

Modulation of premotor mirror neuron activity during observation of unpredictable grasping movements

Massimo Gangitano,^{1,*} Felix M. Mottaghy^{1,†} and Alvaro Pascual-Leone¹

¹Laboratory for Magnetic Brain Stimulation, Division of Behavioural Neurology, Department of Neurology, Beth Israel Deaconess Medical Center, Harvard Medical School, 330 Brookline Ave., Boston, MA 02215, USA

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Abstract

Using transcranial magnetic stimulation, we explored the properties of premotor mirror neurons during the passive observation of a reaching–grasping movement in human subjects. Two different experiments were run using video-clips as visual stimuli. Video-clips showed a normally performed (control stimulus) or an anomalous reaching–grasping movement executed by delaying the time of the appearance of the maximal finger aperture (experiment 1), or substituting it with an unpredictable closure (experiment 2). Motor evoked potentials were recorded at different time-points during the observation of the video-clips. Profiles of cortical excitability were drawn and compared with the kinematic profiles of the corresponding movement. Passive observation of the natural movement evoked a profile of cortical excitability that is in concordance with the timing of the kinematic profile of the shown finger movements. Observation of the uncommon movements did not exert any modulation (experiment 1) or evoked an activity that matched, at the beginning, the modulation obtained with observation of the natural movement (experiment 2). Results show that the resonant motor plan is loaded as whole at the beginning of observation and once started tends to proceed to its completion regardless of changes to the visual cues. The results exclude the possibility of a temporal fragmentation of the resonant plan, because activation of different populations of mirror neurons for each phase of the ongoing action. They further support the notion of the role of the mirror system as neural substrate for the observing–execution matching system and extend the current knowledge regarding mechanisms that trigger the internal representation of an action.

Introduction

In recent years, a large body of neurophysiological evidence has provided insights into the visuo-motor properties of premotor cortex for the planning and control of goal-directed movements (for a review see Rizzolatti *et al.*, 2002). In monkey, the majority of premotor neurons (canonical neurons) become active when the animal plans a finalized movement or observes three-dimensional objects congruent to the shape of the hand that they code during actual grasping (Murata *et al.*, 1997; Rizzolatti & Fadiga, 1998). Other neurons (mirror neurons) are active not only during the actual execution of a movement but also during the passive observation of the same action that they plan (DiPellegrino *et al.*, 1992; Gallese *et al.*, 1996; Rizzolatti *et al.*, 1996a).

Comparable properties are present also in humans. Using transcranial magnetic stimulation (TMS), it was shown that passive observation of movements induces an increase in motor evoked potentials (MEPs) in the muscles involved in the actual execution of the same

movements (Fadiga *et al.*, 1995). These findings have been confirmed and extended in other TMS (Strafella & Paus, 2000; Baldissera *et al.*, 2001; Maeda *et al.*, 2002), magnetoencephalography (Hari *et al.*, 1998; Nishitani & Hari, 2000) and functional imaging studies (Grafton *et al.*, 1996; Rizzolatti *et al.*, 1996b; Grezes *et al.*, 1998; Iacoboni *et al.*, 1999; Buccino *et al.*, 2001).

In a previous TMS study, we showed the presence of a strict temporal coupling between cortico-spinal excitability and the dynamics of a reaching–grasping movement passively observed (Gangitano *et al.*, 2001). MEPs recorded from the first dorsal interosseus (FDI), at different times, during the passive observation of a pincer grasping action matched in time the dynamics of the pinch's kinematics of the actual grasping. We hypothesized that this modulation was the expression of the visual processing of movement features performed by mirror neurons (Gangitano *et al.*, 2001). However, it was not possible to disentangle whether this pattern of modulation was the consequence of comprehensive loading of the resonant plan at the beginning of the observation or whether the plan was fractioned in different phases sequentially recruited during development of the ongoing action.

The present study aims to address this question. We adopted a set-up comparable with that one used in our previous study (Gangitano *et al.*, 2001). A pair of visual stimuli were presented in two distinct experiments. The first stimulus was a video-clip of a natural reaching–grasping movement. The second video-clip represented an anomalous movement in which the temporal coupling between reaching and grasping components was disrupted, changing the time of appearance

Correspondence: Dr A. Pascual-Leone, as above.

E-mail: apleone@caregroup.harvard.edu

**Present address:* Dipartimento di Neurologia, Oftalmologia, Otorinolaringoiatria e Psichiatria, Unità di Neurologia e Riabilitazione Neurologica, Università degli Studi di Palermo, Italy.

†*Present address:* Department of Nuclear Medicine (Radiology III), University Hospital Ulm, Germany.

