeuwen, van Baaren, Martin, Dijksterhuis, & Bekkering, 2009). In this context, inhibition represents the other side of the coin to prevent motor resonance from implementing movements of the body.

In a recent paper (Gianelli et al. 2020) comparing the time course of corticospinal excitability in response to the presentation of manual actions involving tools, either by means of pictures or by means of sentences, we showed that MEPs were suppressed as compared to baseline and control conditions in which a fixation cross or the tool alone were presented, respectively. In that study, stimulus presentation consisted of a combination of two still images, the first with a tool alone, and the second one with a hand holding that tool as if using it. The rapid succession of the two images gave the observer the sensation of a brisk action of the hand with the tool (for a similar presentation technique, see Cavallo, Heyes, Becchio, Bird, & Catmur, 2014; Ortigue, Sinigaglia, Rizzolatti, & Grafton, 2010). This presentation strategy combines the advantage of using movies (i.e. inducing the perception of motion) with that of using a still image of an action (i.e. the motor content is available all at once). In fact, using still images prevents the possibility to predict in advance the upcoming action. In turn, this is important when studying the time course of motor resonance to avoid precocious activations of the motor system due to prior knowledge (Kilner, Vargas, Duval, Blakemore, & Sirigu, 2004). However, in that previous study, it was not clear whether MEP suppression stems from just processing apparent motion per se or rather from processing the motor content of the second stimulus. Clarifying this point is not trivial because apparent motion may not be considered a fully naturalistic stimulus and this in turn may question the ecological validity of the results.

In addition, inhibition started as early as 150 ms and lasted at least 500 ms, that coincided with the duration of the stimulus displaying hand action. Consequently, it remains to be established if MEP suppression continues beyond the last investigated timing (500 ms) or instead a return toward baseline level occurs. In this regard, it is worth noting that, in humans, EEG mu rhythm is considered a marker of the mirror neuron system (Altschuler, Vankov, Wang, Ramachandran, & Pineda, 1997). Mu rhythm dynamics show the occurrence of event-related synchronizations in the alpha and beta bands about 500–700 ms after hand action observation which, in turn, suggest a recovery from activation in sensorimotor cortex (Avanzini et al., 2012; Caetano, Jousmäki, & Hari, 2007; Muthukumaraswamy & Johnson, 2004). It is then possible that this recovery from activation reduces the need for corticospinal inhibition to prevent overt movements. This hypothesis may be verified by testing corticospinal excitability at a timing compatible with mu rhythm synchronization after action observation.

Given the above premises, the objective of the present study is twofold. First, we addressed whether the inhibitory effects observed in the original paper of Gianelli and colleagues (2020) may represent generic apparent motion effects resulting from the combination of two pics or, rather, result from the observation of hand actions in which motion is not showed but only implied. To this aim, we presented to participants only the original pics displaying a hand holding different tools as if using them (second pic of the Gianelli et al.'s combination). In this way, if these pics were sufficient to elicit a MEP suppression comparable to that of the original study, it would be clear that actions with implied motion have a causative role in inducing inhibitory effects and that inhibition does not necessarily depend on perception of apparent motion. The second objective was to evaluate whether the duration of corticospinal inhibition correlates with the dynamics of mu rhythm synchronization after the observation of bodily movements. To this aim, we lengthened the investigated temporal window by adding a further timing for TMS stimulation, covering up to 700 ms after stimulus onset. We expected to replicate the MEP suppression of the original study up to 500 ms and to observe a partial or complete recovery from suppression in the last part of selected time window (that is, at 700 ms). As a control, a group of participants observed stimuli with no motor content and, in this case, we did not expect any modulation of corticospinal excitability.

2. Methods

2.1. Participants

We enrolled 20 subjects (11 males) in the Experimental group and 15 others (9 males) in the Control group of this TMS study, which started at the Magna Graecia University of Catanzaro and was then completed at the University of Milan. They had normal or corrected-to-normal vision and no history of neurological or psychiatric disorder. The mean age of the volunteers was 21 years (range: 19–29) for the Experimental group and 23 years (range: 20–34) for the Control group. All of them were right-handed, as ascertained by the Edinburgh Handedness Inventory (Oldfield, 1971). The research was conducted according to the principles expressed in the Declaration of Helsinki and it was approved by both the Comitato Etico Regione Calabria and the Comitato Etico di Ateneo dell'Università degli Studi di Milano, Italy. Before the experiment, participants gave their written informed consent.

