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Anticipatory Postural Adjustments: from posture to movement

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INTRODUCTION

When performing a complex multijoint action, many components can be identified which contribute to the movement as a whole.
Central nervous system develop several parallel commands, each one with a specific goal, all organized in order to be included in the same action. (Arbib, 1981).
"In fact, the motor act might be compared to an iceberg, the apparent being the movement and the hidden part, which is often the most important, being the maintenance of reference values" massion 1992
Hess (1943) proposed that all the movements are composed by two main parts: the goal oriented movement and the postural related component which acts by assuring the equilibrium maintenance.
The existence of postural adjustments related to voluntary movement has been known since Babinski's publication (Babinski 1899), and many studies have reported changes in the activity of postural muscles associated with simple or complex voluntary movements.
Belenkiy et al. (1967) was the first that described the activation of leg postural muscle that precede (50-100ms) the activation of the prime mover in a arm movement. These anticipatory postural adjustments were interpreted as feedforward parallel commands aimed at minimizing the equilibrium disturbance associated with movement performance.
Since this pioneering study, anticipatory postural adjustments have been studied by axial movements, arm movements and leg movements.

Axial movements
The axial "synergies" associated with upper trunk movements described by Babinski (1899) belong to this category.
When a subject perform an upper limb movement the onset of the kinematic erector spinae or the rectus abdominis, and of leg muscles such as the hamstring-triceps suralis in the case of backward and the quadriceps-tibialis anterior in that of forward upper trunk movements. In the figure we can see the prime mover activation is preceded by the leg muscles activation, indicating the occurrence of a feedforward descending postural command.
Hip and knee movements occur simultaneously but opposite in direction to the upper trunk movements. It has been evaluated that, for this kind of movement, a 9cm displacement of

the CG would be expected (Crenna et al., 1987), nevertheless this postural control the result in a smaller displacement of cg (less than 2cm).

This results highlight the role of the APA that in this case well stabilize the CG that does not vary significantly during the movement execution.

Another example of axial synergies aimed at maintaining the stability of the CG projection onto the ground is provided by respiratory movements: the rhythmic trunk displacements are compensated for by hip displacements in the opposite direction and as a result no change in the center of pressure is to be observed in phase with respiration when the subject is standing on a force platform (Gelfand et al., 1971; Gurfinkel and Elner, 1973). Only under pathological conditions are these synergies lost (Gurfinkel and Elner, 1988).
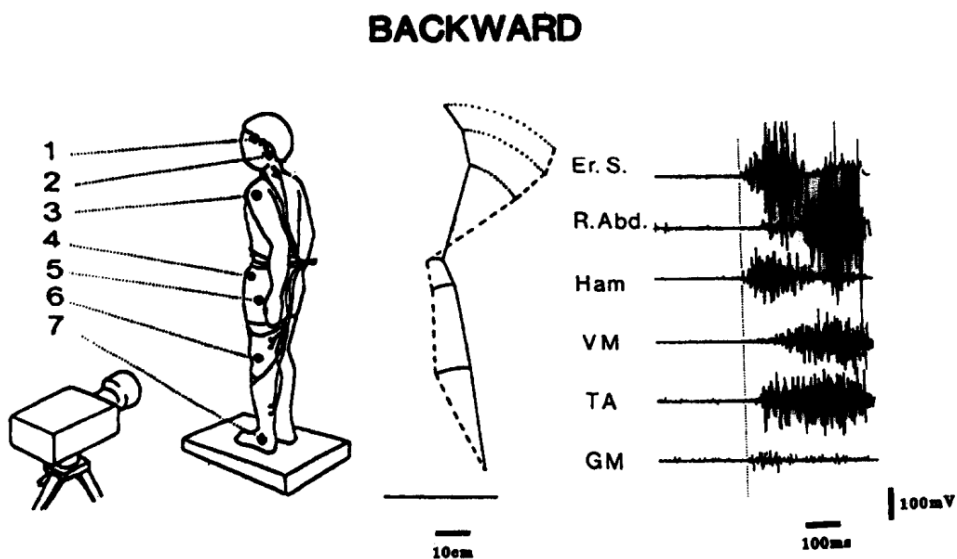
BACKWARD

Fig. 1 Backward upper trunk movement. Stick diagram shows that the backward movement of the trunk is accompanied by a forward hip and knee displacement. Continuous line, initial position; striped line, final position. A set of muscles in the back of the trunk and leg are activated fairly synchronously at an early stage after the "go" signal. Er.S. (erector spinae), Ham (Hamstring), GM (gnstrocnemius medius). The antagonist muscles are activated during the braking phase. (R. Abd., rectus abdominis; VM,vastus medialis; TA, tibialis anterior)

Arm movements

Several experiments of Bouisset and Zattara (1981, 1987b, 1988, 1990) deeply investigate the effect of the acceleration forces exerted on various body segments.

They observed that a unilateral or bilateral shoulder flexion is associated to APAs that create a movement with a force of inertia which will balance the inertial forces due to the movement itself.

Also in this case the CG displacement of the body, caused by the arm movement, is counteracted by the action of the postural actions.
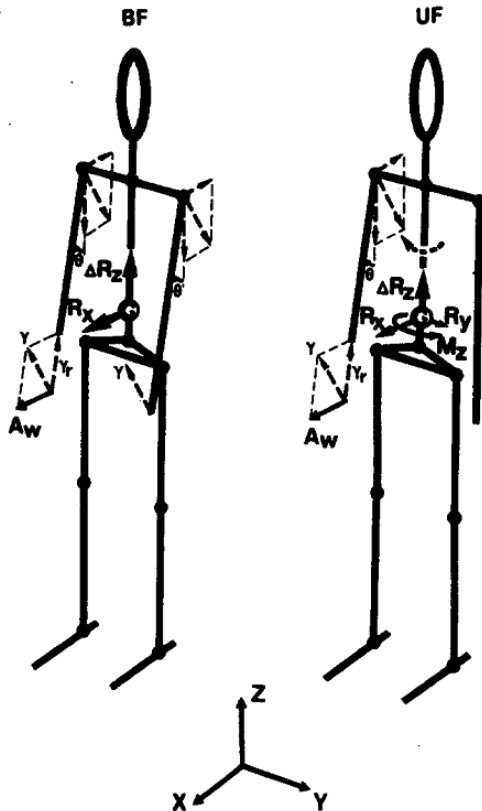

Fig.2 Interpretation of the purpose of the anticipatory postural adjustments (APA) associated with unilateral (UF) and bilateral (BF) arm raising. The filled arrows correspond to the actual recorded biomechanical data, and the dashed arrows correspond to theoretical parameters. 0, angular displacement of the upper limb(s). Aw, ~, r, and ~,, tangential, radial and total upper limb acceleration. Rx and ARz, antero-posterior and vertical acceleration of the body center of gravity, G.Mz, resulting momentum about the vertical axis crossing G. From this analysis, it can be assumed that APA tends to create inertial forces which, when the time comes, will counterbalance the disturbance to postural equilibrium due to the forthcoming intentional movement (Bouiset and Zattara, 1987b)

It's also worth noting that, considering the reaction time paradigm used in a arm raising movement with and without a load, the latency between the activation of the postural adjustment and the go signal remain constant while the prime mover activation was delayed when a load was added (Bouisset and Zattara, 1986, 1988, 1990; Zattara and Bouisset, 1986a, b).

The movement performed with the load is associated to longer APA (that increase the velocity of CG displacement), in this way the greater perturbation that would be caused by the load is counterbalanced from a more efficient postural action.

Leg movements

The equilibrium maintenance during leg movement is a particularly interesting case of postural control since the moving limb is the same that support the body.

Leg movement by changing the support condition cause a change in the CG position that anticipate the movement onset.

A general characteristic of leg movements is that they consist of a sequence where the CG first has to be displaced toward the remaining supporting limbs, and the movement onset is delayed until the CG displacement has reached a given value.

Alexeiev and Naidel (1972) were the first that described the APAs generated by a leg movement in human.

They described the anticipatory activation of tibialis anterior (TA) or triceps suralis associated to the dorsiflexion or ventroflexion on other side.

When a leg is rapidly flexed an initial displacement of the center of pressure (CP) toward the flexing leg is observed which is initiated by an early activation of the gluteus medius of that leg(Rogers and Pai, 1990).

Mouchnino et al. (1990) found analogous results in experiments where the subjects were asked to move a leg laterally to a height of 45 ° as fast as possible.

The first step in this motor act consists of displacing the body weight onto the supporting leg, by externally rotating the supporting leg around the ankle joint (anteroposterior axis). There again, an initial thrust exerted by the moving leg occurs prior to the CG displacement toward the supporting leg: a burst from the gastrocnemius medius (GM) occurs in the moving leg before the CP thrust. In most cases, the postural adjustment associated with leg movements serves to displace the CG projection toward a place which is compatible with equilibrium maintenance during the displacement of the moving limb. Here we have a motor act which is characterized by a sequential control where two different goals are achieved successively, the first consisting of displacing the CG projection toward the feet surface of the supporting leg or toward the tip-toes or toward the heels and a second of raising the moving leg. The movement onset is delayed until the CG displacement is achieved or about to be achieved.

This initial thrust seems to be also correlated with the center of mass acceleration toward the opposite leg. With slow movements, the initial thrust of the moving leg is absent and the knee extensor activity in the supporting leg first increases.

Stabilization of the position of given segments

Another role played by APAs is the the stabilization of different body segments, when performing movements.

During a bimanual load lifting task the anticipatory adjustment of forearm flexors acts counterbalancing the perturbation caused by the voluntary movement (Hugon et al., 1982; Dufoss et ; Paulignan et al., 1989).
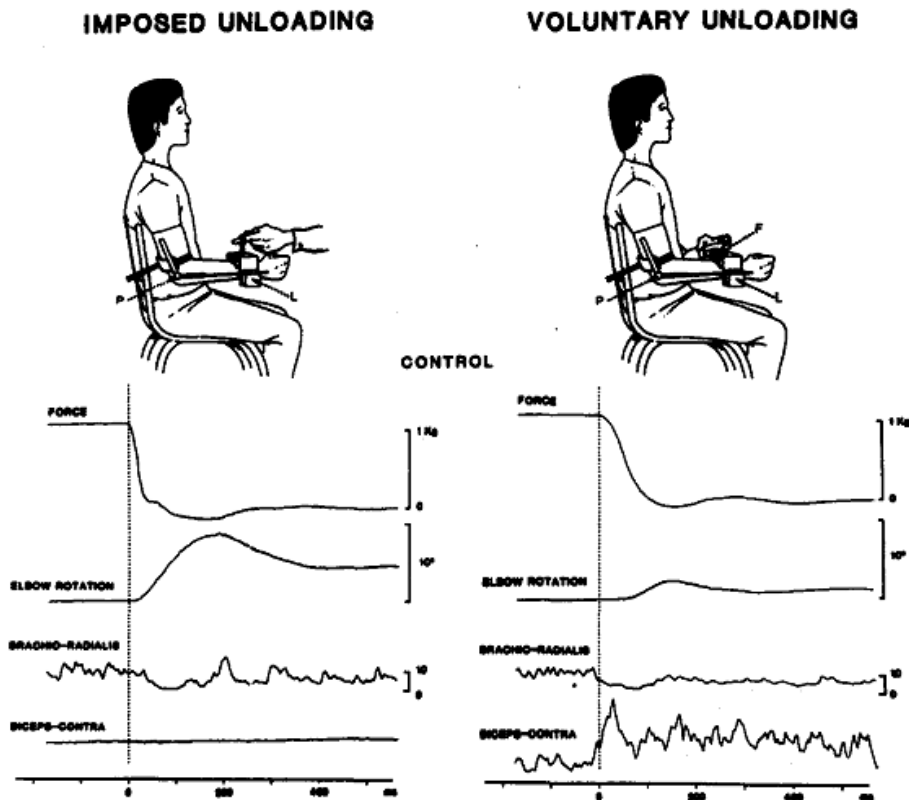

Fig. 3 Comparison between imposed unloading and voluntary unloading. The sitting subject maintains a loaded forearm (1 kg) in a horizontal position. The load is lifted either by the experimenter (imposed unloading) or by the voluntary movement of the other hand (voluntary arm). The vertical dashed line shows the onset of unloading. On the left, imposed unloading: the experimenter lifted the weight supported by the postural forearm. On the right, voluntary unloading: the subject himself lifted the weight with his other hand. Note that during voluntary unloading, the position of the postural forearm was maintained and a deactivation of the postural forearm flexors occurred before the onset of unloading. Average of 20 trials.

The unloading, the "postural" forearm by a voluntary movement of the subject's other arm is accompanied by an anticipatory inhibition of the postural forearm flexors, which is time locked with the onset of biceps contraction in the voluntary forearm. The

anticipatory adjustment was observed in a deafferented patient (Forget and Lamarre, 1990). It thus constituted a feedforward control aimed at minimizing the disturbance of the forearm position due to the unloading. Manipulation of heavy objects is, in fact, an old habit learned during childhood and the stabilization of the forearm position is a prerequisite for a careful exploration or manipulation of objects.

Body scheme

The proprioceptive Ia signal is the source of information necessary to create the representation of the body geometry. Roll and Roll (1988) suggested that the spindle input forms a continuous kinematic chain from the feet to the eyes.

Roll and Roll (1988) and Lackner, (1988) demonstrated that the internal representation of the body geometry could be changed by stimulating the proprioreceptors system at different level. Tendon vibration caused illusion of movement or generates postural adjustments.