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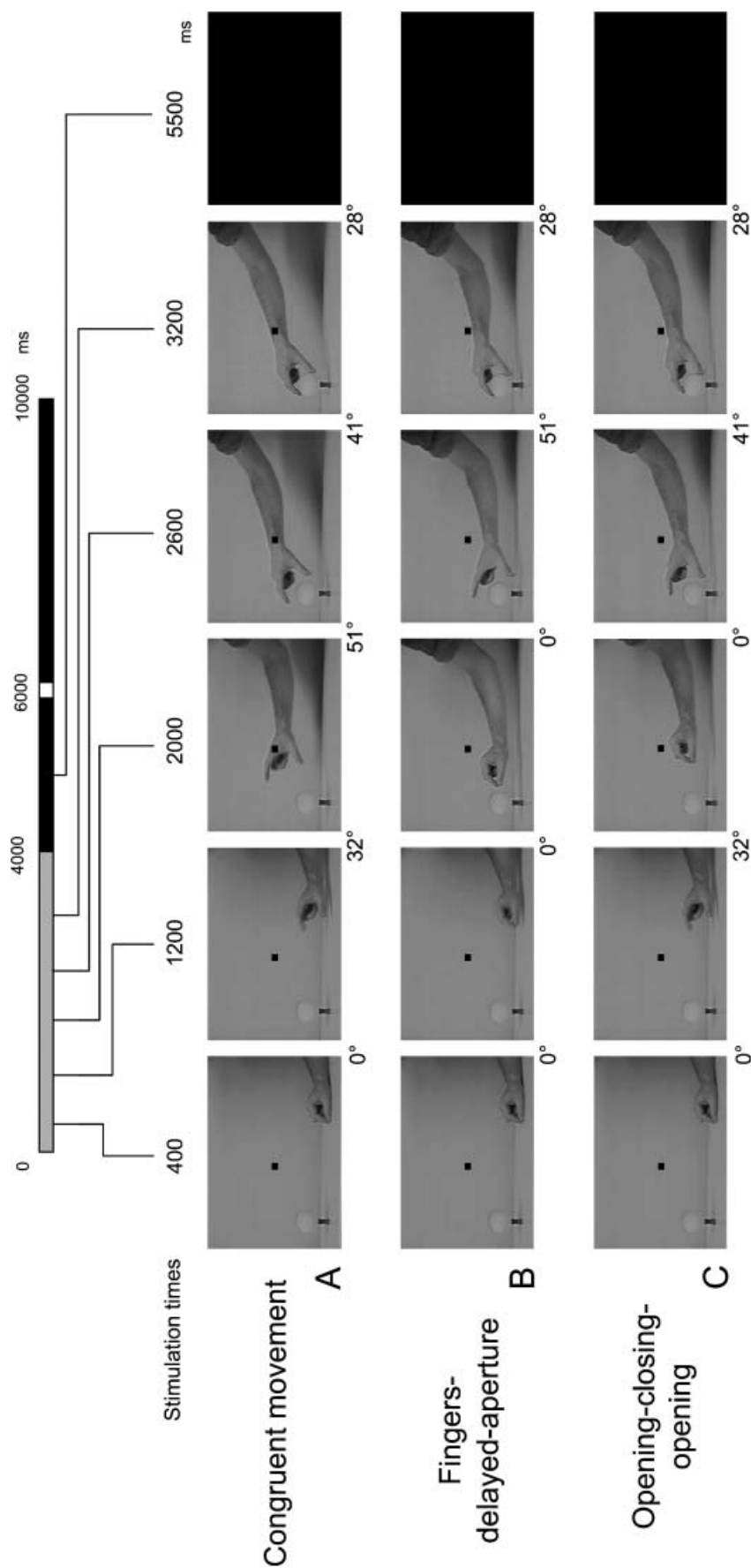


FIG. 1. The sequence of events during a single trial. Pictures are snap-shots obtained during delivery of single pulses in experiment 1 (video-clip A and video-clip B) and experiment 2 (video-clip A and video-clip C). At the top of the figure the continuous bar represents the duration of the trial (10 000 ms); the light grey bar is the duration of the video-clip (4000 ms), and the black bar is the black screen period (6000 ms). The vertical bars indicate the six time-points at which a TMS pulse could be delivered (400, 1200, 2000, 2600, 3200 and 5500 ms). Fingers aperture is calculated by referring their displacement with respect to anatomical reference points and is expressed as degrees of aperture, with 0° degrees at the starting position.

of maximal finger aperture. This effect was realized either by keeping the hand closed throughout the whole reaching and opening it just in proximity to the target (experiment 1) or substituting part of the natural finger opening with a sudden movement of closure (experiment 2).

Our reasoning is that if the changes of cortical excitability passively follow the temporal dynamics of the abnormally modified movements (i.e. peak of cortical activity at the same time as the delayed maximal finger aperture) it can be hypothesized that there are different populations of mirror neurons, each population coupled for each phase of the movement. On the other hand, if the motor output modulation is the result of deployment of the motor plan of the mirror cells at the initiation of the expected movement, there might be a disparity between cortico-spinal excitability and finger-aperture profiles.

Experiment 1

Materials and methods

Subjects

Eight right-handed subjects (five males and three females, mean age 30 ± 5.8 years) participated in this experiment after giving written,

informed consent. All subjects were unaware of the aim of the study. None had contraindications to TMS (Wassermann, 1998). The local Institutional Review Board (IRB) approved the study.

Stimuli

Stimuli were two digital video-clips (video-clips A and B) presented on a PC monitor. Both video-clips showed the right arm of a person reaching and grasping a ball. Grasping was a precision grip movement executed by means of the thumb and the index finger of the hand. The ball could be reached in two different ways: (i) in video-clip A (congruent movement; Fig. 1A) the reaching movement of the arm was coupled to a natural opening-closing movement of the index and the thumb; (ii) in video-clip B (fingers-delayed-aperture; Fig. 1B) the natural reaching movement was coupled to a delayed aperture of the fingers that opened only in proximity to the target. The starting point of the arm and target of movement were located on the same plane.

Beginning and end of movements were synchronized. Video-clips A and B had the same duration, i.e. 4000 ms sampled in 120 frames. In their first sequences, video-clips A and B were indistinguishable. Consequently, no cues were given to the subjects to guess how the action could develop until the movement was started. The end of the