The minimum required sample size (N = 17) of the Experimental group was determined by a power analysis using the following parameters: alpha = 0.05, power = 0.85, effect size = 0.8 (Cohen's d). The effect size was derived from results of the original study with similar stimuli and design (Gianelli et al. 2020). Data collection slightly exceeded calculated sample size allowing to account for data loss due to technical reasons. Experimenters were blind to the results till the end of data collection.

2.2. Stimuli

As Experimental stimuli, we employed a subset of stimuli used in a previous study (Gianelli et al. 2020). Briefly, they consisted of 12 pictures showing a right hand interacting with common tools (see Fig. 1). The hand was postured in the appropriate fashion required to effectively make use of the tools. Pictures' resolution was 440 x 440 pixels, with the tool presented roughly at the center of the image and a right hand coming from the right side.

As Control stimuli, we used a set of scrambled images. These were obtained by manipulating the original experimental images with the G'MIC plug-in (Tschumperlé, Fourey, & Osgood, 2025) running in GIMP 2.10 (https://www.gimp.org). Each experimental image was first sliced into 400 equal tiles according to a 20x20 grid. Subsequently, tiles' order was randomly shuffled so that resulting images maintained a certain degree of visual complexity (e.g. colors, contours, brightness) but the original stimuli were no longer recognizable (see Fig. 1). All stimuli are available at reasonable request.

2.3. Procedure

Participants sat on a comfortable chair in front of a 19 in. PC screen (1920 × 1080 pixel resolution and 60 Hz refresh rate). The eye-to-screen distance was about 90 cm. They were requested to maintain the right