Gurfinkel et al. (1988a) outlined a possible interpretation about the relationship between of the postural adjustment and the proprioceptive input. Previously Lund and Broberg, (1983) shown that stimulation of the vestibular system cause different postural reaction on the base to different position of the head with respect to the trunk.

Gurfinkel et al. (1988a) discovered that the neck muscle Ia inputs provide information about the head position in fact during unilateral vibration of neck muscles, which mimicks head rotation toward the opposite side, the postural reaction to vestibular stimulation is that which would be observed if the head was actually rotated.

One of the hypotheses proposed by Nashner (1977) and Cordo and Nashner (1982) in man and by Gahry and Massion (1981) in the cat was that there exists a repertoire of synergies providing a stable muscle pattern and that this repertoire may be utilized by sensory inputs associated with an external disturbance and by internal inputs associated with voluntary movement as well. This organization would reduce the number of degrees of freedom and simplify the problem of motor control in the domain of postural adjustment in line with the concepts of Bernstein (1967) and Gelfand et al. (1971). This hypothesis was based on the fact that a restricted number of muscle patterns could be observed during both postural reactions and anticipatory postural adjustments and that some of them were common to both

types of postural adjustments. According to Cordo and Nashner (1982), comparable patterns were observed when pulling a lever by the arm and when inducing a backward sway of the supporting platform. The possibility that fixed synergies may be the basis of postural reactions or anticipatory postural adjustments was further discussed in several studies who proposed that for each muscle pattern, a fixed synergy should be identified on the basis of the reproducibility of the spatial distribution of the pattern. In fact, as far as the anticipatory adjustments are concerned, the patterns described by most authors were found to be fairly reproducible. Several other sets of data have also suggested that the synergics are not fixed but flexible both in the case of anticipatory and reactional postural adjustments. For example, the distal muscles involved in the lower limb muscle activation associated with upper trunk movement can vary among subjects and even in the same subject (Pedotti et al., 1989; Oddsson, 1990). However, this is observed mainly during the first few trials, and the subject then tends to reproduce the same pattern and to create a "habit". Changes in the synergies were reported as a result of a change in the support conditions. For example, when a platform disturbance is delivered while a subject is standing on a short support base, a new pattern, the hip synergy, replaces the previous ankle synergy observed with normal support. This hip synergy corresponds to a new type of kinematic change centred in thc hip displacement which maintains the CG projection onto the ground (Horak and Nashner, 1986). This new synergy results from short term learning. Changes in synergy were also reported when upper trunk movements were performed by subjects standing on a narrow support basis (Pedotti et al., 1989). There, the change was found to occur only in trained subjects (gymnasts) and persisted for some time after the return to normal standing conditions. This suggests that the change in synergy resulted from short term learning.

Magnitude of vouluntary movement and apas
The process of generation of APAs is affected by a major factors: the features of the primary movements.
Fast movements characterized by a consistent inertia of the moving segments are associated to greater reaction forces (Lee et al, 1987), on the other side slower movements do not need apparent anticipatory postural adjustments (Horak et al., 1984, 1989b; Crenna et al., 1987; Oddsson, 1990).
The experiment demonstrated that a self-triggered perturbation is always associated with anticipatory postural adjustments, and that the magnitude of the adjustments may

be scaled with respect to the magnitude of a motor action used to induce the perturbation. (Aruin et al. 1995b)

APAs amplitude is augmented when the movement speed (e.g. Shiratori and Aruin, 2007; Lee et al, 1987) or the mass of the moved segment (e.g. Friedli et al, 1984) is increased and when the movement is performed against a resistance (e.g. Baldissera et al, 2008a). In the opposite conditions, APAs are decreased in intensity or even abolished. The relationship between APAs intensity and intensity of the prime mover contraction can be easily explained since APAs should depends on the amount of the expected perturbation, which in turn depends on the strength of the prime mover activation. In fact, strong prime mover contractions increase both the force exerted by prime mover proximal tendon and the interaction torques acting on the proximal segments: the perturbation of the segmental equilibrium is therefore amplified and so is the modification in whole-body geometry, eventually resulting in an increased displacement of the whole body centre of mass. Moreover, when the segment mass (e.g. Zattara and Bouisset 1988) or the movement speed (e.g. Horak et al, 1984) is increased, APAs latency becomes more and more anticipated with respect to the prime mover onset and, by contrast, when the movement is slow and/ or small masses are displaced, APAs onset can be even delayed after the prime mover activation. By combining the anticipated APAs onset with the larger APAs intensity, a greater counter-perturbation (see above) can fully develop in order to efficiently counteract the larger equilibrium disturbance caused by the stronger movement.

Direction and apas

A careful analysis on the basis of accelerometric recordings performed at the level of the various joints was carried out by Bouisset and Zattara (1987a, b, 1988) during uni and bilateral arm raising. These authors measured the acceleration forces associated with arm movements, calculated the reaction forces at the shoulder level and measured the acceleration forces and torques generated by the anticipatory postural adjustments prior to the movement onset. They showed that these "anticipatory" forces were directed in the opposite direction to the reaction forces associated with movement performance and thus served to minimize the postural disturbance caused by the movement.

The effect of the direction of perturbation on APAs was also studied in experiments where the subjects were instructed to perform bilateral shoulder movements in different directions, from forward to backwards with increments of 30 deg (Aruin et

al. 1995a). Bilateral shoulder movements gradually changed the magnitude of postural perturbations in the sagittal plane.
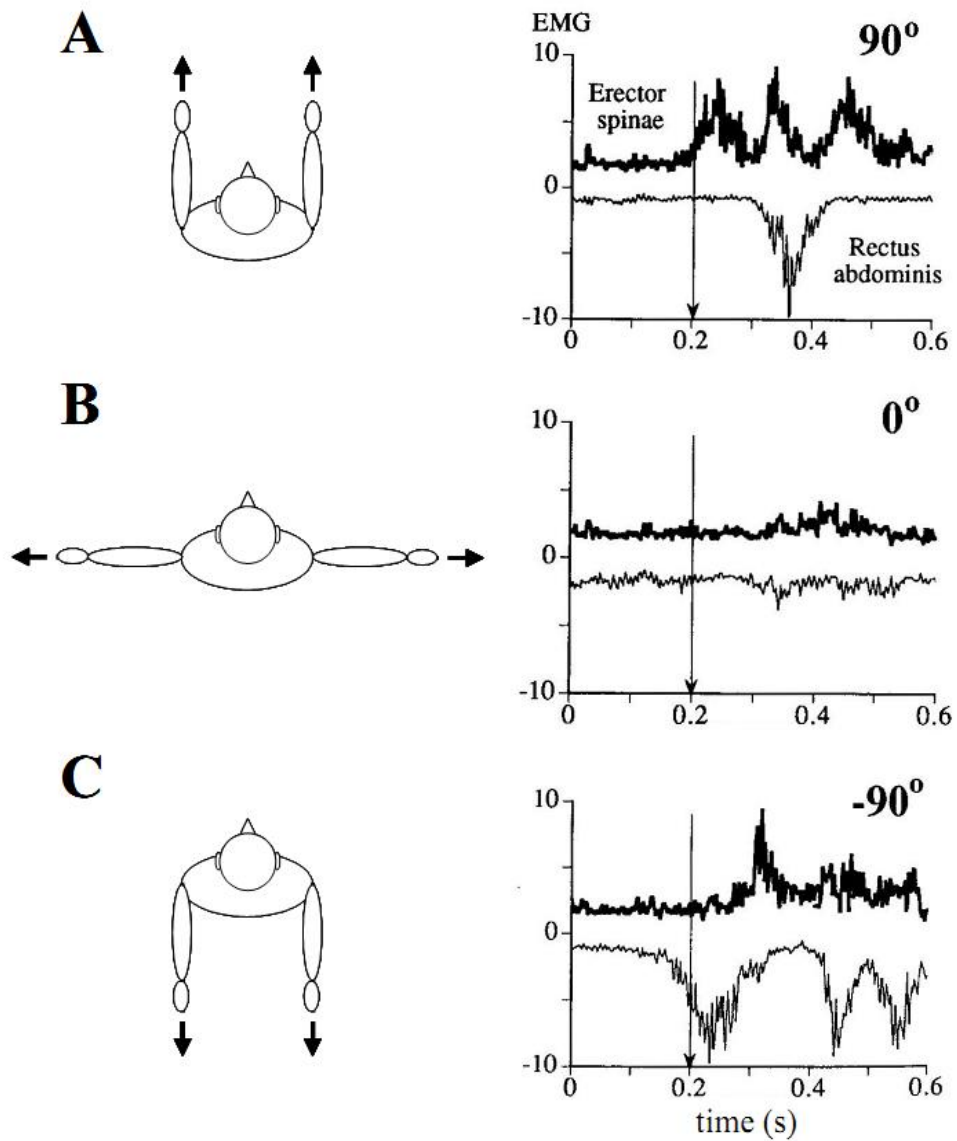

Fig. 4 movement direction reversal reverts APAs sign. Subjects performed bilateral shoulder movements in 3 different directions (flexion, A; abduction, B; extension, C). Graphs on the right show APAs recorded in the EMG of two antagonist trunk muscles (Erector Spinae, ES and Rectus Abdominis, RA). Shoulder flexion is accompanied by an excitatory APA in ES and no APA in RA; on the contrary, shoulder extension is preceded by RA activation and no APA in ES. No APA can be seen in the EMG traces when bilateral shoulder abduction are performed. Adapted from Aruin and Latash, 1995

Proximal postural muscles demonstrated the largest anticipatory increase in background activity during movements in one of the two opposite directions (forward or backwards). These changes progressively decreased when movements deviated from the preferred direction, and frequently disappeared during movements in the opposite direction. The patterns in distal muscles generally demonstrated larger anticipatory changes during movements forward and backwards as compared to movements in intermediate directions. The results demonstrated that a change in the direction of voluntary bilateral shoulder movements leads to changes in anticipatory EMG activity of both proximal and distal muscles of the dorsal and frontal parts of the trunk and legs.

An increase in movement velocity leads to earlier and more pronounced changes in the activity of postural muscles (Horak et al. 1984,Lee et al. 1990), while a decrease in movement velocity leads to smaller anticipatory postural adjustments, or even to their disappearance (Bazalagette et al 1987). The experiment demonstrated that anticipatory postural adjustments could be scaled with respect to the magnitude of a self-triggered perturbation(Aruin et al. 1996).

Speed and apas

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Posture and apas

The majority of studies of the effect of the postural task on APAs while standing were performed by modulating the stability of the body, using unstable surfaces or reducing the plantar support (Do et al. 1992, Gantchev et al. 1996, , Aruin et al. 1998).

The effect of postural task on APAs was obtained when lower extremities were involved in a perturbation. In particular, APAs were studied during leg flexion (Noiullot et al. 2000), lateral leg raising task (Mouchnino et a. 1992, Mille et al. 1998), while rising on tiptoe (Lipshits et al. 1981, Nardone et al. 1988). APAs were also studied in subjects standing on one leg (Aruin et al. 1988, Nouillot et al. 2000) and during standing inclined

forward. In the latter case, inclination was induced in the ankle joints, and a perturbation was induced by a load release from extended hands (Aruin et al. 1998).

The anticipatory EMG burst typically observed in the calf muscles was reduced when the subjects performed the same task while standing on a small plate placed on a narrow support (Pedotti et al. 1989).

Anticipatory postural adjustments were smaller when the posture was unstable. Instability in the sagittal plane had larger effects on anticipatory postural adjustments than instability in the frontal plane.

On a board with a small area of support the greater effects were on the magnitude of anticipatory postural adjustments (Aruin et al. 1998).

This study demonstrated that the magnitude of anticipatory postural adjustments, in conditions of a standard perturbation induced by a standard motor action by the subject, depends on two factors related to the postural task: the plane of postural instability and the area of support.

It would seem that in conditions of postural instability the central nervous system may be "unwilling" to generate strong anticipatory postural adjustments in order to avoid subjecting the fragile equilibrium to another source of perturbations.

In other experiments the subjects performed fast bilateral shoulder extension movements while standing. Body configuration was modified by instructions to the subjects to stand vertically or with a forward bend (Aruin 2002). The electrical activity of postural muscles and displacements of the center of pressure were recorded. Results indicated that APAs were modified with changes in the angular position of the upper body. The results of studies of anticipatory postural adjustments in conditions of postural instability taken together allow us to formulate the hypothesis (Aruin et al. 1998) that anticipatory postural adjustments themselves may be perturbations to balance, and that the lack of anticipatory postural adjustments in conditions of postural instability represents a defensive strategy of the CNS.

 Load and apas

In experiments where a load was added to the arm to be raised, it has been shown that the intensity of anticipatory postural adjustments is graded as a function of the postural disturbance expected from the movement (Horak et al. 1984, Dick et al. 1986, Bouisset et al. 1987, Aruin et al. 1995a).

Intra-limb APAs

From the sixties a large amount of literature has been mainly devoted to ''inter-limb'' APAs (see Cordo and Gurfinkel 2004). Conversely, the effects of focal movements on the segmental posture have been less investigated. Few examples concern movement of one arm and the related postural fixation in the contralateral side(see Hugon et al. 1982; Baldissera et al. 2008) or ''intralimb'' APAs developing in the ipsilateral arm prior to arm (Zattara and Bouisset 1988) or hand movements (Chabran et al. 2001). It should be also underlined that both inter-limb and intra-limb APAs precedes fast (Cordo and Nashner 1982) and large movements (Aruin and Shiratori2004), usually involving large mass segments, i.e. one or both arms, the trunk or the whole lower limb.