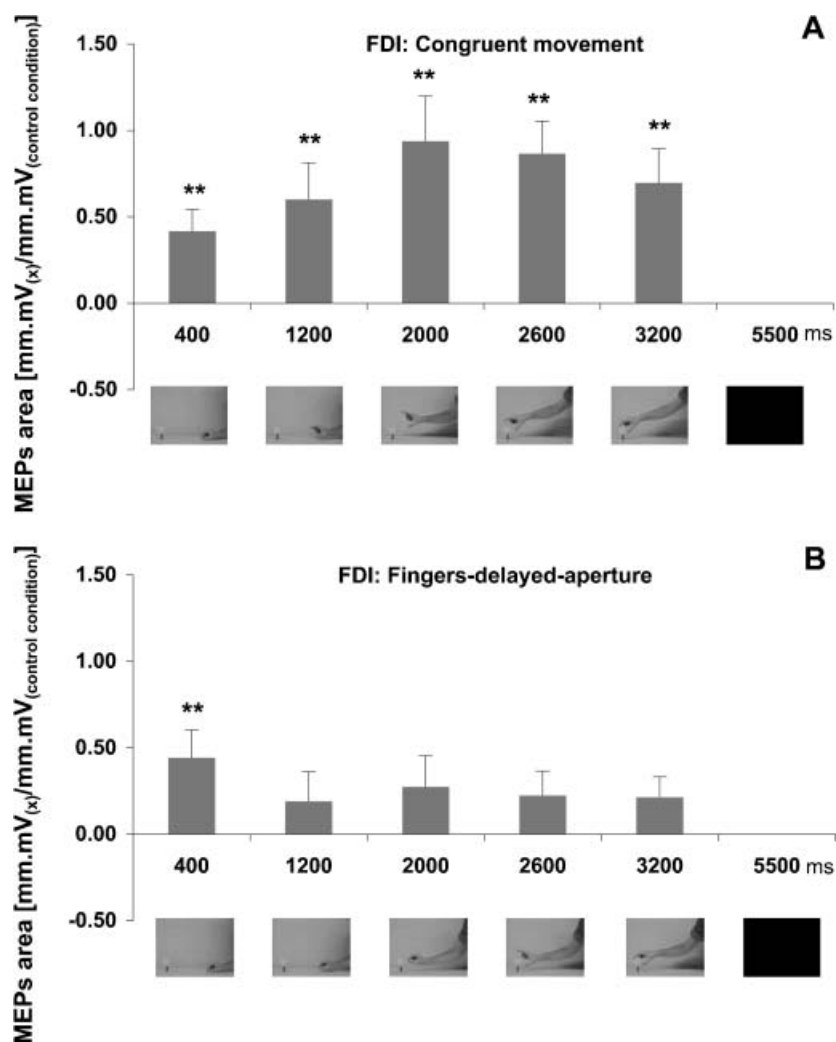


FIG. 2. Averaged MEP area of FDI collected, across the subjects, at the different time-points. (A) MEPs collected during presentation of video-clip A (congruent movement); (B) MEPs collected during the presentation of video-clip B (fingers-delayed-aperture movement). Whiskers are standard errors of means (SEM). Asterisks indicate significant variations ($P < 0.05$) with respect to basal values.

movements was immediately followed by the presentation of a black screen for 6000 ms. Each clip lasted 10 000 ms in total.

Procedure and task

Subjects were seated in a dimly illuminated room in a comfortable chair placed 1 m in front of a PC monitor. The graphic viewport for stimuli presentation was 18° of the subjects' visual angle.

A visuo-auditory task was designed in order to ensure that subjects paid attention to the stimuli. Subjects were asked to judge the position of the hand displayed on the video relative to a reference point on the screen at the time of presentation of an auditory cue (a modulated tone of 210 ms duration with a peak frequency of 4075 Hz). The two possible, alternative responses were: (i) hand on the right side of the reference point or (ii) hand on the left side of the reference point. Subjects were forced to choose one of the two responses even if they were unsure of the position of the hand at the time of the auditory cue. The reference point was a small red square shown between the hand starting position and the target location. The red square was turned off at the appearance of the black background. The PC generated the auditory cue in a pseudo-randomized order, at 500, 1000, 1500 or 2000 ms after beginning of the trial. Times of presentation of the auditory cue were chosen considering the position

of the hand relative to the position of the red square on the screen: in two of the trials (500 and 1000 ms) the auditory cue was presented before the hand reached the red square (hand on the right side of the screen), whereas in the other two (1500 and 2000 ms) the auditory cue was presented after the hand passed the red square (hand on the left side of the screen). Responses were given by pressing the right or the left button of a mouse with the index or the middle finger of the left hand, when a yellow square appeared on the screen 2000 ms after the end of the video-clips, during the period of presentation of the black background. Consequently, the period allowed for the response was 4000 ms (the time course of the experimental procedure is shown in Fig. 1).

TMS procedure

In each trial, a single TMS pulse was delivered at the time of appearance of one of six different preselected kinematic features of the congruent reaching-grasping movement as shown in video-clip A. Times of TMS pulses were: (i) at 400 ms with the hand still on the starting position; (ii) at 2000 ms, at the appearance of the maximal finger aperture; (iii) at 3200 ms, at the completion of the closure phase; (iv) a further stimulation was delivered at 5500 ms, i.e. 1500 ms after the disappearance of the video-clips with the black