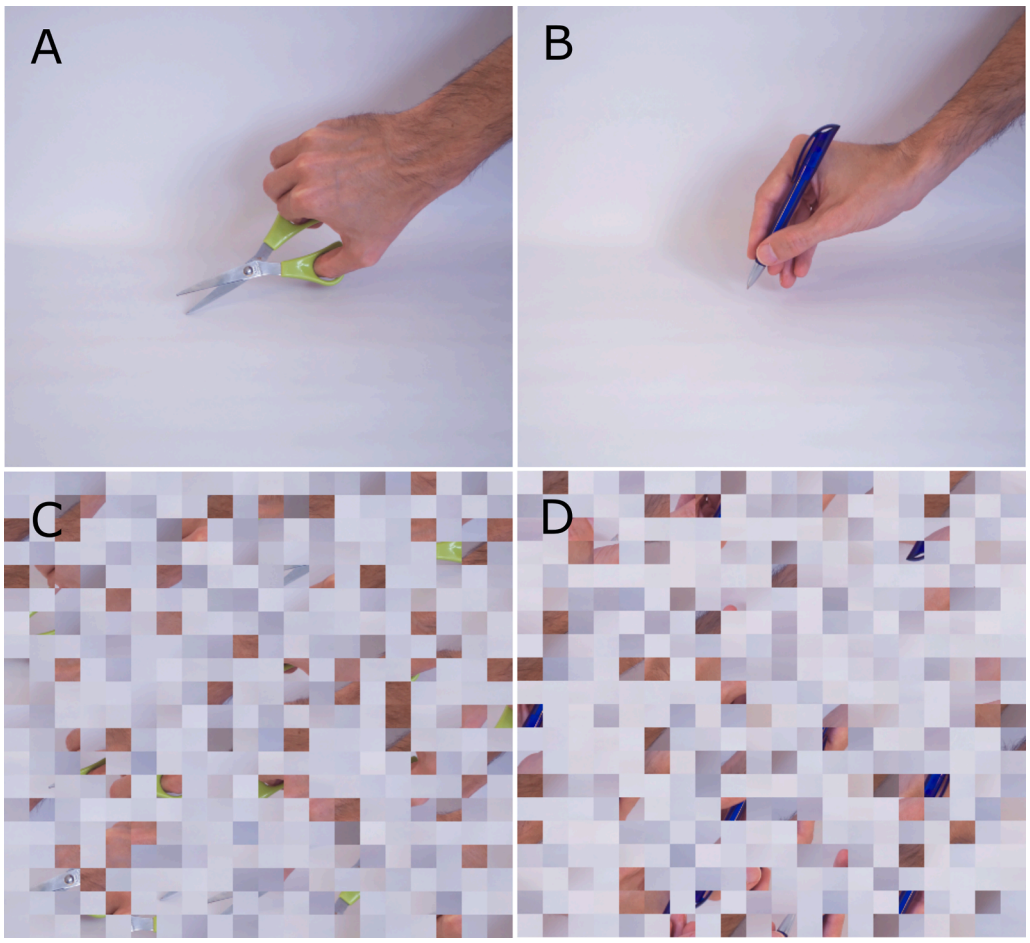

Fig. 1. Stimuli. Stimuli presented to participants. A, B: examples of static pics showing a right hand in the act of using common tools (implied actions). C, D: corresponding scrambled versions.

arm flexed at the elbow (90°) on a pillow and relax while carefully attending to the presented stimuli. To maintain participant's attention, in 10 % of the trials a written question randomly appeared right after the experimental stimuli disappeared. Questions were related to basic features of the stimuli (e.g., "Does the presented image contain the hue blue?") and participants responded vocally. Each trial started with a white fixation cross displayed at the center of a black background (duration: 2 s). Subsequently, a picture was presented (1 s), followed by a black screen serving as intertrial (duration 6 s). Each experiment consisted of one session of 60 trials. For each trial, a single TMS pulse was given at one of five possible timings: one Baseline, at 1000 ms after the onset of the fixation cross, and four experimental timings at 150 ms (T1), 350 ms (T2), 500 ms (T3) and 700 ms (T4) after the onset of the picture, respectively (see Fig. 2). The software Eprime 3.0 (Psychology Software Tools, Inc.) controlled stimulus presentation and triggering of the TMS. In conclusion, the experimental design consisted of 2 groups (Experimental group observing hand actions vs. Control group observing scrambled pics) X 5 Timings (Baseline, T1, T2, T3, T4).

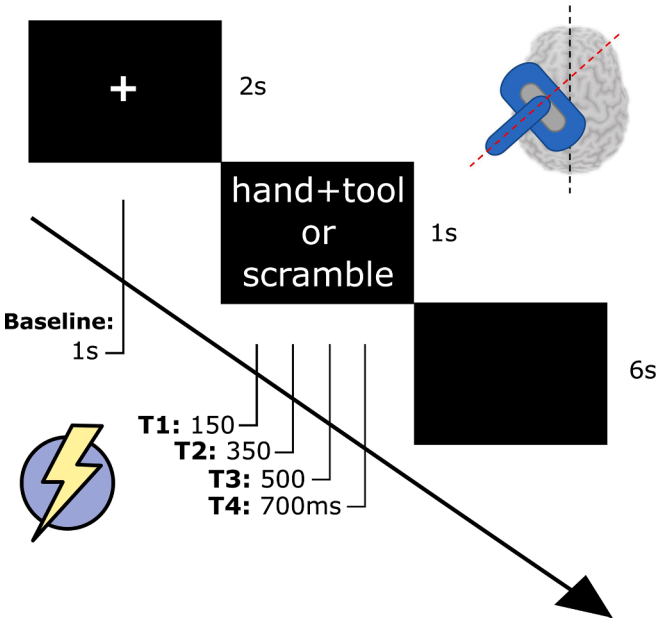

Fig. 2. Procedure. Experimental procedure with images' duration and TMS stimulation timings used in the present study.