However, given that every movement should produce interaction torques on the supporting segments, our group were interested in answering the following major questions: (1) are interaction torques engendered by a gentle flexion of the index finger able to perturb the whole arm posture? (2) are interaction torques engendered by a gentle flexion of the index finger effectively compensated by an appropriate chain of APAs?

Caronni and Cavallari positively answered the first question by simulating a finger flexion in a four joint arm model and also by measuring, in a real arm, the reactive displacements ensuing to an electrically induced passive index finger tap. Subsequently, they observed that the postural changes predicted by the mechanical model and found in the in vivo situation, were, as expected, effectively minimised by APAs on the proximal joints. Lastly, they used the mechanical model to prove that interaction torques produced by finger movement may effectively change the final fingertip position. This was the first demonstration that large postural adjustments characterised by an important anticipation from the prime mover may be evoked by a gentle flexion of one single finger. These ''intra-limb'' anticipatory adjustments show striking similarities to the classical ''inter-limb'' APAs (see Cordo and Nashner 1982).
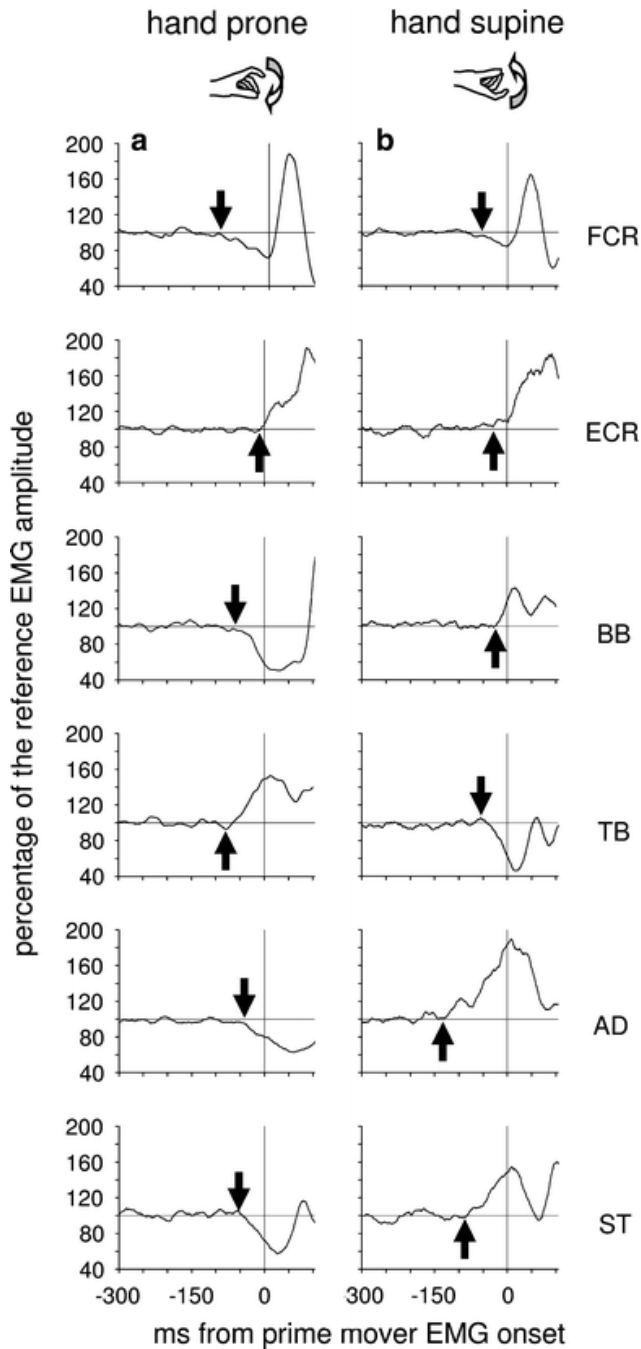
Fig. 5 Postural adjustments in upper-limb muscles preceding an index finger tap with the prone or supine hand. Each graph displays the APA onset (arrow) and its development on the tonic EMG from different postural muscles, of a single representative subject, with the hand resting prone (a) or supine (b). The vertical line at 0°ms marks the onset of the prime mover activity. Note that in the muscles acting at the elbow, the shoulder and the trunk APAs reverts in sign when hand posture changes from prone to supine. EMG is rectified, integrated and averaged (75 trials) and its size expressed in percentage of the mean EMG level recorded 1 s before the go signal

First, they are distributed to several upper-limb muscles and created a postural chain preventing the effects of the interaction torques generated by the focal movement (see Massion 1992). Second, they reverted in sign when hand posture was changed from prone to supine, i.e. when the direction of the focal movement was inverted (Aruin and Latash 1995). However, a special comment deserves the posture-independent anticipatory action observed in FCR (EMG reduction) and ECR (EMG increase). In fact, due to its trans-wrist course, FDS muscle acts as a wrist flexor both with the prone or the supine hand, thus its mechanical action, and the associated interaction torques on the wrist do not change with hand posture. Third, they changed in amplitude according to the level of postural stability (Nardone and Schieppati 1988; Aruin et al. 1998; Slijper and Latash 2000). In conclusion, according to these considerations and on the basis of the most common definition of equilibrium (i.e. a condition in which all acting influences are cancelled by others, resulting in a stable, balanced, or unchanging system) the natural anticipatory synergies coupled to gentle finger tap should be considered as genuine APAs.

Pointing

In those motor tasks in which the whole-body balance is not threatened, the importance of an accurate segmental stabilisation may look less obvious. Previously has been shown (Caronni and Cavallari, 2009a and 2009b), that when the index-finger is flexed, an APA chain develops in several upper-limb muscles to stabilise the segmental equilibrium of the arm. According to a mechanical simulation showing the consequences of a poor APA control on the movement trajectory Caronni and Cavallari proposed that APAs could be crucial to guarantee movement precision by an appropriate stabilisation of the proximal segments.

On these bases a successful pointing movement needs that prime mover activation and postural control are tuned each other; i.e. accuracy may be obtained by i) tailoring the prime movement to compensate for a loose APA chain, or by ii) tailoring the APA chain to aim a pre-set pointing movement.

A growing body of evidence clarifies the contribution of the proximal segments on both movement trajectory and speed of distal segments (e.g. Kaminski et al., 1995; Archambault et al., 1999; Pigeon et al., 2000; Pozzo et al., 2002; Bortolami et al., 2008; Kim et al., 2009). Conversely, at our knowledge, information about the linkage between APAs and voluntary movement accuracy may only be indirectly figured out from, e.g., Bonnetblanc et al. (2004), Nana-Ibrahim et al. (2008) and Bertucco and Cesari (2010). Indeed, these studies show that Fitt's law (movement time increases with movement

amplitude and decreases as target size increases, i.e. when the accuracy constraint loosens) applies both to prime movement speed and to the associated APAs. This observation actually provides an indirect suggestion that APAs are involved in attaining the movement precision necessary to accomplish the imposed accuracy constraint, however this is not a proof because APAs are also well known to be scaled according to movement speed (Shiratori and Aruin, 2007; Lee et al., 1987). The linkage between target and APAs size (e.g. Bonnetblanc et al., 2004) could then be just an epiphenomenon of the former relationship.

Immobilization

It is well known that joint immobilization leads to significant modification to the muscular and nervous system. Over the past few decades several authors have documented modifications in skeletal muscle properties such as atrophy (White et al. 1994; Hather et al. 1992), increasing in the intra-muscular connective tissue (Józsa et al. 1990) and reduction of the maximal voluntary contraction strength (Veldhuizen et al. 1993; Hortobàgyi et al. 2000) after cast immobilization.

In contrast, only a few papers have examined the neural adaptations to immobilization. Liepert et al. (1995) showed that immobilization leads to reduction of the cortical area of the inactivated muscle, while Facchini et al. (2002) demonstrated a decrease of the cortical excitability, without affecting nerve or muscle excitability, after only four days of motor restriction. Huber et al. (2006) found that these changes may even occur after twelve hours of immobilization. Moisello et al. (2008) demonstrated that short-term immobilization affect inter-joint coordination by acting on feed-forward mechanisms, while Avanzino et al. (2011) showed that a brief period (10 h) of right-hand immobilization decreased the excitability of left primary motor cortex and reduced interhemispheric inhibition from left-to-right hemisphere. Finally Langer et al. (2012) illustrated that the left cortical thickness of the sensorimotor cortex changed during right arm immobilization. Anticipatory postural adjustments (APAs), may play a key role in the process leading to the motor impairment induced by immobilization.

Mirror

Since the first description of neurons in the ventral premotor cortex of the monkey that fire both when the animal performs a certain action and when it observes the experimenter or another monkey performing the same action (''mirror neurons''; di Pellegrino, Fadiga, Fogassi, Gallese, & Rizzolatti, 1992), a number of investigations in monkeys as well as in

humans have been inspired by this discovery. Human research in this field has expanded quickly on the assumption that the ''motor resonance'' observed in man, i.e., the subliminal activation of the motor system when watching actions performed by others, were mediated by ''mirror'' neurons. In many of the studies exploring the possible role of this type of neurons in cognitive functions, the label ''mirror'' was often extended to neural mechanisms underlying any type of motor resonating responses, not only those to vision of body movements but also those to other sensory modalities (e.g., hearing), and even vegetative or emotional reactions (Warren et al., 2006; Wicker et al., 2003). Only a few investigations were focused instead on the mechanisms that supposedly transform   of their functional role, both as ''motor'' and as ''mirror'' neurons.

All these considerations underline the importance of investigating how the discharge of mirror neurons is translated into a motor act.

AIM

In all the experiments we investigated the relationship between the voluntary movement and the postural control. We concentrate our attention in particular on the role played by the APAs since the general hypothesis is that the postural feed forward control of the movement is strictly bound to the voluntary movement itself in a complex and flexible way. The thesis will be organized by dividing in the principal section (Introduction, Aim, Methods, Discussion And Conclusion), giving detailed description for each experiment. Then, I'll dedicate the last common part giving a general pictures that rise from a global vision of the described experiments.

Experiment 1

We tested the hypothesis that APAs preceding an upper-limb target reaching movement could play a role also in controlling the movement accuracy. Aim of this study was to seek a direct proof of the relationship between the APAs amplitude and the endpoint of a target reaching movement.

Experiment 2

The aim of this study was to determine whether a short term immobilization (12 h) interferes in parallel with both the activation of the prime mover muscle, responsible for a given movement, and with the postural muscles that are recruited to stabilize the limb.

Experiment 3

In this experiments we wanted verify if the postural activation is affected by the phenomenon of the motor resonance as well how already described for the prime mover activation.

METHODS

Experiment 1

Material and Methods

Ten right-handed subjects (4 females) were engaged (mean age ± SD: 26.9 ± 3.28 years). They reported no history of orthopedic or neurological disorder; none of them reported a reduction in the visual acuity. Each volunteer gave his/her informed consent to the experiment, which was approved by the local Ethics Committee in accordance to the 1964 Declaration of Helsinki. A variation of the "Belen'kii paradigm" (1967) has been adopted: standing subjects were asked to point-and-touch a target placed in front of them. The task was also performed while wearing and after doffing prismatic goggles.

Motor task

Subjects stood barefoot on a force platform, according to their spontaneous upright stance, while keeping both upper-limbs along the body. After an acoustic go signal, delivered every 5 s, subjects were instructed to perform a self-paced index-finger pointing movement, using right shoulder flexion, which was as fast and accurate as possible. Subjects were asked to hit the target, watch the final position they attained for no more than 1 s, return to the initial position at their preferred speed and finally relax before starting a new movement. By monitoring the EMG traces during the experiment, it was apparent that they returned to their baseline within 3 sec from the movement onset. The target consisted in two lines drawn on a Plexiglas screen, 1 mm thick x 2 cm long, one vertical and one horizontal, so that its center was clearly visible. The anterior-posterior, vertical and lateral position of the target was regulated for each subject: the target was positioned at the shoulder height on the subject's midline, one upper-limb length from his/her feet.

Experimental design

A couple of prismatic lenses (Fresnel 3M® Press-On, 20 diopters) were mounted on conventional safety goggles to produce a rightward shift of the binocular eye-field of about 11°. This corresponds to a 12cm linear shift of the target, placed at 60cm (about one arm length) in front of the subject. While wearing prisms, subjects realized the shifting in the binocular eye-field only when they performed the first pointing movement. Indeed, only when the finger reached the Plexiglas screen they became aware they had missed the target, also because the fingertip underwent the same visual position bias as the target

itself. Subjects were asked to close their eyes when donning and doffing goggles, and not to move their arms before the new recording session started.

Each experiment was arranged into three sessions of repeated target reaching: before donning (BEFORE), while wearing (DURING) and after doffing (AFTER) goggles with prismatic lenses. In each session, 25 trials of the target reaching task were performed. Between two subsequent sessions, subjects did rest for 5 to 10 min. Subjects donned the goggles just before starting the DURING session and kept them on throughout the following rest period. Goggles were removed only right before starting the AFTER session. No subject reported fatigue. They were allowed to familiarize with the motor task by practicing at least 15 target reaching movements before the first experimental session.

Recordings

In each experiment, electromyographic (EMG) activity, right upper-limb movement, target position and ground reaction forces were synchronously recorded.

EMGs were recorded from two muscles of the right upper-limb (Anterior Deltoid, AD; Biceps Brachii, BB) and four muscles of both the right and left lower-limbs (Quadriceps, Q; Hamstring, H; Tibialis Anterior, TA; Soleus, SOL). For each muscle, conventional disposable bipolar electrodes (1 cm diameter) were glued 25 mm apart on the skin covering the muscle belly. EMG signals were amplified (1-10 k) and band-pass filtered (30 to 500 Hz).