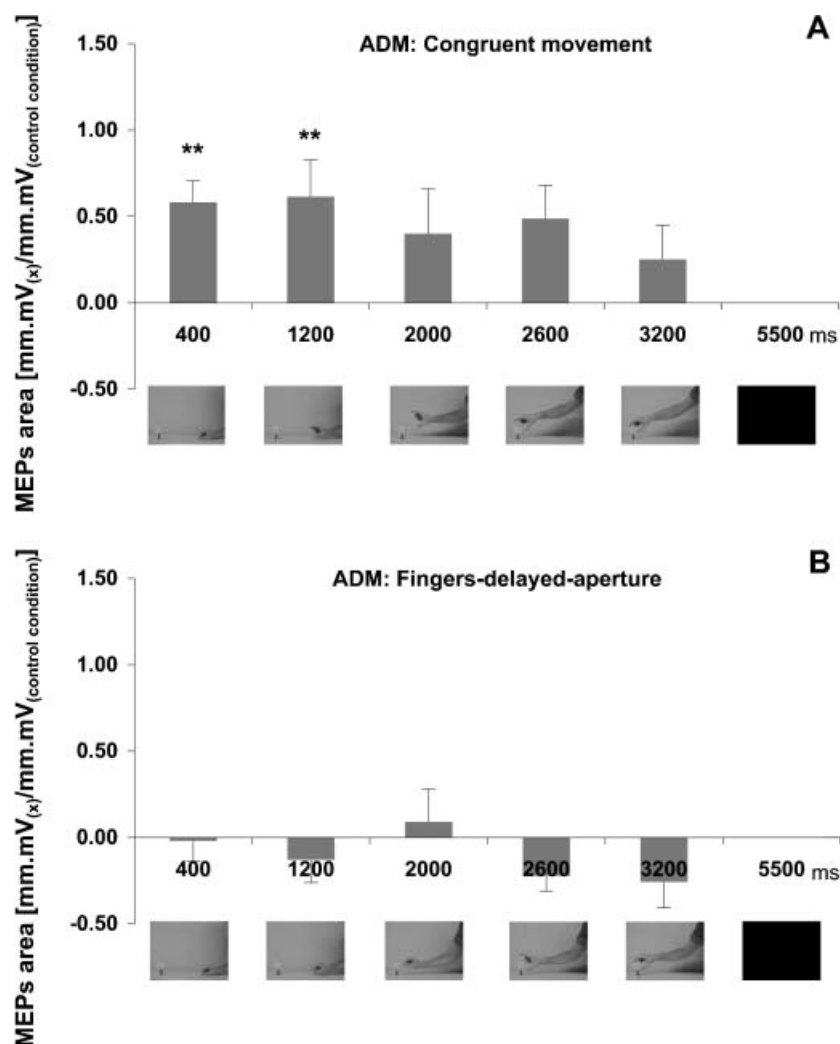


FIG. 3. Averaged MEP area of ADM collected, across the subjects, at the different time-points. Other details are as in Fig. 2.

background on the screen; (v) at two intermediate times between stimulation times 1–2 and 2–3 (1200 and 2600 ms). The 2600 ms stimulation time was chosen based on the timing of the appearance of the maximal finger aperture in video-clip B. The 5500 ms stimulation time was considered the control condition (see Fig. 1). The trials were executed successively. The minimum period between two TMS pulses was set at 5000 ms in order to minimize the potential risk of carry-over effect of a TMS pulse on the subsequent one.

Each video-clip was presented 96 times: the entire session was composed of 192 trials, 16 presentations for each of the six TMS conditions.

TMS stimulation and EMG recording

TMS was performed using a Magstim 200 transcranial magnetic stimulator (Magstim Co., Whitland, UK) and a 70-mm figure-of-eight coil. Four disposable surface electrodes were placed on the tendon and on the bellies of the right FDI and of the abductor digiti minimi (ADM) muscles. Circular ground electrodes were placed on the subjects' forearms and linked to common grounds. FDI was chosen because it is involved in the index finger–thumb pinch, whereas ADM was used as control muscle. EMG and MEPs were collected with a Dantec Counterpoint electromyograph (Dantec, Skovlunde, Denmark). The signal was amplified, filtered with a band pass of 20–1000 Hz, digitized using PowerLab 16S (AD Instruments Ltd, Hastings, UK) at a sampling rate of 2 kHz, and stored on a computer for off-line analysis. TMS pulses were delivered on the left hemisphere with the TMS coil held over the optimal scalp position for the induction of MEPs of maximal amplitude in the contralateral FDI muscle. With this TMS coil position it was also possible to record a stable signal from ADM in all subjects. The site of stimulation was marked on the surface of tightly fitting Lycra swimming caps that subjects wore during the experimental session. The coil was positioned tangentially to each subject's head surface, with the handle pointing occipitally, held at 45° from the mid-sagittal axis of the subject's head. This placement induces an electric current flowing perpendicular to the orientation of the central sulcus, and this has been shown to be optimal to achieve the lowest motor threshold (MT) (Brasil-Neto *et al.*, 1992; Mills *et al.*, 1992).

Single TMS pulses were delivered to the optimal scalp position in order to define the individual MT. MT was defined as the minimal intensity of stimulation capable of inducing MEPs of peak-to-peak amplitude greater than 50 μ V in at least six out of ten consecutive trials (Rossini *et al.*, 1994). Subjects were instructed to keep their right hand as still and relaxed as possible throughout the task. TMS pulses were applied at 110% of the individual MT.

MEPs were rectified, and the area under the curve was calculated and normalized with respect to the control condition. Data were submitted to a $2 \times 2 \times 6$ analysis of variance (ANOVA) with the recorded muscles (FDI vs. ADM), stimuli (video-clip A vs. video-clip B) and TMS stimulation times (400, 1200, 2000, 2600, 3200 and 5500 ms) as factors. Planned comparisons (LSD Fisher test) were employed as *post hoc* tests. Significance level were set to $P < 0.05$. Finally, by using a simple regression analysis, percentages of MEPs with respect to the control condition were related to the amount of finger aperture shown at the time of TMS application.

Results

Subjects gave a correct response, regarding position of the hand, at the time of the auditory cue in 98.6% of the trials.

In the three-way ANOVA, the interaction between video-clip, recorded muscles and TMS stimulation times was significant ($F_{5,35} = 3.78$, $P < 0.007$). Video-clip A was more effective in increasing cortical excitability than video-clip B ($F_{1,7} = 20.03$, $P = 0.02$).

During presentation of video-clip A, MEPs collected from FDI at the first five stimulation times were significantly different from those collected at control stimulation at 5500 ms ($P < 0.01$; Fig. 2A). MEPs recorded from ADM were significantly different with respect to the reference MEPs only at delays of 400 and 1200 ms ($P < 0.05$; Fig. 3A). When the delayed aperture was shown (video-clip B), MEPs from FDI were different from the control condition only at 400 ms ($P < 0.01$; Fig. 2B). No significant modulation of ADM across the stimulation times was found with video-clip B (Fig. 3B).