2.4. Transcranial Magnetic Stimulation

TMS pulses were delivered by a Magstim Rapid stimulator (Magstim Company, Whitland, UK) with a standard 70 mm figure-of-eight coil placed on the skull with a medio-lateral orientation (handle pointing backwards). MEPs were recorded from the first dorsal interosseous (FDI) muscle of the right hand. Participants wore a piece of elastic tubular bandage covering the head with a grid of 1-cm resolution drawn on it. Following the international 10–20 EEG system, the coordinate origin

was fixed at the Vertex. Moving the coil on the grid by 1-cm steps (starting from the location of the electrode C3 in the 10/20 EEG system with an intensity of 55 % of the stimulator output), the hand motor area was localized on the left hemisphere at the beginning of each session. The optimal stimulation site and the resting motor threshold (rMT, lowest intensity able to evoke 5 out of 10 MEPs with peak-to-peak amplitude of at least 50 μV) were determined on FDI and subsequently used to evoke MEPs at an experimental intensity of 120 % of rMT (mean stimulator output across all experiments = 64.3 %). EMG recordings were performed with a Biopac MP36 (BIOPAC Systems, Inc., Goleta, USA) and data was continuously recorded by means of surface Ag-AgCl electrodes in a belly-tendon montage. EMG signal was acquired at a sampling frequency 2000 Hz, band-pass filtered (5–1000 Hz) and stored offline for further analyses. For each trial, the peak-to-peak amplitude of MEPs was calculated (Biopac Student Lab, v.4.1).

2.5. Analyses

EMG traces were visually inspected, and 2 participants of the Experimental group were discarded because of poor muscle relaxation or signal loss due to coil displacement during the recording. In the remaining participants (N = 33), trials showing abnormal background EMG activity (i.e. > 50 μV within a 150 ms time window before the TMS pulse) and trials where the peak-to-peak amplitude within a 150 ms time window after the TMS pulses was < 50 μV were removed from further analyses. Of the retained MEPs, those with amplitudes exceeding subject's mean ± 3 SD were considered as outliers. In total, we retained an average of 58 MEPs for each participant. Data were then log-transformed and for each participant and Timing (T1, T2, T3, T4), mean MEP amplitudes were expressed as difference from the Baseline MEP (log MEP_{Tx}-log $MEP_{Baseline}$). The means of log-transformed raw data were used for the statistical comparisons.

According to the experimental hypotheses, to compare groups and examine whether MEP amplitude turned out to be modulated over time, MEPs were submitted to a repeated measures analysis of variance (ANOVA) with Timing (T1, T2, T3 and T4) as within-subject factor and Group (Experimental and Control) as between-subject factor. Where required, post hoc comparisons were performed with Newman-Keuls test. Significance level was set at $\alpha = 0.05$. As a measure of effect size, η_p^2 is reported for each significant effect. Normality assumption was verified by means of Shapiro-Wilk tests. When required by sphericity violation (Mauchly's test), a Greenhouse-Geisser correction for sphericity was applied and corrected F values are reported. In addition, to investigate whether MEP amplitude differed from the Baseline level for each timing and group, we planned 8 (2 groups X 4 timings) one sample t tests (Hp_0 test value = 0). Significance level was set at $\alpha = 0.05$. As a measure of effect size, Cohen's d is reported for each significant effect. Normality assumption was verified by means of Shapiro-Wilk tests. Statistical comparisons were performed by means of the software Jamovi (v.2.3.28).

3. Results

Mean values of log-transformed MEPs for Group and Timing are reported in Table 1. For completeness, also raw MEPs data are reported in the same table. An overview of the statistical comparisons is reported in Table 2. Fig. 3 shows the temporal dynamics of MEP amplitude in the 2 groups. Shapiro-Wilk tests did not show violation of normality.

As for the ANOVA, analyses showed a significant main effect of Group ($F_{1,\ 31} = 4.456$, $p < 0.05$; $\eta_p^2 = 0.13$), indicating that MEP amplitude was smaller when observing hand actions than scrambled images. In addition, a main effect of Timing was observed (Greenhouse-Geisser corrected $F_{2.24,\ 69.59} = 4.618$, $p = 0.01$; $\eta_p^2 = 0.13$). Post-hoc t-tests showed a significant MEP decrease when timings T2, T3 and T4 were compared with T1 ($p = 0.04$, 0.007 and 0.008, respectively). Conversely, the timings T2, T3 and T4 did not significantly differ from each other ($p > 0.33$). Finally, no significant interaction was detected (Greenhouse-Geisser corrected $F_{2.24,\ 69.59} = 0.472$, $p = 0.65$; $\eta_p^2 = 0.01$).