A 3D motion analysis system (SMART-D, BTS$^®$; 6 infrared cameras) was used to record both the right upper-limb movement and the target position. Reflecting spherical markers (1.5 cm $\varnothing$) were taped to the dorsal aspect of the metacarpo-phalangeal joint of the second finger, radius distal end, olecranon and acromion. Reflective tape was applied directly to the distal phalanx of the index-finger so as to resemble a hemispherical marker. This avoided placing a marker directly on the fingertip, which could interfere with the pointing movement. This method allowed the kinematics acquisition device to track the centre of the fingertip as all the other markers, with the same accuracy (±0.5 mm). A positive deflection on the marker trace indicates a leftward (x, right-left axis), forward (y, posterior-anterior axis) or downward (z, up-down axis) displacement. To identify target position, two hemispherical markers were glued on the Plexiglas screen, equidistantly above and below the target cross. Thus, target position was recognized as the xyz coordinates of the "virtual" marker placed mid-way on the line connecting the two.

A dynamometric platform (AMTI$^®$ OR6-7) was used to record forces (F) and moments (M) discharged to the ground with reference to the above axes.

Data acquisition was accomplished by the SMART-D workstation. EMG and platform signals were A/D converted at a sampling frequency of 1120 Hz, while cameras sampling rate was 70 Hz. EMG, kinematics and force signals were digitalized with 16 bit resolution and stored on a PC for offline measurements.

Data analysis

As stated above, subjects were asked to start the target reaching movement with a right shoulder flexion. Thus, AD muscle will be referred to as the pointing prime mover.

Movement onset (0 ms) was identified as the time when the y-position trace of the elbow marker crossed a threshold (set at the mean elbow position in the 500 ms preceding the go signal +2 SDs) for at least 100 consecutive ms. Movement end was instead identified, by the same threshold method, from the y-coordinate of the target "virtual" marker, signaling the impact of the index-finger on the Plexiglas screen. Systematically, timing measurements were visually checked and independently confirmed by two of the Authors (AC and FB). The pointing movement was assumed to terminate when the finger touched the Plexiglas screen.

We define pointing error the distance between the index-fingertip position on the screen and the target (see, for example, Ronchi et al. 2011, Chapman et al. 2010, Luauté et al. 2009). For each trial, the horizontal (x) and vertical (z) components of the pointing error were measured. Movement onset was chosen as reference point to leave enough time for APAs to fully develop (a similar approach is also applied elsewhere, e.g. Bouisset & Zattara 1987, Zattara & Bouisset 1988). Moreover, this reference allowed us to quantify the amplitude and latency of the anticipatory prime mover activation. In addition, this analysis allowed quantifying APAs amplitude in their mechanical actions on the ground, which develop together with or even after the prime mover activation.

Platform recording analysis was conducted on the three components of the forces exerted to the ground (Fx, Fy and Fz), on the displacement of the centre of pressure (CoPx, CoPy) and on the torque exerted about the z axis passing through the CoP (Tz). The position of the CoP and the value of Tz were derived from the recorded platform signals; Tz was calculated according to the following formula:

$$Tz = Mz + CoPy*Fx - CoPx*Fy$$

with Mz: moment about the vertical axis passing through the platform centre; CoPx and CoPy: right-left and posterior-anterior CoP co-ordinates in the platform plane, respectively.

For each trial, both EMGs and platform recordings were re-aligned on movement onset (0 ms). EMGs were rectified and then smoothed by a running average (time window 35 ms).

In each session, analysis of EMG and platform recordings was performed on trials 1-5 (1st BLOCK), in which pointing error resulted to be significantly different among sessions, and trials 11-15 (2nd BLOCK), in which the pointing error was similar in the three sessions.

The EMG and the platform traces within each block were then averaged to obtain a block mean trace (BMT). For each muscle, BMTs were normalized on the mean amplitude of the BMT recorded in the 1st BLOCK of the BEFORE session, thus allowing comparison between EMGs recorded from different subjects. Background activity (i.e. the mean amplitude of the BMT from -1000 to -500 ms) was finally subtracted from the EMG and platform BMTs. Voluntary EMG onset in arm muscles and APA onset in postural muscles, as well as in force platform traces, were identified as the time when the BMT crossed ± 2 SDs of the mean background activity level, and remained above that threshold for at least 50 ms. All onset timings were visually checked and independently confirmed by two of the Authors (AC and FB). APAs amplitude, or amplitude of pre-movement activation in AD and BB, was quantified as the mean amplitude of the BMT in a time window arbitrarily set from -25 to 0 ms (see also Caronni & Cavallari 2009a). It is also worth to note that, in the great majority of recordings (except for left Q in BEFORE) APAs and prime mover activation level at the 0ms corresponded to the highest pre-movement amplitude, and that APAs onset in EMG and platform recordings were, on average, much earlier than -25 ms. Data were analysed by a custom-made software.

Statistical analysis

A two-way repeated measures ANOVA, with prisms (BEFORE, DURING and AFTER) and trials (1-25) as factors was applied to horizontal and vertical pointing errors. A similar test prisms (BEFORE, DURING and AFTER) x blocks (1st and 2nd BLOCK) was also applied to i) the amplitude and timing of APAs, or of pre-movement activation in AD and BB and ii) the mean movement duration of trials 1-5 and 11-15. When ANOVA resulted in a significant main effect and/or interaction, Tukey HSD test was used for post-hoc comparisons. Significance level was set at 0.05.

Experiment 2

Materials and methods

Experiments, carried out in 5 male and 5 female adult volunteers, were approved by the Ethical Committee of the University of Milano, School of Medicine, in accordance with the standards laid down in the 1964 Declaration of Helsinki. All subjects gave a written consent

to the procedure, after being informed about the nature of the experiments; none of them had any history of neurological disease. Their mean (±SD) anthropometric characteristics were age, 29.4 ± 9.4 years; weight, 64.8 ± 14.2 kg; height, 169 ± 10 cm; index finger length, 9.2 ± 0.7 cm; and arm length, 76.9 ± 7.1 cm.

Experimental procedure

The subject was sitting in a chair with his dominant arm lying along the body, the elbow flexed at 90, and the prone hand in axis with the forearm. The index finger was kept extended and in contact with a proximity switch (CJ10-30GK-E2, Pepperl and Fuchs Mannheim, Germany), so that the metacarpophalangeal joint angle was about 180, all other fingers hanging. Subjects had their eyes open throughout the whole experiment and were explicitly asked to keep their back supported, the upper limb still, and both feet on the ground throughout the experiment. A wrist weight of 0.5 kg (Domyos Gym Weight, Decathlon, Villeneuve d'Ascq, France) was then wrapped around the distal end of the forearm. The subject position was always visually controlled by the experimenter. Subjects were asked to flex their index finger at the metacarpophalangeal joint so as to gently tap and rest on a flat surface. Each movement was self-paced and performed after an acoustic signal. The time between the beep and the movement onset varied according to the subject will. This procedure was adopted to exclude any reaction time.

In each experiment, index finger flexion was performed 120 times, divided into 4 sequences of 30 movement trials. The 30 trials were accomplished in a temporal window of about 2 min, and then the subject had time to rest (about 3 min) before undergo a new sequence. Subjects never complained about fatigue.

Movement and EMG recordings

The onset of the fingertip movement was monitored by the proximity switch. Flexion–extension of metacarpophalangeal and elbow joints was recorded by strain-gauge goniometers (mod. F35 and SG110, respectively, Biometrics Ltd, Newport, UK) fixed to the respective joint. Angular displacements were DC-amplified (P122, Grass Technologies, West Warwick, Rhode Island, USA), A/D converted at 2 kHz with 12-bit resolution (PCI-6024E, National Instruments, Austin, Texas, USA), and stored. Goniometer calibration was undertaken before each experimental session. Couples of pre-gelled surface electrodes, 24 mm apart (H124SG, Kendall ARBO, Tyco Healthcare, Neustadt Donau, Germany), were used to record the EMG signal from the prime mover Flexor Digitorum Superficialis (FDS) and from the following muscles that had an anticipatory postural activity: Biceps Brachii (BB), Triceps Brachii (TB), and Anterior Deltoid (AD). A good selectivity of the EMG

recordings was achieved both by a careful positioning of the electrodes and by checking that activity from the recorded muscle, during its phasic contraction, was not contaminated by signal from other sources. FDS activity was selectively recorded by positioning the electrodes on its course in the distal third of the forearm. To probe both excitatory and inhibitory APAs, the subject was requested to maintain a constant level of EMG activation in the BB muscle against the 0.5 kg weight suspended to the distal end of the forearm. The EMG was AC-amplified (IP511, Grass Technologies West War-wick, Rhode Island, USA; gain 2–10 k) and band-pass filtered (30–1000 Hz, to minimize both movement artifacts and high-frequency noise), A/D converted at 2 kHz, 12-bit (PCI-6024E), visualized, and stored for further analysis.

Mmax-wave recording

The amplitude of the surface EMG reflects the impedance of the structures interposed between the electrodes and the muscle (skin and fat). To exclude impedance variability from subject to subject, EMG amplitude was normalized to the maximal motor response (Mmax) evoked by orthodromic nerve stimulation (S8800 stimulator ? SIU5 isolation unit, Grass Technologies, West Warwick, Rhode Island, USA). Mmax was recorded at the beginning of each experiment. The subject was seated with both arms at sides and muscles relaxed; the experimenter positioned the stimulating anode dorsal to the clavicle and the cathode in the underarm. The M-wave was recorded through the same pre-gelled electrodes used during the experiment. The intensity of stimulation (duration 0.8 ms) was gradually increased, by controlling the resulting M-wave on an oscilloscope. Once exceeded the intensity necessary to elicit Mmax, the track was acquired at 10 kHz, 12 bits (PCI-6024E), and stored.

M-waves were measured peak-to-peak in mV.

Immobilization

At the end of the first part of the experiment (session PRE), performed during the morning hours, the subject was disconnected from the instruments, but the pre-gelled electrodes and the electrogoniometers were left on the skin.

In the late afternoon, the metacarpophalangeal and radio-carpal joints were then immobilized by a splint made of synthetic plaster (Dynacast, BSN medical, Hamburg, Germany), closed by an elastic bandage, whose length was equal to the distance between the distal ends of the fingers and the proximal third of the forearm. The splint kept wrist and

fingers in mid-range (neutral) position. Anyway, this apparatus allowed flexion–extension and pronation–supination at the elbow level.

The subject was then free to return to his/her normal daily activities. After 12–14 h, that is, during the successive morning, the splint was removed and the subject again performed 4 sequences of 30 finger flexions (session POST), followed by the evaluation of the Mmax. Data analysis In each session, the 120 EMG traces of the prime mover and those simultaneously recorded from the postural muscles were digitally rectified and integrated (time constant: 25 ms). Traces collected from each recorded muscle were then averaged in a fixed temporal window (-2000 to 300 ms from the onset of index finger flexion, detectedby the proximity switch). The period from -1500 to -1000 ms, free from APAs, was utilized to calculate a mean reference level that was subtracted from for each EMG trace. In each experiment, latency and amplitude of the postural activity were measured off-line on the averaged EMG trace. The onset of an effect in the postural muscle was identified by a software threshold set at ±2 SD of the reference signal level and visually validated. Latency of the APA was referred to the movement onset, thus assuming negative values. APA amplitude was measured as mean level of the trace in the temporal window from APA to movement onset and normalized to Mmax. In the same temporal window, for each subject, the SD resulting from averaging the EMG traces was used to estimate their within-session variability. Pooled within-session variability for each muscle was then computed as the RMS of the

individual values.

To assess whether APAs well-balanced the reaction forces of the primary movement, the peak-to-peak angular excursion of the elbow joint was measured from the onset of finger flexion to the moment when flexion started to be braked, that is, when its acceleration zeroed.

All comparisons between the two experimental sessions were performed either by paired t tests (session PRE vs. session POST) or by a repeated measures ANOVA with factors muscle (BB vs. TB vs. AD) 9 session (PRE vs. POST).

Experiment 3

Subjects were sitting 2 meters in front of a 45" computer screen, the elbow flexed at 90 degrees and the hand lying in supine position. Subjects were requested to keep a still position and to look attentively at a video-clip showing a human right upper-limb, with the

elbow flexed at 90 degrees and the hand, supine performing a ballistic hand flexion (see photo on the right). During the video the flexion movement was repeated three times.

The excitability of the motor pathways innervating the observer's FCR and the BB motoneurones was tested by measuring the amplitude of Motor Evoked Potentials (MEPs) evoked simultaneously in the two muscles. MEPs were elicited in FCR and BB during the third flexion movement, by using TMS (8-shaped coil) positioned on the contralateral cortex. They were recorded by conventional bipolar surface electrodes placed over the muscles belly, amplified, band-pass filtered, digitally converted and analyzed off line by a custom-made software.. The motor threshold (MT; intensity needed for evoking a visible MEP in 5 over 10 stimuli) was measured in the less excitable muscle (BB) and the stimulator output was set at about 120% MT. MEPs were elicited 220, 90, 60, 30ms before movement onset, at the movement onset (0ms), and 40, 100 and 160ms after. In each trial, the video was presented 16 consecutive times. During each presentation a MEP was elicited in BB and FCR at one of the above delays in random alternation, so that each interval was tested twice per trial. Each trial was repeated 5 times, to avoid fatigue, the subject could rest for 5min between two consecutive trials. For each subject, 5 trials were repeated. The background EMG in BB and FCR, as well as the elbow position were monitored on a oscilloscope and kept stable throughout all the video presentation

All subjects taking part to the experiment were also asked to imitate the wrist ballistic movement shown in the video clip, moving as much as possible in synchrony with it. This was done in order to disclose the temporal pattern of activation in FCR and BB and the occurrence of the postural activation during the overt movement. MEPs peak to peak amplitude was measured and averaged, in each trial, with those obtained at the same delay. Within each trial, the mean MEP amplitude at each delay was expressed in percentage of the MEPs amplitude evoked 220ms before the movement onset. For each delay a grand average of all trials in a single subject, and average value of all subjects was then calculated. A paired t-test was used to find significant variation from the baseline value.