Video-clip A resulted in a significant correlation between angles of finger aperture, calculated at the different stimulation times, and MEPs recorded from FDI ($r = 0.357$, $P < 0.02$; Fig. 4A). No correlation was present between MEPs recorded from ADM and amount of finger aperture ($r = 0.097$, $P = 0.55$). During the presentation of video-clip B, correlations between finger aperture and MEPs were not significant both from FDI ($r = 0.23$, $P = 0.1$; Fig. 4B) and from ADM ($r = 0.05$, $P = 0.7$).

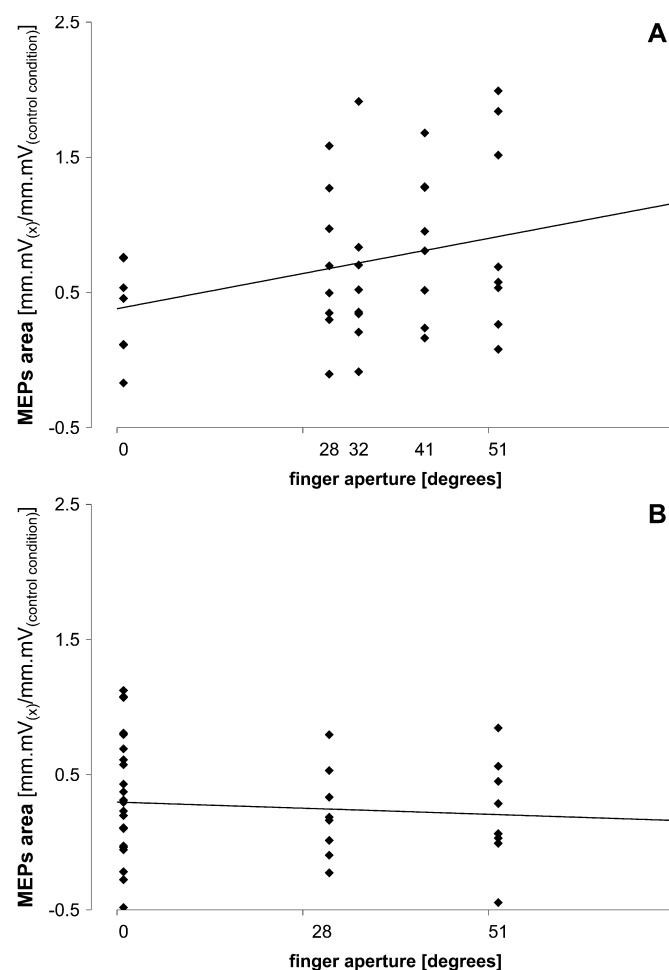


FIG. 4. Correlations between MEP area (vertical axis) of FDI and degrees of finger aperture (horizontal axis) during presentation of video-clip A (A) and video-clip B (B) in experiment 1. MEPs are normalized with respect to the control condition (5500 ms).

Experiment 2

In this experiment a new video stimulus (video-clip C) was used. Video-clip C matched video-clip A for the first 1200 ms and was characterized by a sudden closure movement inserted at the time of the expected natural, maximal finger aperture (opening–closing–opening, Fig. 1C). Times of movement beginning and end were synchronized. This manipulation was made in order to avoid the predictability of the ensuing movement by analysis of the first few frames of the video clips, as might have been the case in experiment 1.

Eight right-handed subjects (eight males, mean age 31.2 ± 4.6 years) participated in this experiment. None had participated in experiment 1. Subjects' selection criteria were the same as in experiment 1.

The apparatus was the same as in experiment 1. Stimuli were the same sequence of the natural reaching–grasping movement presented in experiment 1 (video-clip A) but with video-clip C instead of video-clip B was used.

Experimental conditions and procedures, data transformation and statistical design were the same as in experiment 1. During the presentation of video-clip C the 2000 ms TMS pulse was delivered

when the presented frame showed the fingers completely closed (see Fig. 1C).

Results

Subjects gave a correct response regarding position of the hand, at the time of the auditory cue, in 97.9% of the trials.

During the presentation of video-clip A, a significant modulation of MEPs from FDI was present from 400 to 3200 ms ($P < 0.01$; Fig. 5A). This confirms our results from experiment 1. However, presentation of video-clip C induced a significant modulation of MEPs from FDI only at the first three times, i.e. at 400, 1200 and 2000 ms (Fig. 5B). No significant modulation of ADM was found in both stimuli conditions (Fig. 6).

Regression analysis also confirmed the results of experiment 1. MEPs obtained during the congruent-movement observation were significantly related to the amount of FDI aperture ($r = 0.384$, $P < 0.01$; Fig. 7A) whereas no correlation between MEPs recorded from ADM and finger aperture was present ($r = 0.012$, $P = 0.93$). There was no significant correlation between finger aperture and MEPs collected both from FDI ($r = 0.088$, $P = 0.58$, Fig. 7B) and