As for the one-sample t tests, when observing pics of hand actions with tools, MEPs differed from the Baseline at 150 (T1, t = -3.56, p = 0.002; Cohen's d = -0.84), 350 (T2, t = -4.36, p = < 0.001; Cohen's d =

Table 2
Overview of Statistics.

ANOVA	SS	df	MS	F	p	η_p^2
Intercept	1,193	1	1,193	16,788	0,000	0,351
GROUP	0,317	1	0,317	4,456	0,043	0,126
Error	2,203	31	0,071			
TIMING	0,245	3	0,082	4,618	0,005	0,130
TIMING*GROUP	0,025	3	0,008	0,472	0,703	0,015
Error	1,644	93	0,018			

SS: Sum of Squares
df: Degrees of Freedom
MS: Mean of Squares
η_p^2: Partial Eta Square

ONE-SAMPLE T TEST	Student's t	df	p	Cohen's d
HAND PICs				
150	−3,56	17	0,002	−0,84
350	−4,36	17	<.001	−1,027
500	−2,74	17	0,014	−0,646
700	−3,49	17	0,003	−0,823
SCRAMBLED				
150	0,972	14	0,348	0,251
350	−0,517	14	0,613	−0,134
500	−1,911	14	0,077	−0,494
700	−2,377	14	0,032	−0,614

Note. $H_a\ \mu \neq 0$

Table 1
Mean Log-transformed and Raw MEP data.

MEP VALUES									
Mean Log Raw					***Mean Raw (mV)***				
	HAND ACTIONs		SCRAMBLED			HAND ACTIONs		SCRAMBLED	
	Mean	SD	Mean	SD		Mean	SD	Mean	SD
Baseline	0.045	0.272	−0.029	0.365	**Baseline**	1.545	1.164	1.595	1.474
150	−0.033	0.269	−0.004	0.374	**150**	1.357	1.136	1.668	1.581
350	−0.118	0.316	−0.051	0.402	**350**	1.205	1.184	1.552	1.434
500	−0.130	0.328	−0.126	0.366	**500**	1.129	0.947	1.254	1.096
700	−0.118	0.276	−0.122	0.317	**700**	1.117	0.937	1.191	0.911
150-Baseline	−0.077	0.092	0.025	0.100	**(150/Baseline)*100**	88.470	15.911	103.377	24.514
350-Baseline	−0.163	0.159	−0.022	0.162	**(350/Baseline)*100**	75.470	24.162	96.874	24.792
500-Baseline	−0.175	0.271	−0.097	0.196	**(500/Baseline)*100**	78.477	34.833	82.897	24.387
700-Baseline	−0.163	0.198	−0.092	0.150	**(700/Baseline)*100**	76.837	28.677	84.174	27.966