RESULTS

Experiments 1

Prismatic lenses induce pointing errors in a target reaching movement

Before wearing prismatic lenses, subjects were very accurate in reaching the target: in each of them the index-finger final position scattered within a circle of 15 mm radius from the target centre (white dots in Figs. 6a, b). When the same movement was performed while wearing goggles (Fig. 6b, black dots), the index-fingertip hit the Plexiglas screen to the right of the target. However, within ten trials (prisms adaptation, see Fig. 6c, d) pointing fell again within the 15 mm circle. Pointing after goggles removal (Fig. 6b, grey dots), caused the subject to hit to the left of the target and, also in this case, the error faded away (recovery from prisms after-effect) after a few movement repetitions.

The mean time-course of the horizontal and vertical components of the pointing error (H and V, respectively) for each of the three experimental conditions (BEFORE, DURING and AFTER wearing prisms), is detailed in Fig. 6c, d. When subjects performed the motor task before wearing prismatic lenses, neither the mean H-error (2.0 ± 1.6 mm, mean ± SEM) nor the mean V-error (0.9 ± 1.0 mm) were different from 0 (one sample t-test, $P > 0.25$ for both variables). When subjects performed the first target reaching trial while wearing the prismatic lenses, a large rightward H-error occurred (-100.3 ± 16.4 mm; $P < 0.001$), while V-error was not different from 0 (-26.7 ± 23.9 mm; $P > 0.25$). Also after doffing goggles a considerable H-error occurred (33.5 ± 7.3 mm; $P < 0.001$), but now all subjects ended the pointing to the left of the target. Conversely, V-error (5.7 ± 5.1 mm; $P > 0.25$) was again not different from 0.

Two-way ANOVA on H-error showed a significant effect of both prisms ($F_{2,18} = 26.19$, $P < 0.001$) and trials ($F_{24,216} = 8.18$; $P < 0.001$), as well as a significant interaction ($F_{48,432} = 14.92$, $P < 0.001$). Post-hoc comparisons revealed that trials (i.e. time) had no effect in the BEFORE session, while there was a significant difference between BEFORE and DURING sessions for trials 1 to 5 (P always < 0.001) and, only for trial 1, between BEFORE and AFTER ($P < 0.002$). Note also that the adaptation process (trial 1-5 with prisms) is considerably longer as compared to the after-effect recovery (trial 1, after prisms).

Since a two-way ANOVA showed no significant modification on the V-error, the following analysis will focus on the H-error only.
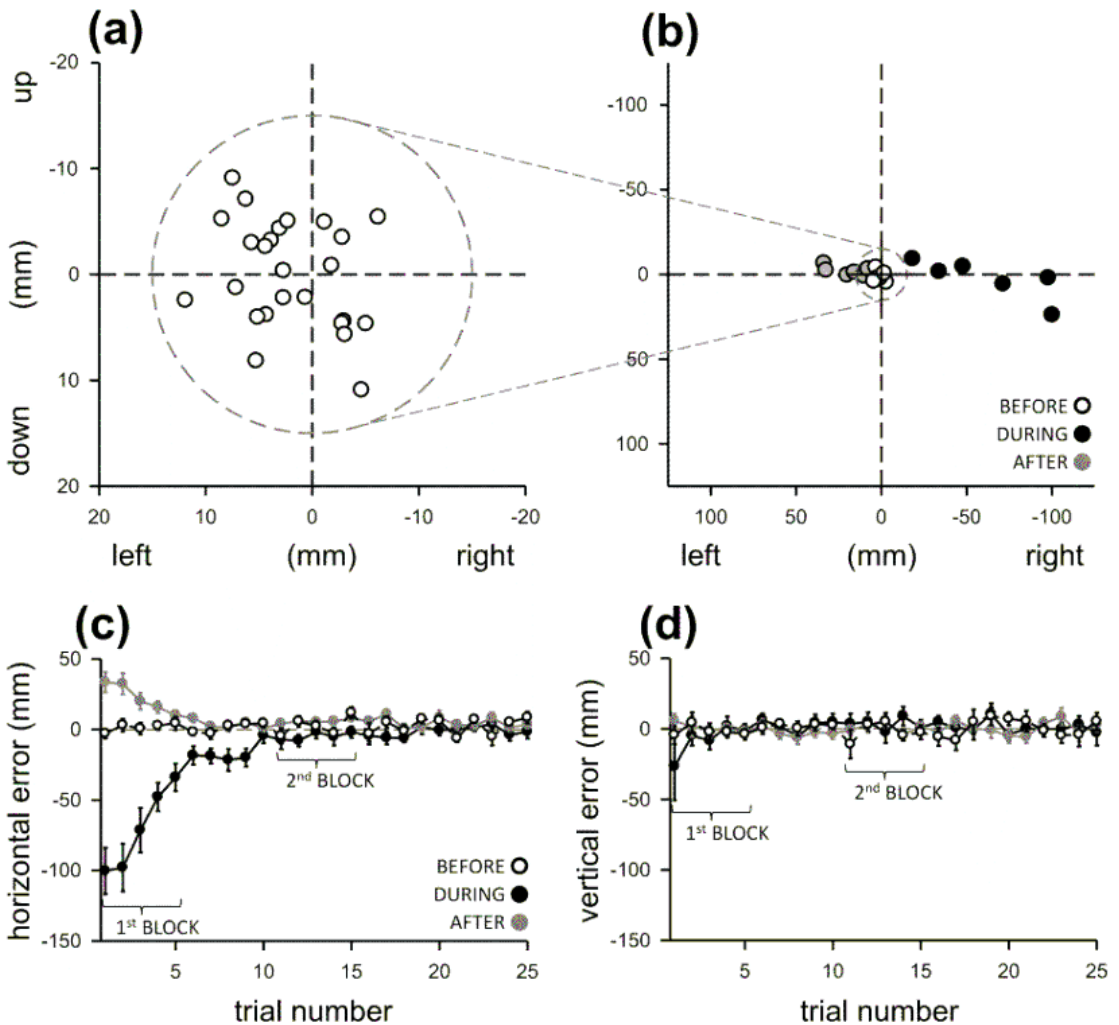
Fig. 6(a) Final position of the index-finger (average of all subjects) in each of the 25 pointing movement trials performed BEFORE wearing goggles with prismatic lenses (white dots). All points fell within a circle of 15 mm radius from the target centre (grey dashed circle). (b) Final position of the index-finger (average of all subjects) in the first 6 pointing trials, performed in the three successive experimental sessions (BEFORE, DURING and AFTER wearing prismatic goggles). While wearing prisms (DURING, black dots), the index-fingertip missed the target and pointing terminated on its right. After doffing prisms (AFTER, grey dots), the error reversed and pointing terminated on the target left, signalling an after-effect. (c, d) Average horizontal and vertical pointing errors (± inter-subject SEM) in each movement trial for the three experimental sessions (same labels and symbols as in b). The rightward horizontal pointing error observed in the DURING session recovered, in about 10 trials, to values comparable to those of the BEFORE session. The leftward error in the AFTER session had a lower amplitude than that in the DURING one, and recovered more quickly. A *prisms* x *trial* ANOVA found that the horizontal pointing error was significantly different among sessions only in trials 1 to 5. The same ANOVA design, instead, did not found any significant change in vertical pointing error. Thus, EMG and platform data from trials 1 to 5 (1st BLOCK) were matched to those of other 5 trials (11-15, 2nd BLOCK), in which pointing error was comparable among sessions, i.e., both adaptation to prisms and recovery from after-effect were completed.

Prismatic lenses modify APAs of a target reaching movement

As shown above, pointing errors of trials 1 to 5 were significantly different among sessions, thus a comparison of EMG and platform data from these trials (1st BLOCK – BEFORE, DURING and AFTER sessions) was carried out.

Similar between-sessions comparisons were also drawn in a second block of 5 trials (11-15, 2nd BLOCK), in which adaptation to prisms and after-effect recovery were apparently completed, and pointing errors were comparable among sessions.
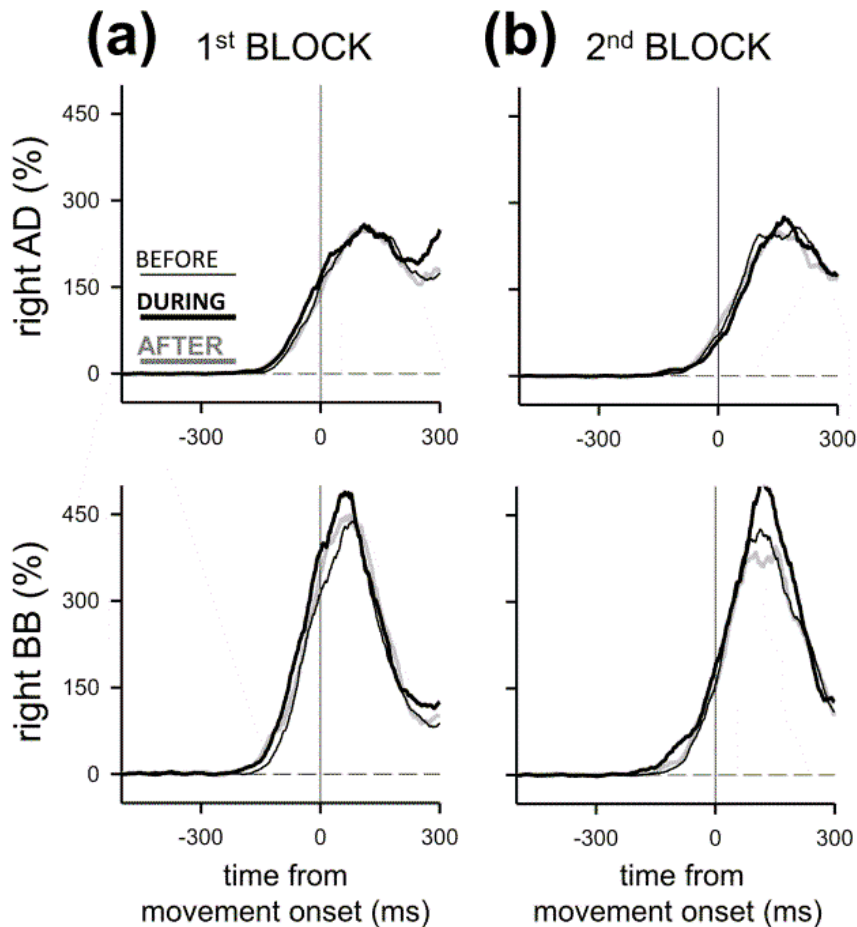

Fig. 7 Rectified and smoothed (see Methods) EMG recordings from the prime mover Anterior Deltoid (AD) and from the Biceps Brachii (BB) muscles. Averaged traces of one representative subjects, obtained in trials 1-5 (a - 1st BLOCK) and in trials 11-15 (b - 2nd BLOCK) of the three experimental sessions: BEFORE (thin black line), DURING (thick black line) and AFTER (thick grey line) wearing prismatic goggles. Note that, in each BLOCK, the pre-movement (before 0 ms) EMG activity was nearly indistinguishable in all sessions.

EMG recordings. The EMG activity recorded in the prime mover AD and in BB muscle when the right shoulder is flexed and the index-finger points to the target is shown in Fig. 7 for a representative subject. Note that in both muscles, pre-movement activity in the 1st BLOCK (before 0 ms) was nearly indistinguishable in all sessions, thus pointing errors were not due to changes in the prime mover activation.

For what concerns lower-limb muscles (Fig. 8), when the reaching movement was performed without prisms, so that pointing was accurate, excitatory or inhibitory APAs developed in all muscles, except left-H and SOL (1st BLOCK, BEFORE). The latter two will be then ruled out from analysis, since EMG amplitude at movement onset was not significantly different from the background activity (paired t-test, $P > 0.5$ for both muscles). When reaching was performed immediately after donning prisms and the subject's index-fingertip hit the Plexiglas screen to the right of the target (1st BLOCK, DURING), APAs in Q and TA of both sides increased in amplitude (compare thick to thin black lines). When prisms were removed and subjects missed the target to the left (1st BLOCK, AFTER), APAs in Q and TA of both sides decreased to values similar to those observed in the BEFORE session. In contrast, both when adaptation to prisms completed and when after-effect recovered (2nd BLOCK; Fig. xxxb and xxxb, d), pre-movement EMG activity in AD and BB, as well as APAs in lower-limb muscles, were similar in all experimental sessions.