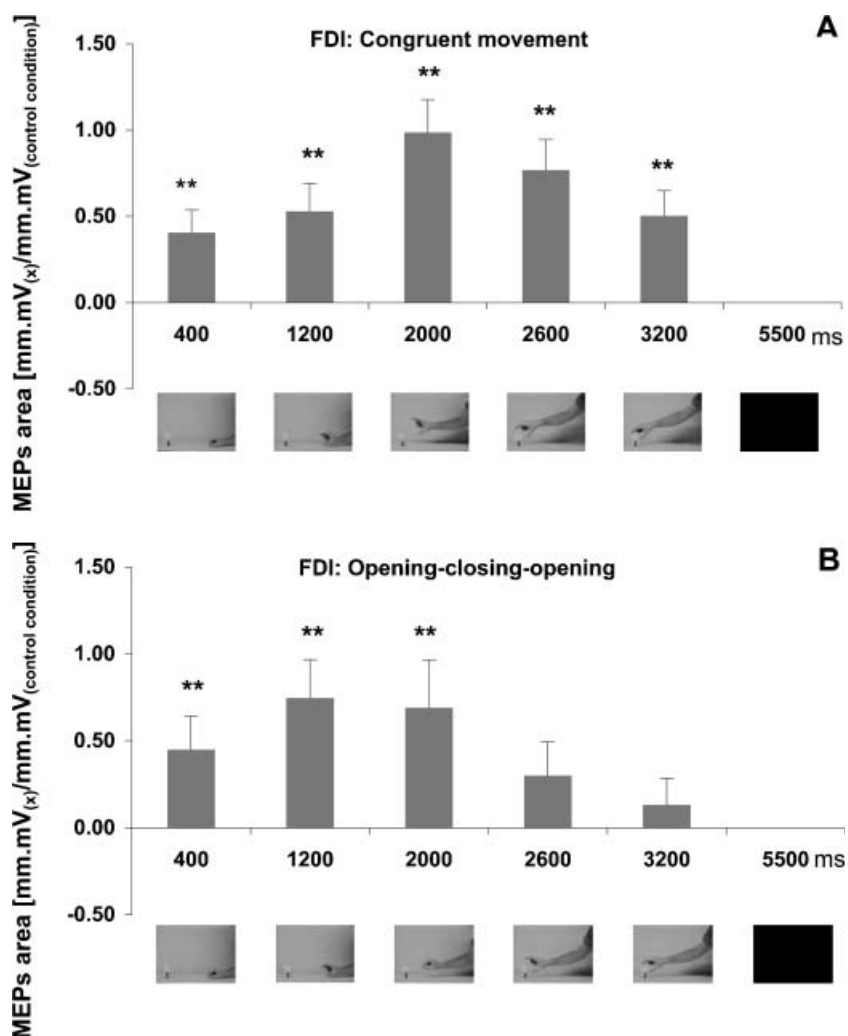


FIG. 5. Averaged MEP area of FDI collected, across the subjects, at the different time-points. (A) MEPs collected during the presentation of video-clip A (congruent movement); (B) MEPs collected during the presentation of video-clip C (opening–closing–opening movement). Whiskers are standard errors of means (SEM). Asterisks indicate significant variations ($P < 0.05$) with respect to basal values.

from ADM ($r = 0.07$, $P = 0.58$) during observation of the aperture–closure–aperture movement.

Discussion

In this study we explore the temporal dynamics of the modulation of cortico-spinal excitability during the passive observation of a reaching–grasping movement. We assume that this modulation is secondary to the input from premotor mirror neurons on motor cortical outputs. Specifically, we address the question of whether such a modulation is triggered in accordance with an expected motor plan and once initiated proceeds to the end regardless of the visual cues, or whether instead, it can be modified on a moment-to-moment basis by virtue of the observed movement.

Both these hypotheses can be sustained by several neurophysiological remarks. As shown using single-cell recording studies, canonical premotor neurons describe the action according to its goal (Rizzolatti *et al.*, 2001). In this case, the program may contain the sum of all kinematic components from its initial activation. However, action coding neurons can be clustered into different subsets, each one responsible for different aspects of the temporal segmentation of the movement (Rizzolatti *et al.*, 1988; Rizzolatti & Fadiga, 1998). For

example, a subset of neurons can discharge during the whole action, others can fire during the opening phase and others during finger closure (Rizzolatti *et al.*, 1988).

Mirror neurons have comparable properties. They usually describe the observed action in a comprehensive manner. However, whereas some show a broad depiction of the goal, others encode in detail the terms of how and when the goal is achieved (Gallese *et al.*, 1996; Rizzolatti & Luppino, 2001). It has been shown that some mirror neurons can discharge at the appearance of the final epochs of a movement, for example showing a stronger activity in the last phase of grasping (Umiltà *et al.*, 2001) or stop firing when the target is achieved or continuing to discharge after the end of the action during the holding phase (Gallese *et al.*, 1996). These last results suggest that mirror neurons may be able to encode some aspects of the timing of an action. Therefore, our initial hypothesis of the possible presence of different clusters of resonant cells coupled to different phases of a movement that are sequentially recruited cannot be excluded *a priori*. Moreover, mirror neurons seem to have different properties in humans and monkeys. In fact, in humans they can be activated also during action imitation (Iacoboni *et al.*, 1999; Nishitani & Hari, 2000) whereas in monkey this condition is ineffective (Gallese *et al.*, 1996).