SD: Standard Deviation

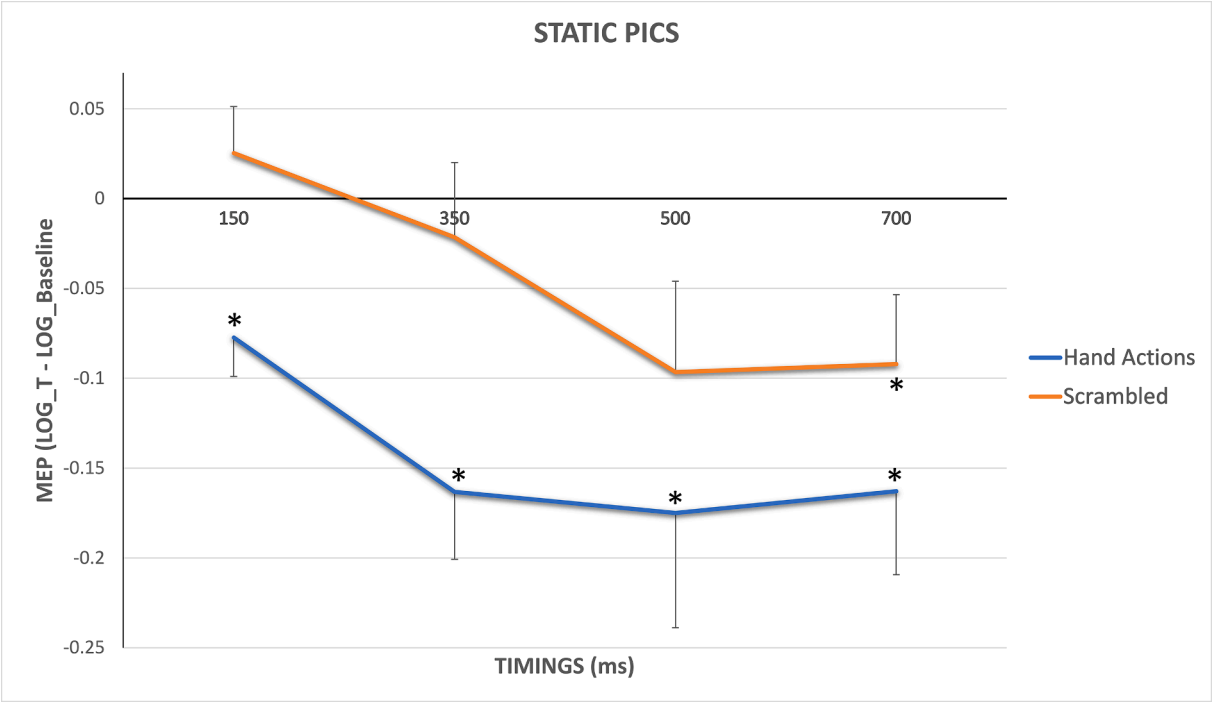

Fig. 3. Results. Mean amplitudes (Log_{Tx}-$Log_{Baseline}$) of log-transformed MEPs recorded from the right FDI muscle during observation of static pics displaying the hand in the act of using common tools (blue curve) and the corresponding scrambled stimuli (orange curve). Error bars indicate SEs. The different timings (ms) of TMS stimulation are reported on the horizontal axis. Note that '0' on the vertical axis corresponds to a MEP amplitude equal to that of the Baseline. Asterisks indicate significant comparisons at one-sample *t* test on log-transformed data. When significant, this test indicates that MEP amplitude in each group at a given stimulation timing is different from MEP amplitude during fixation cross observation (i.e. from Baseline, see text for details). (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

-1.03), 500 (T3, t = -4.36, p = < 0.001; Cohen's d = -0.65) and 700 ms (T4, t = -3.49, p = 0.003; Cohen's d = -0.82). At these timings, as indexed by the negative sign of the t statistics, MEP amplitude is significantly smaller than the Baseline. When observing pics of scrambled images, only T4 (t = -2.38, p = 0.03; Cohen's d = -0.61) showed a significant difference from the Baseline.

Overall, these findings indicate that observation of hand actions significantly suppressed corticospinal excitability as compared to observation of scrambled pics, while from 350 ms onwards, corticospinal excitability was suppressed irrespective of the group. Hand action-related suppression below the baseline clearly indicate the presence of early and enduring motor inhibition.