In summary, when prisms induce a pointing error, the movement seems to be associated to stronger lower-limb APAs, not paralleled by changes in the activation of the prime mover.
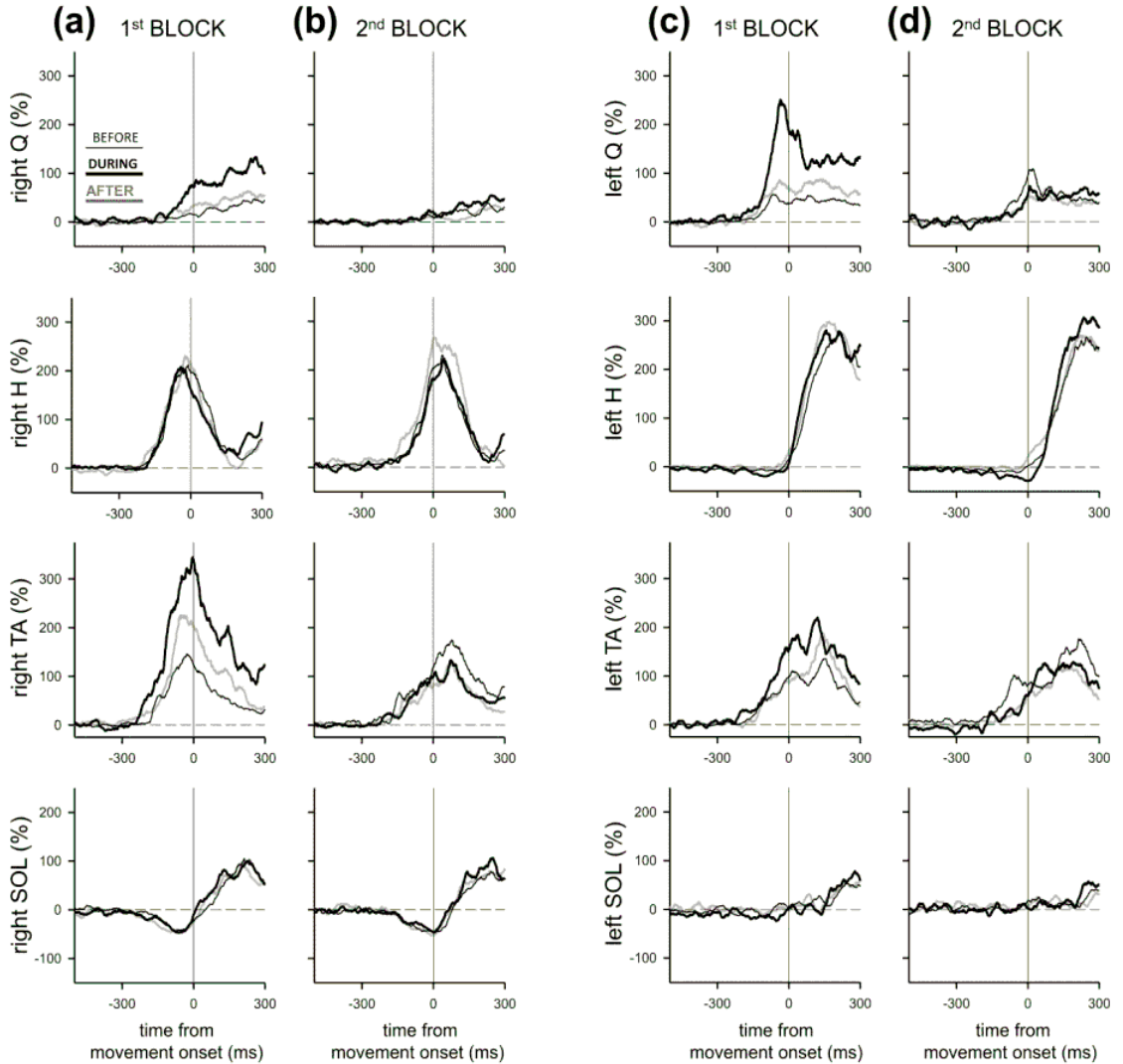
Fig. 8 Rectified and smoothed EMG recordings from the lower-limb muscles Quadriceps (Q), Hamstring (H), Tibialis Anterior (TA) and Soleus (SOL) of the right (a, b) and left (c, d) sides. Averages of the traces of one representative subjects, obtained in trials 1-5 (a, c - 1st BLOCK) and in trials 11-15 (b, d - 2nd BLOCK) of the three experimental sessions: BEFORE (thin black line), DURING (thick black line) and AFTER (thick grey line) wearing prismatic goggles. BEFORE wearing prisms, excitatory or inhibitory APAs developed, prior to the movement onset, in all muscles except left H and SOL. In the 1st BLOCK of the DURING session, APAs in Q and TA of both sides increased, while when prisms were doffed (1st BLOCK of AFTER session) APAs decreased to values similar to those of the BEFORE session. In contrast, in the 2nd BLOCK, when adaptation to prisms and after-effect recovery were completed, APAs in lower-limb muscles were similar in all experimental sessions.

Quantitative analysis of the EMG recordings.

Figure 9a, c shows the mean amplitudes of the pre-movement activation in AD and BB and of the APAs in lower-limb muscles. Two way ANOVAs (prisms x blocks), computed on pre-movement EMG amplitude of the two upper-limb muscles, showed no prisms nor interaction effect, while a significant blocks factor resulted for both AD ($F_{1,9} = 117.13$, P < 0.001) and BB muscles ($F_{1,9} = 52.74$, P < 0.001). Two-way ANOVAs (prisms x blocks) on the right-Q and the right-TA resulted in a not significant prisms main effect, a significant blocks main effect ($F_{1,9} = 5.67$, P < 0.05 and $F_{1,9} = 11.72$, P < 0.01, respectively) and a significant interaction ($F_{2,18} = 3.57$, P < 0.05 and $F_{2,18} = 4.81$, P < 0.05, respectively).
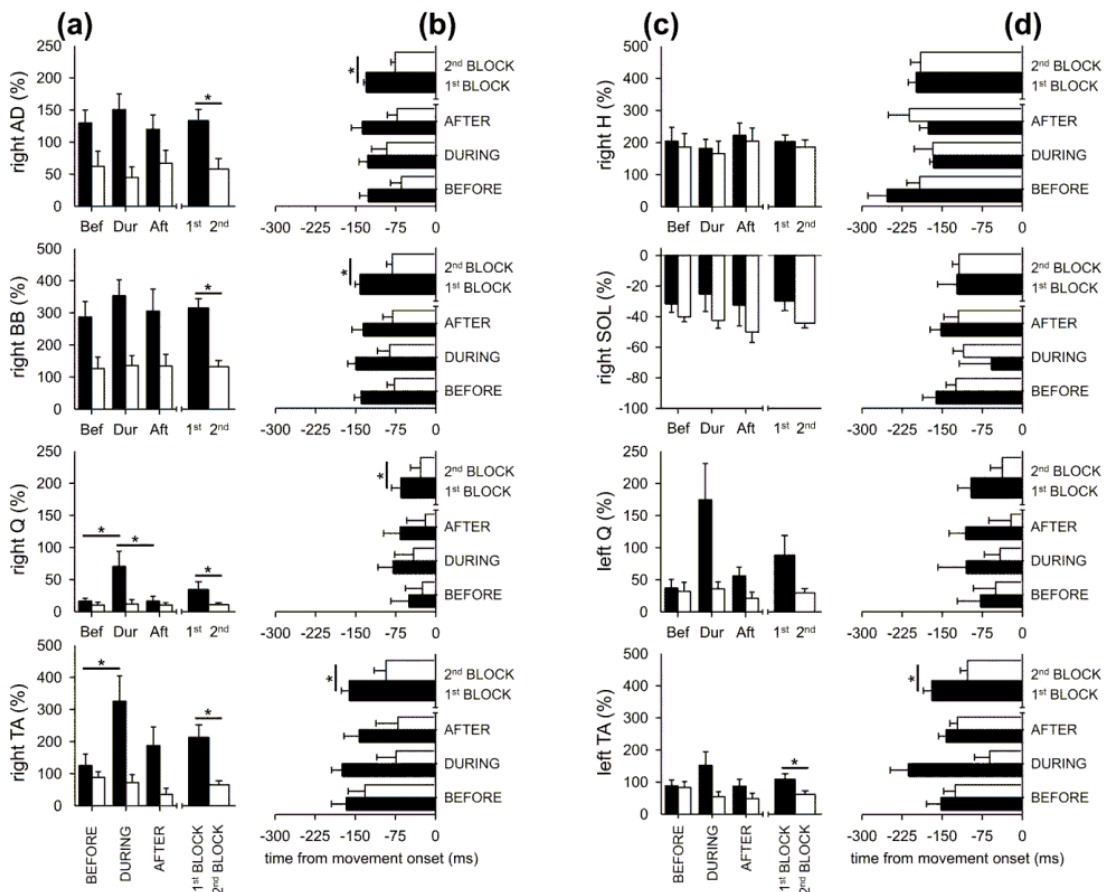

Fig. 9 Mean normalized amplitude (a, c) and mean latency with respect to movement onset (b, d) of pre-movement activation in right arm muscles and of APAs in lower-limb muscles. Plotted values refer to the 1st and 2nd BLOCK (black and white bars, respectively) of each experimental session (BEFORE, DURING and AFTER wearing prismatic goggles). Error bars mark the inter-subject SEM. Significant differences (*prisms x block* ANOVA on each muscle) are marked by asterisks. For each BLOCK, the average of the values recorded among the three sessions is also reported, after the axis break, so as to illustrate the main effect of the ANOVA *block* factor. The APAs increase in Q and TA muscles of both legs observed in the 1st BLOCK of the DURING session reached significance only in the right limb. Moreover, a significant *block* factor was observed in the size and latency of pre-movement activations in AD and BB as well as in the size and latency of APAs in right Q and in right and left TA. Finally, in the 2nd BLOCK no significant effect of prisms was found on size or latency of pre-movement activations or of APAs.

According to the post-hoc comparisons, prisms affected only the 1st BLOCKS, in which right-Q EMG was larger in the DURING than in both BEFORE and AFTER sessions, while right-TA was larger in DURING than in BEFORE session only. ANOVAs on right-H, right-SOL and left-Q showed neither main effects nor interactions, while a significant blocks main effect was found in left-TA ($F_{1,9} = 7.6$, $P < 0.05$). Despite the strong increase in APAs strength in left-Q and left-TA, prisms did not produce significant changes.

Figure 9 summarizes also the latencies of the pre-movement activation and of the APAs. Two-ways ANOVAs (prisms x blocks) resulted in no significant prisms main effect nor interaction, while blocks factor was significant in AD, BB, right-Q, right-TA and left-TA (in all cases, $F_{1,9} > 7$, $P < 0.05$), i.e. those muscles that showed blocks main effect on the EMG amplitude.
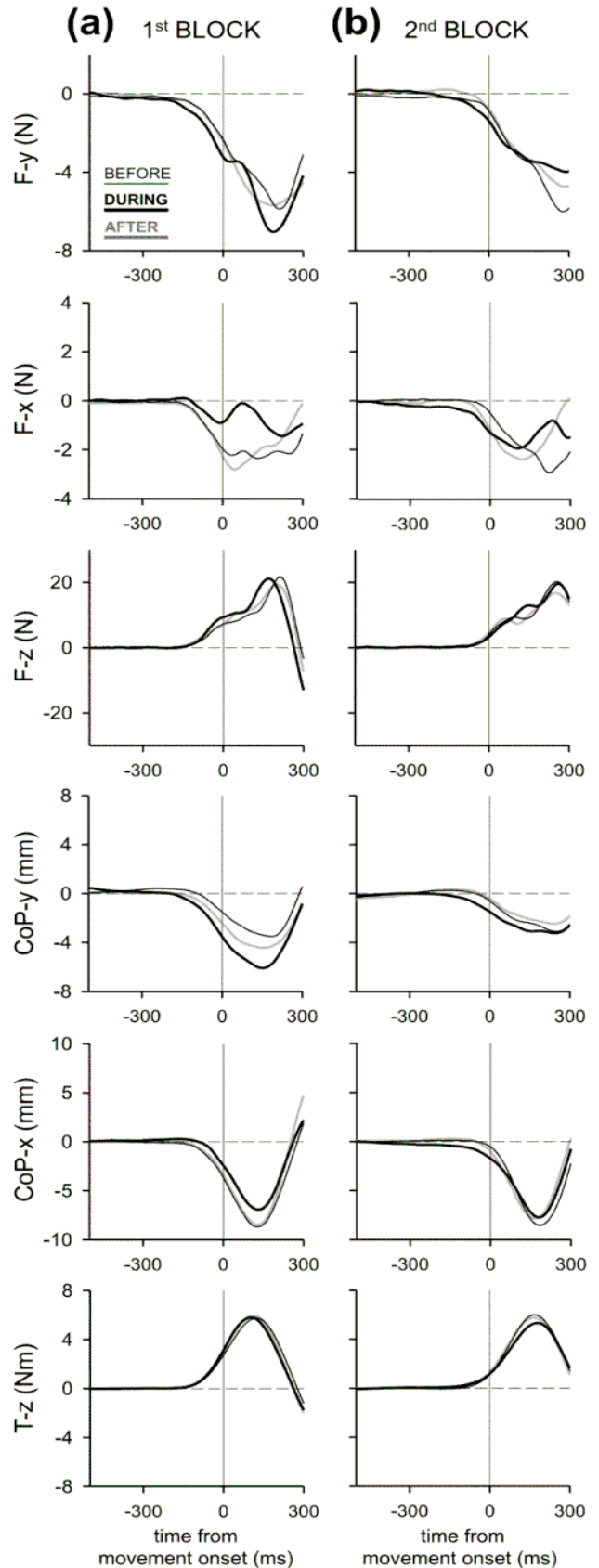
Platform recordings.

Before wearing prisms, platform recordings changed prior to the movement onset, when all of them significantly differed from the corresponding background level (paired t-test, P always < 0.05). As shown in Fig. 10a - 1st BLOCK, the force vector pointed backward, rightward, and downward; the CoP moved backward and rightward, and the vertical torque turned clockwise. The largest prisms effect occurred in the APA revealed by the CoPy (antero-posterior) displacement. When prisms were just donned (Fig. 10a - 1st BLOCK), the anticipatory displacement increased, while after doffing them it reduced in size. When adaptation to prisms and after-effect recovery were completed (Fig. 10b - 2nd BLOCK), the APA size in CoPy was again comparable among the three sessions.