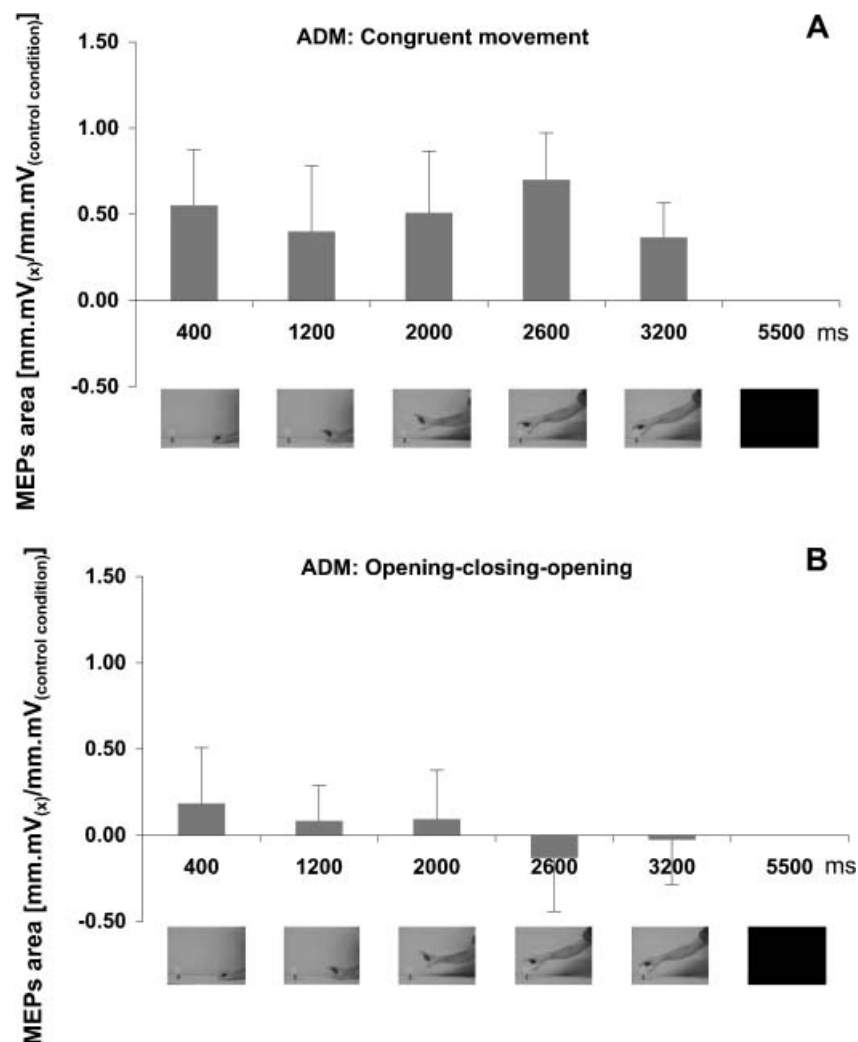


FIG. 6. Averaged MEP area of ADM collected, across the subjects, at the different time-points. Other conventions are as in Fig. 5.

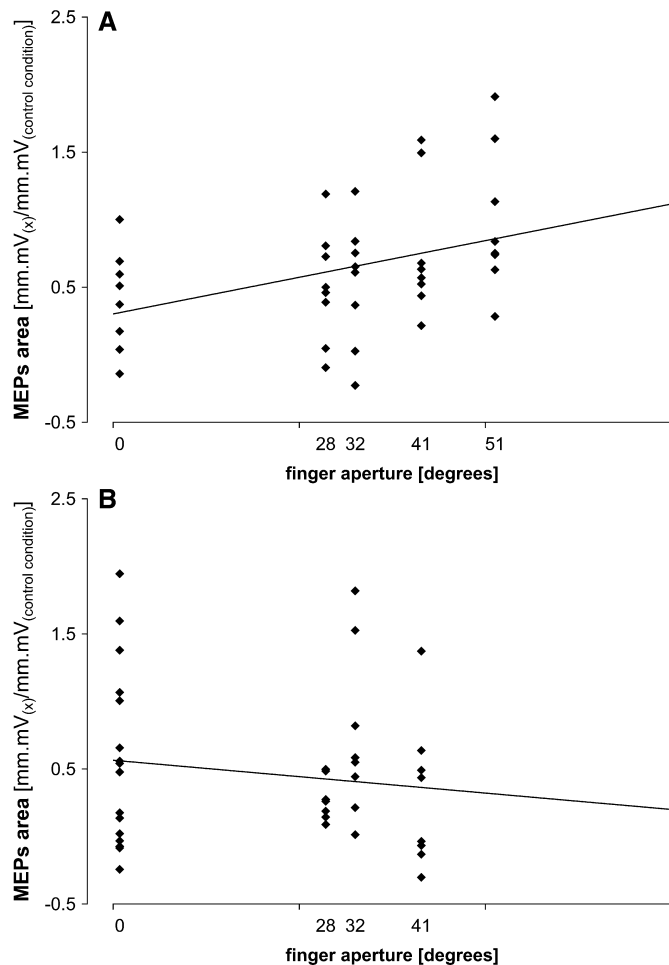


FIG. 7. Correlations between MEP area (vertical axis) of FDI and degrees of finger aperture (horizontal axis) during the presentation of video-clip A (A) and video-clip C (B) in experiment 2. MEPs are normalized with respect to the control condition (5500 ms).

In a previous study (Gangitano *et al.*, 2001), adopting a naturally performed reaching–grasping movement as visual stimulus, we found that the kinematic landmarks of the finger aperture were temporally correlated to the modulation of cortico-spinal excitability as indexed by the amplitude of TMS-induced MEPs. The amplitude of MEPs evoked by TMS pulses delivered at different time points during the passive observation of the movement was related to the amount of finger aperture. All MEPs were also matched in time to the kinematics of fingers: the smaller values were recorded when the fingers were closed and still placed at their starting position, whereas the time of appearance of the peak of cortico-spinal excitability was related to the time of appearance of the maximal finger aperture.

Grasping is a complex action that follows well-defined rules (Jeannerod, 1988). In the opening phase the fingers are shaped according to the physical features of the object to be grasped. The time during which the grip size is largest is fixed and is temporally coupled to the reaching (Jeannerod, 1984; Gentilucci *et al.*, 1991, 1992; Jeannerod *et al.*, 1995). This fine-tuned control is based on the existence of strong cortico-cortical facilitatory connections from ventral premotor cortex to primary motor cortex, especially from regions of hand representation (Cerri *et al.*, 2003; Shimazu *et al.*, 2004). These outputs have a primary role in the coupling of premotor and motor cortices during the execution of grasp-related actions (Cerri

et al., 2003; Shimazu *et al.*, 2004). It seems reasonable to hypothesize that comparable circuits could be activated not only during the actual grasp but also when a mirror activity is evoked. For these reasons, our findings of a correspondence between the dynamics of finger kinematics and cortical excitability during the passive observation of a grasp movement have to be considered highly significant and worthy of further investigation.

In the present study some features of ‘abnormality’ were artificially introduced to the stimuli by delaying the appearance of maximal finger aperture just before the final contact with the object (video-clip B) or substituting part of the maximal aperture phase with a sequence representing a sudden closure (video-clip C).