4. Discussion

The observation of pics with a still human hand in the act of using different tools (implied motion) induced a clear corticospinal suppression as compared to observation of scrambled pics. This suppression was already present at 150 ms from pic onset, peaked at 350 ms and was still observed up to 700 ms. Moreover, hand action observation induced a significant inhibition of the motor system as compared to the Baseline level (observation of fixation cross) at all investigated timings. These findings are in line with a previous study (Gianelli et al. 2020) showing that the sensation of a brisk action of the hand upon the same tools employed here (apparent motion) induced a similar MEP suppression lasting at least 500 ms in hand muscles involved in the observed action. The present study suggests that giving the observer the illusion of seeing a brisk hand action or presenting a still pic just displaying a snapshot of that action (i.e. the hand postured as if using the tool) basically produce very similar effects at the motor system level. This, however, does not mean that combining two pics instead of using just one is useless. In Gianelli and colleagues (2020), for instance, it was shown that at 200 ms after their onset, corticospinal excitability is not affected by the presentation of tools alone, while it is suppressed after the subsequent pic, showing the hand in the act of using the tool, is presented. It seems, then, that both the agent of the action (e.g. the hand) and its target (e.g. the tool) are necessary to suppress the MEPs. Showing two still pics to generate an apparent motion may then be considered as a valid alternative to the presentation of videoclips displaying the natural unfolding in time of bodily actions, especially when one intends to investigate the time course of motor resonance. While, on the one hand, videoclips were successfully employed to disentangle the instantaneous effect of observing specific stages of the ongoing movement, such as, for instance, the opening rather than the closure phase of the fingers during hand actions upon objects (e.g. Gangitano et al. 2001, 2004), on the other hand, the ongoing character of naturalistic actions may confound the measurement of the motor resonance time course with the extent of movement that has taken place (Cavallo et al. 2014). In fact, each subsequent timepoint in a videoclip will differ from the preceding one in terms of the amount of movement that has occurred, preventing the possibility to track the temporal evolution of the motor system response to a specific movement amount, unless no more changes in movement follow the one under investigation. This problem does not apply when presenting snapshots of bodily actions because the extent of movement, and thus the action phase, remains the same at all the investigated timepoints after the stimulus onset (here, a hand interacting with a tool). Moreover, since the amount of movement is available all at once, interference effects of predictive processes are ruled out. Predictive abilities are important during action processing (Kilner et al. 2004) but may obscure information about the motor resonance time course. Summing up, presenting still pics is a suitable strategy for studying the motor resonance time course.

In line with the literature on inhibitory motor control (e.g. Cai et al. 2011; Duque, Greenhouse, Labruna, & Ivry, 2017; Filevich, Kuehn, & Haggard, 2012), the functional role attributed to corticospinal inhibition in the observers is to prevent activation of motor representations,

elicited by the presented hand actions, from resulting in overt movement (e.g. Keysers, & Gazzola, 2010). In this context, cortical mechanisms leading to MEP suppression may consist of either the inhibition of M1 motor neurons or their disfacilitation. This interpretation is supported by studies showing the existence in monkey premotor and primary motor cortex (Kraskov, Dancause, Quallo, Shepherd, & Lemon, 2009; Vigneswaran, Philipp, Lemon, & Kraskov, 2013) and in humans (Mukamel, Ekstrom, Kaplan, Iacoboni, & Fried, 2010) of mirror neurons which were suppressed during action observation. However, another study employing static snapshots suggesting object manipulation (Urgesi et al. 2010), which are similar to the set of stimuli used in the present study, reported corticospinal facilitation for the presentation of still frames depicting a hand in the initial or intermediate phases of a grasping action, as compared to the end phase in which the action has been completed. Their results may seem at odds with the inhibition found in the present study. Nevertheless, facilitation and inhibition strictly depend on the reference used, namely the baseline condition against which the experimental conditions are contrasted. It has been shown (Hannah et al. 2018) that the baseline obtained prior to an action observation task turned out to be significantly smaller than the baseline measured within that task context (extra- vs. intra-task baseline difference of these authors). This finding points to a contribution of top-down processing in modulating corticospinal excitability, for example by attention- or arousal-related processes. Keeping this in mind, the facilitatory effects of Urgesi and colleagues (2010) could be at least partially explained by the use of an extra-task type of baseline. Moreover, reduced MEP amplitudes from a hand muscle were shown also when participants just imagined that their arm was paralyzed (Hartmann, Falconer, Kaelin Lang, Mueri, & Mast, 2022). Consequently, motor inhibition has been reported at neuronal and system levels, and in multiple functional domains; it then appears to be a substantial part of the engagement of the motor system also in action observation and motor imagery contexts. In addition, the finding that apparent motion is not essential for modulating corticospinal excitability offers possible insights about the neural underpinnings of such phenomenon. It is well known that movement-related aspects of visual information are processed by parietal areas and conveyed to premotor cortices via the dorsal stream, while visual features aiding object recognition reach temporal and then frontal areas via the ventral stream (e.g. Goodale, & Milner, 1992; Yang, He, Han, & Bi, 2020). It could then be argued that the processing of a still image displaying both an object and a hand properly interacting with it, following the ventral stream triggers the retrieval of a motor memory of the implied gesture in premotor areas. If so, the inhibition observed in the present study may reasonably be aimed at suppressing the ensuing hand action, irrespective of the route followed to activate motor representations. Finally, a decay of corticospinal excitability was observed also for scrambled stimuli which did not display hand actions. However, their overall ability to induce motor inhibition was significantly lower, as suggested by a significant decrease of MEP amplitude relative to the Baseline only at 700 ms. Since these stimuli do not display a clear image, it may be hypothesized that there is a progressive decrease of attention that could explain the facilitation decay. This, for instance, could be investigated by directly probing cortical inhibition (see below).