Quantitative analysis of platform recordings.

Figure 11a shows the mean amplitudes of the APAs in platform recordings. Two-way ANOVA (prisms x blocks) on APA amplitude in CoPy resulted in a not significant prisms main effect, a significant block main effect ($F_{1,9} = 4.93$, $P < 0.05$) and a significant interaction ($F_{2,18} = 13.19$, $P < 0.001$). According to the post-hoc comparisons, prisms affected only the 1st BLOCK, in which the anticipatory displacement was larger in the DURING than in both BEFORE and AFTER sessions. Two-way ANOVAs on the remaining recordings highlighted only a significant block main effect in all traces ($F_{1,9} > 10$, $P < 0.01$). The same ANOVA design on APAs latencies (Fig. 11b) showed no prisms main effect nor interaction, but a significant block main effect in Fy, Fz and CoPy ($F_{1,9} > 5$, $P < 0.05$).

Fig. 10 Force platform recordings: components of the force exerted on the ground along the three Cartesian axes (Fy, positive when directed forward; Fx, positive leftward; Fz, positive downward), displacement of the Centre of Pressure (CoPy, positive forward; CoPx, positive leftward) and torque about the body vertical axis passing through the CoP (Tz, positive clockwise). Averaged traces of one representative subjects, obtained in trials 1-5 (a - 1st BLOCK) and in trials 11-15 (b - 2nd BLOCK) of the three experimental sessions: BEFORE (thin black line), DURING (thick black line) and AFTER (thick grey line) wearing prismatic goggles. BEFORE wearing prisms, all platform recordings changed prior to the movement onset, thus showing APAs. Prismatic goggles induced the largest change in the APA of CoPy, which increased when prisms were just donned (1st BLOCK, DURING) and reduced in size after doffing them (1st BLOCK, AFTER). When adaptation to prisms and after-effect recovery were completed (2nd BLOCK), the size of APA in CoPy was again comparable among the three sessions.
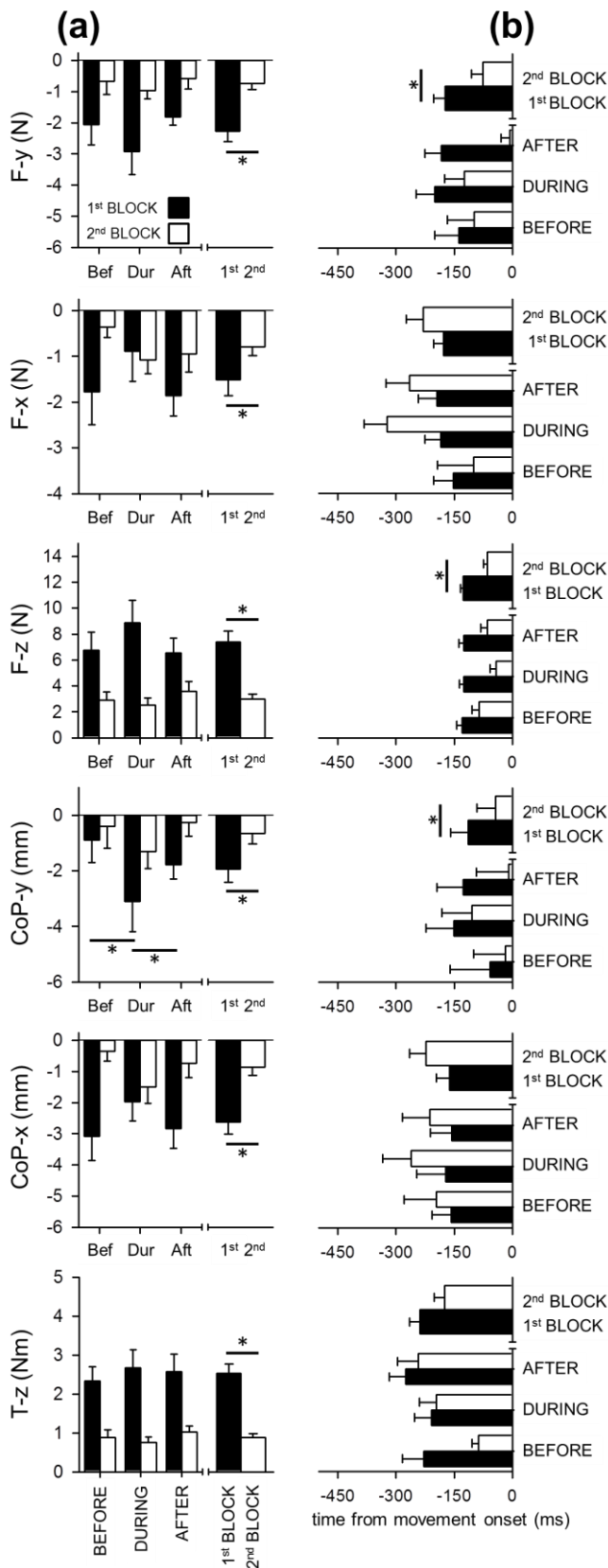
Fig. 11 Mean amplitude (a) and mean latency with respect to movement onset (b) of APAs in the force platform recordings. Plotted values refer to the 1st and 2nd BLOCK (black and white bars, respectively) of each experimental session (BEFORE, DURING and AFTER wearing prismatic goggles). Error bars mark the inter-subject SEM. Significant differences (prisms x block ANOVA on each force platform recording) are marked by asterisks. For each BLOCK, the average of the values recorded among the three sessions is also reported, after the axis break, so as to illustrate the main effect of the ANOVA block factor. The CoPy APA increase observed in the 1st BLOCK of the DURING session reached significance. Moreover, a significant block effect was observed in the size of APAs in all platform variables and also in the latency of APAs in Fy, Fz and CoPy. Finally, in the 2nd BLOCK no significant effect of prismatic goggles was found on size or latency of APAs.

Control analysis - Duration of the target reaching movements

Since it is widely reported that APAs are scaled in amplitude to movement duration (faster movements are associated to larger APAs, Shiratori & Aruin 2007, Lee et al. 1987) we carefully verified if this parameter changed during the experiment. Figure xxxx shows the mean duration of the target reaching movement in the two blocks of trials, collected in the three experimental sessions. Two-way repeated measures ANOVA (prisms x blocks) highlighted only a significant blocks main effect ($F_{1,9} = 63.45$, $P < 0.001$). Thus, within each block, movement duration was similar in the three sessions, indicating that the APAs changes observed in the EMG and platform recording, within the 1st BLOCK, should not be ascribed to changes in movement velocity. On the other hand, all the APAs changes between the two blocks (1st BLOCK vs. 2nd BLOCK comparisons) may be affected as well by the increase in movement duration observed in the second block.
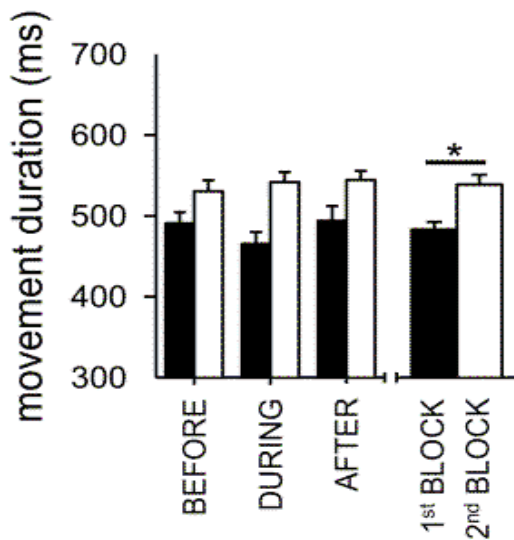

Fig. 12 Mean duration of the target reaching movement. Plotted values refer to the 1st and 2nd BLOCK (black and white bars, respectively) of each experimental session (BEFORE, DURING and AFTER wearing prismatic goggles). Error bars mark the inter-subject SEM. Significant differences (*prisms* x *block* ANOVA) are marked by asterisks. For each BLOCK, the average of the values recorded among the three sessions is reported, after the axis break, so as to illustrate the main effect of the ANOVA *block* factor. Note that the only significant difference was an increase in movement duration in the 2nd *vs.* 1st BLOCK, while prisms had no effect in either BLOCK.

Experiment 2

APAs prior to index-finger flexion with the hand prone

Figure 13 shows the pattern of APAs, observed in a representative subject, when an index-finger tap is performed. The activation of the prime mover Flexor Digitorum Superficialis (FDS) was constantly preceded by APAs at the elbow level: a clear inhibition of Biceps Brachii (BB) EMG was mirrored by a facilitation of Triceps Brachii (TB). These two effects were similar for both timing and (normalized) amplitude. An important anticipatory reduction in the EMG activity could also be observed in the more proximal Anterior Deltoid (AD) muscle.

Thus, the reciprocal modulation at the elbow and the inhibitory effect at the shoulder would in fact counteract both the arm flexion and the scapular elevation which is developed when the index-finger is flexed, so that the full APA chain stabilises the more distal joints (Caronni and Cavallari 2009a).

APAs after 12 hours immobilization: EMG recordings

Figure 13 reports also the comparison of the rectified and integrated EMG traces recorded before (PRE) and after (POST) immobilization. It is apparent that in both conditions, activation of the FDS muscle, which starts about 14 ms before the index-finger flexion, is clearly preceded (by about 30 ms) by a major postural inhibitory activity in BB and AD muscles, which is almost synchronous to an excitatory postural adjustment in TB muscle. Note however that, in the time window between APAs and movement onsets, the inhibition of BB and AD is increased after immobilization, while the activity remains unchanged in FDS; on the other hand, TB excitation is reduced. These data testimony that a short-term immobilization of the distal joints interferes with the motor program that generates the proximal components of the postural control.
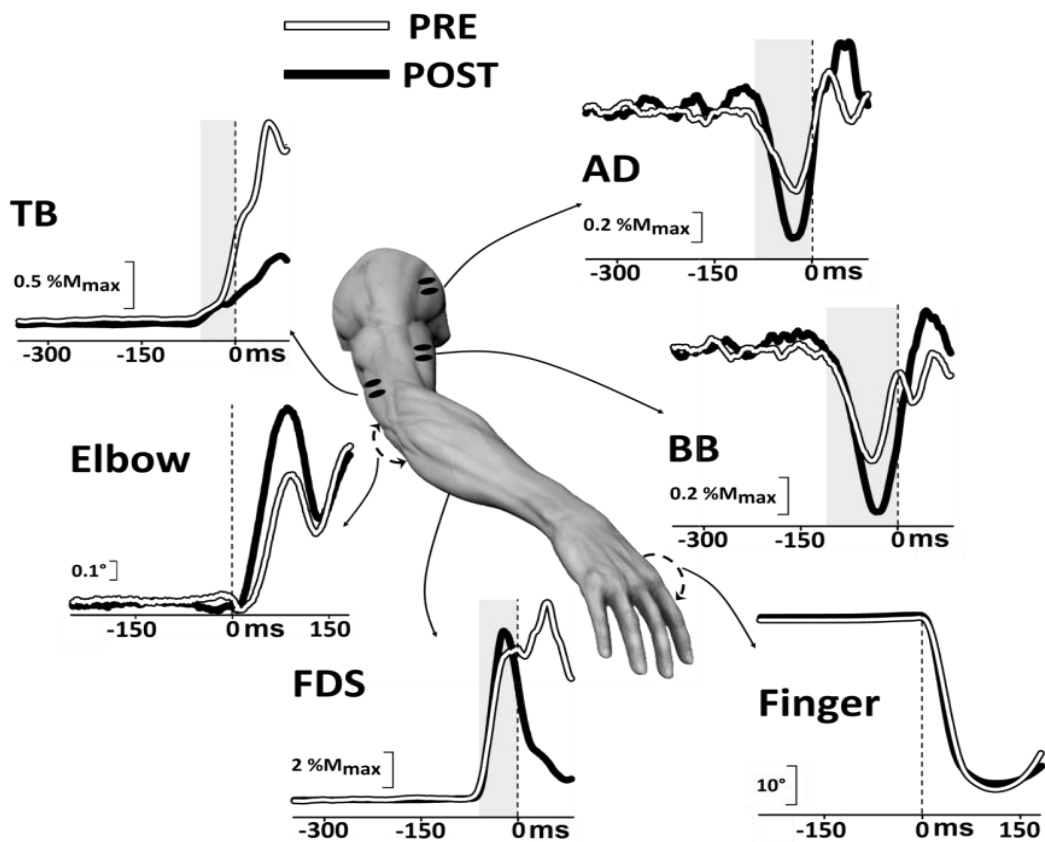
Fig. 13 Intra-limb APAs change after short-term immobilization. Average recordings in a representative subject before (PRE, white) and after (POST, black) immobilization. When rapidly flexing the index-finger (Flexor Digitorum Superficialis, FDS, prime mover), the arm equilibrium is preserved thanks to APAs (shaded area) which are excitatory in Triceps Brachii (TB) and inhibitory in Biceps Brachii (BB) and Anterior Deltoid (AD). After 12h immobilization, FDS activation preceding the movement onset and index-finger movement was unchanged. Instead, inhibitory APAs in BB and AD apparently increased, while excitatory APA in TB was marginally decreased. The changes in the postural chain lead to a less effective fixation of the elbow joint, which showed a larger displacement during index-finger acceleration.