In experiment 1, during the observation of video-clip B, MEP modulation was generally absent. These results make the hypothesis of a step-by-step modulation of cortico-spinal excitability as a result of the sequential activation of different populations of mirror neurons unlikely. In such a case, the time of the peak of MEP activity should strictly depend on the time of presentation of the maximal finger opening, regardless of the moment of its appearance (late or early finger opening).

However, the lack of cortico-spinal modulation during the observation of video-clip B could also be explained by considering that an improbable movement such as that shown in the experiment 1 could not match any of the motor plans resident in the premotor cortex because of its novelty. In monkeys, highly skilled movements have stronger and more extended cortical representation than broader and less defined actions (Rizzolatti & Luppino, 2001). It is reasonable to suppose that novel, unnatural movements may have weak representations. It cannot be ruled out that an unnatural movement, such as that represented in video-clip B, once familiar, would eventually modulate cortico-spinal activity as achieved by natural grasping (video-clip A). Experiment 2 was designed to address this issue.

The observation of video-clip C induced a clear, significant modulation of cortico-spinal excitability. This modulation was limited to the first time-points checked, but in a comparable manner as induced by the natural movement (video-clip A). The modulatory effect was clearly suppressed by the appearance of the sudden finger closure and was not substituted by other patterns of modulation. In fact, the unexpected finger closure was not signaled by a depression of cortical excitability, but instead was substituted by a slow decay of the initial activation. The second, late, finger opening did not evoke any increase in activity. This suggests that the initial resonant plan neither took into account the modified features of the observed movement stimulus nor was substituted by a new plan in response to the new grasp. It seems that the original plan, once activated, loaded the depiction of the temporal features of the natural movement that it was discarded when these features ceased to match the visual properties of the observed movement.

It can be deduced that the mirror system is able to infer the goal and the probability of an action during the development of its ongoing features. Other neurophysiologic evidence supports this notion. In a single-cell recording study, Umiltà *et al.* (2001) compared object-directed actions in a full vision condition with when the same action was lacking its final portion. Mirror neurons became equally active in both cases, suggesting that an internal motor representation is generated in the observer’s premotor cortex even when the object of the action can be only inferred.

A biological movement usually follows well-defined rules that give it characteristics of suitability (Jeannerod, 1984, 1988; Jeannerod *et al.*, 1995). In our two experiments the observation of a movement with plausible or normal kinematic features evoked a greater cortico-spinal facilitation than the observation of unnatural movements. The

determination of plausibility of a movement requires access to a pre-existing description of the kinematics of movement and its goal (Rizzolatti *et al.*, 2001). This information reaches the premotor cortex through different routes (Giese & Poggio, 2003). Mirror properties, although with some remarkable differences, have been detected in several brain regions, including Broca's area (Rizzolatti *et al.*, 2001), the superior temporal sulcus (STS) region (Perrett *et al.*, 1990) and the inferior parietal lobule (Fogassi *et al.*, 1998). Mirror neurons in STS are responsive to the sight of biological stimuli but not to their movement (Perrett *et al.*, 1990). Mirror cells in the inferior parietal lobe seem to have intermediate properties between premotor and temporal features (Fogassi *et al.*, 1998). Avikainen *et al.* (2002) proposed that the ability to make proper judgements about motor acts and about their consequences requires access to one's own body schemes, represented in somatosensory cortices. The mutual interaction between premotor activity and sensory processing is evident in a very early stage of mirror response (Rossi *et al.*, 2002).

Movements requiring a high degree of precision, such as the precision grip, require high levels of coding, proportional to the skilfulness of the gesture (Jeannerod, 1988; Jeannerod *et al.*, 1995). If similar properties are present also for mirror neurons, movements demanding high neural control should be expected to recruit particularly high mirror cortical activity. This hypothesis could account for the overall greater activity during observation of the natural movement. Interestingly, mirror neurons are assumed to be the neural interface for action understanding (Rizzolatti *et al.*, 2001). If so, the unnatural movements in our experiments would be predicted to defy understanding as they appear to lack an adequate neuronal interface.

In both experiments the presentation of video-clips showing normal movements failed to induce a modulation of ADM activity. This result was predicted by our experimental hypothesis. Cortico-spinal neurons controlling muscles not directly involved in the active production of a movement, such as ADM, were included as a neutral control of FDI responses. It can be argued that we centered the TMS coil on the optimal spot for FDI and that a suboptimal placement of the coil for induction of MEPs in the ADM may have been responsible for the failure to demonstrate a modulation of its cortico-spinal projection. However, optimal scalp position for both FDI and ADM are within the spatial resolution of the employed coil (approximately 1 cm). Furthermore, the employed intensity of stimulation evoked MEPs of at least 500 mV peak-to-peak amplitude both for FDI and for ADM. Most importantly, suboptimal but consistent stimulation of the motor output to the ADM should have revealed a profile of modulation comparable with the FDI profile although with smaller values. This was not the case. However, it is worth noting that the MEPs induced in the ADM during the presentation of normal movements were larger than those evoked during the presentation of anomalous movements. This can be explained considering a synergic and co-ordinated contraction of the ADM as a postural support for the overall movement of the hand.

Finally, it is notable that observation of the immobile hand in the first frames of the video-clips triggered an unexpectedly characteristic modulation of cortico-spinal activity. This effect is consistent across all experimental conditions and independent of the video-clip presented. A conservative explanation could attribute it to an increase in the level of alertness due to relative temporal closeness of the 400-ms TMS pulse to the beginning of the trial. However, no EMG activity was present during the preceding period. For this reason, it is reasonable to suppose that the observation of a static picture, likely to evolve into a motor act, may be sufficient to induce a minimal, but significant, activation of the premotor circuits. In

other words, just the expectation of the predictably ensuing movement is sufficient to trigger a modulation of cortico-spinal excitability.

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Abbreviations

ADM, abductor digiti minimi; FDI, first dorsal interosseus; MEP, motor evoked potential; MT, motor threshold; TMS, transcranial magnetic stimulation.

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