As for the temporal dynamics of excitability changes, in the present study the investigated temporal span was longer than that of Gianelli and colleagues (2020) and covered up to 700 ms. Based on the similar reactivity of both mu rhythm and mirror neurons in response to action observation and execution, Altschuler and colleague (1997) considered this rhythm as a marker of the mirror neuron system in humans. We then looked at mu dynamics in the literature to get insight about the duration of motor resonance effects. So, since alpha and beta power rebounds in central electrodes start at about 500–700 ms after the end of observed hand movements (Avanzini et al., 2012; Caetano et al. 2007; Muthukumaraswamy & Johnson 2004), and given the fact that the motor content of our still pics is available all at once at the onset of the stimulus and thereafter does not change (as if the movement ends just after its onset), we expected to find at least a reduction in corticospinal inhibition at 700 ms. A weaker or even abolished MEP suppression, as a possible correlate of EEG synchronization, is supported by a study which found that MEP amplitude was facilitated during spontaneous high amplitude mu-alpha waves (Bergmann, Lieb, Zrenner, & Ziemann, 2019; but see Sauseng, Klimesch, Gerloff, & Hummel, 2009). Contrary to our expectations, however, in the present study corticospinal excitability did not vary in the late investigated timings. The observed lack of recovery from inhibition at 700 ms suggests that corticospinal excitability does not correlate straightforwardly with EEG rhythms, at least when mu oscillations are not spontaneous but follow observed actions. It is possible that the recovery from suppression does not start until the offset of the static pic (here, after 1 s). In the future, this hypothesis should be tested by examining MEP amplitude after pic offset. Although the statistical interaction is not significant, the temporal profiles of corticospinal excitability in the 2 groups may however suggest a possible convergence over time. In a follow up, also this aspect could be verified by further lengthening the investigated time span. Finally, we did not explore whether the observed corticospinal suppression is accompanied by a variation in the activity of inhibitory and facilitatory cortical circuits. Measuring intracortical inhibition (ICI), for instance, might allow to evaluate if the present corticospinal modulation is actively instantiated by the cortical motor system to prevent overt movements or rather indicates a passive decay of activation's level. Future studies should better investigate this point.

5. Conclusion

In conclusion, the present study shows that the observation of still pics displaying the hand in the act of using common tools affects corticospinal excitability in a fashion very similar to what has been observed for stimuli with apparent motion of the hand with the same tools. This suggests that apparent motion and implied motion may trigger the same inhibitory motor effects. In addition, we extended previous findings on motor inhibition dynamics during hand action observation by showing a MEP suppression lasting at least 700 ms from stimuli onset. Motor inhibition, along with motor facilitation, shapes action processing not only during execution but also during observation of actions. The temporal profile of recovery from inhibition and the correlation between corticospinal excitability and mu rhythm dynamics still need further investigation.

CRediT authorship contribution statement

Riccardo Dalla Volta: Writing – review & editing, Writing – original draft, Validation, Supervision, Software, Investigation, Formal analysis, Data curation, Conceptualization. **Francesco Scarfone:** Writing – review & editing, Software, Investigation, Formal analysis, Data curation. **Dario Brambilla:** Data curation, Formal analysis. **Roberto Esposti:** Writing – review & editing, Validation, Formal analysis. **Paolo Cavallari:** Writing – review & editing, Supervision.

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Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Data availability

Data will be made available on request.

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