The effect of immobilization has been regularly observed, although to varying degree, in all ten subjects. Figure 14B shows the mean amplitude of the APAs, expressed as a percentage of the average value of the maximum motor evoked potential (M_{max}) recorded in the two sessions. Figure 14A shows that the average level of FDS muscle activation does not reveal significant changes ($t_9 = 1.46$, $P = 0.18$), before and after immobilization. The average inhibitory effects on BB and AD and excitation on TB are instead compared on the right. A two way, muscle x session ANOVA with repeated measures showed a significant main effect for both factors (muscle: $F_{2,18} = 18.65$, $P < 0.0001$; session: $F_{1,9} = 14.73$, $P = 0.004$) and the absence of any interaction ($F_{2,18} = 0.23$, $P = 0.79$). Thus, after immobilisation of the wrist, inhibition of BB and AD significantly increased (+19.4% and +30.3% respectively) and excitation of TB significantly decreased (-36.0%). The only significant Tukey post-hoc tests are those comparing excitation in TB vs. inhibition in BB and AD. Pooled within-PRE and within-POST session variability (in %M_{max}) was: 1.64 and 1.40 in FDS, 0.23 and 0.25 in BB, 0.26 and 0.16 in TB, 0.26 and 0.29 in AD, respectively.
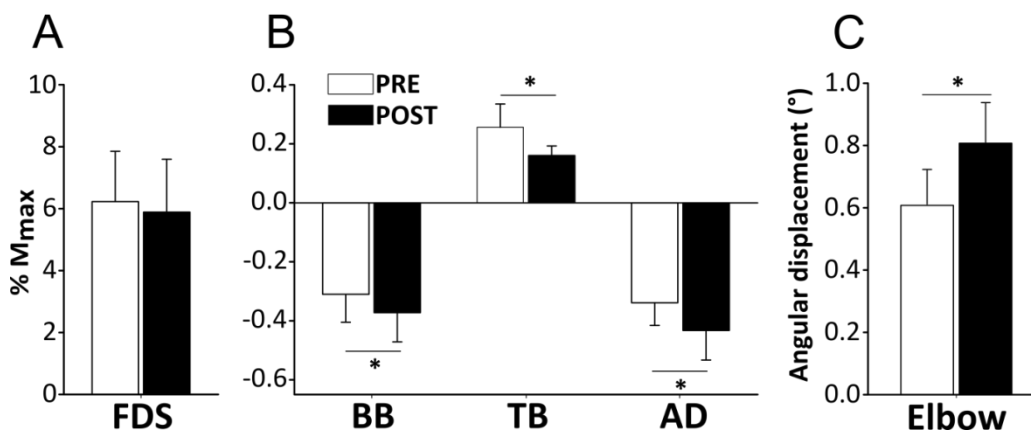

Fig. 14 Amplitude of FDS voluntary activation, of APAs in BB, TB and AD and of elbow displacement, before (PRE, white) and after (POST, black) immobilization. Mean values ± SEM. Paired t-tests found no PRE *vs.* POST difference in FDS activation, but a significant increase of APAs in BB and AD. The change in TB did not reach significance. APAs changes lead to a significant increase of elbow displacement.
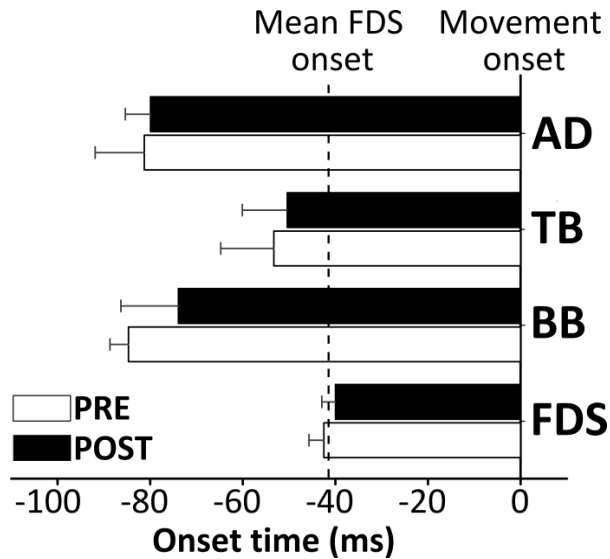
Fig. 15 Latency of FDS, TB. AD and BB activation before (PRE, white) and after (POST, black) immobilization. Mean values ± SEM.

The time-course of the effects was similar in the two experimental sessions. Figure xxx show the average latency of the APA onset in the individual muscles: note that EMG activation in the prime mover is constantly preceded by the APA in BB, TB and AD muscles.

APAs after 12 hours immobilization: goniometric recordings

The clear changes observed in the APAs in response to immobilization are evidently expected to cause an altered fixation of the proximal joints, leading to a less accurate execution of the finger flexion. The kinematics consequences induced by immobilization, were easily highlighted by comparing the angular excursion of the elbow recorded in two experimental sessions. Figure 14C shows that, after removing the plaster, the amplitude of elbow excursion was significantly higher (0.628 ± 0.11 ° versus 0.798 ± 0.12 °; $t_9 = 2.55$, P =0.031).

Control measurements

TABLE

PRE	POST	t-test P	
Finger movement amplitude (°)	46.6 ± 3.4	45.6 ± 3.2	0.38
Finger movement duration (ms)	129.4 ± 15.0	124.4 ± 15.0	0.22
M_{max} in FDS (mV)	1.8 ± 0.3	1.7 ± 0.3	0.62
M_{max} in BB (mV)	3.8 ± 0.9	3.2 ± 0,6	0.49
M_{max} in TB (mV)	2.5 ± 0.4	2.6 ± 0,4	0.58
M_{max} in AD (mV)	2.3 ± 0.3	2.1 ± 0.3	0.29
Background EMG level in BB (%M_{max})	0.9 ± 0.2	1.0 ± 0.3	0.40
Background EMG level in AD (%M_{max})	1.0 ± 0.3	1.1 ± 0.3	0.33

Table 1 reports the control measurements concerning the substantial stability of the amplitude and duration of the index-finger flexion; the amplitude of the M_{max} waves and the level of EMG activity in the muscle that receive inhibition. For all parameters statistics showed no difference between the PRE- and the POST-immobilization values. This will exclude changes in the kinematics parameters of the movement, in the interface between electrodes, skin and muscles and in the level o background motoneurones activity recruited in the two situations.

Experiment 3

Figure 16A showed the timing of voluntary FCR activation and BB postural activation (mean ± SE), scaled with respect to wrist flexion onset (0ms). Wrist flexion and FCR activation are both preceded by an excitatory postural activity in elbow flexor and the BB activation antici pate the activation of the FCR 20ms circa.

 Figure xxxB showed the Integrated EMG traces recorded from the prime mover and the postural muscles (single subjects). The BB postural activity, in this experiment, is excitatory since the mechanical perturbation caused by the voluntary movement is directed toward the opposite direction respect with what we observed in the previous experiment.
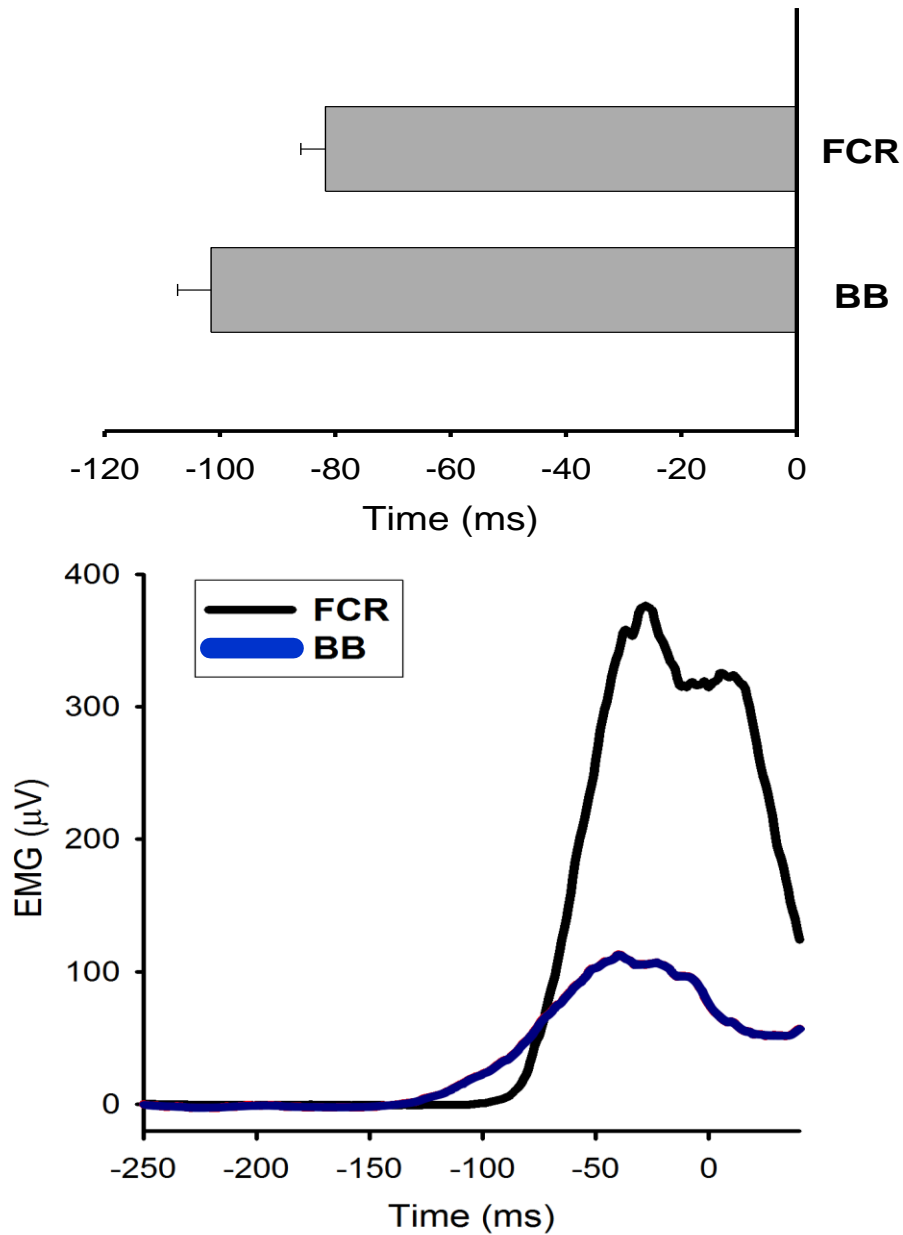
Fig. 16 A) Timing of voluntary FCR activation and BB postural activation (mean ± SE), scaled with respect to wrist flexion onset (0ms). B) Integrated EMG recorded from the prime mover and the postural muscles. Wrist flexion and FCR activation are both preceded by an excitatory postural activity in elbow flexors. Data from a single subject

In Figure 17 is shown the time course of FCR and BB MEPs during observation of wrist flexion. The illustrated trend represent the modulation of the MEPs amplitude in FCR and

BB tested at different intervals from the onset of wrist flexion (0ms) observed on the screen. The 100% value (blue-baseline) refers to the mean MEPs amplitude recorded at -220ms, when nor postural neither voluntary activity is expected.

In FCR, the population MEPs are significantly greater than the baseline at 0, 100, 160ms, while for BB they reach statistical significance at -30 and 0ms intervals. Both FCR and BB excitability results modulated before the onset of the observed movement. Moreover, interestingly, the excitability in BB grow up one interval before that of FCR.
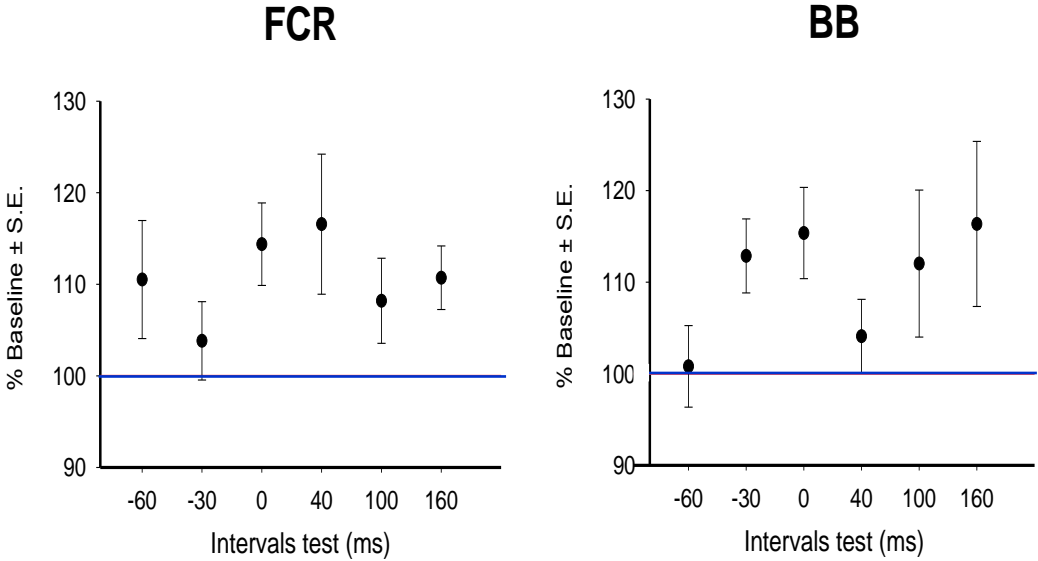

Fig. 17 Time course of FCR and BB MEPs during observation of wrist flexion

DISCUSSION

Experiment 1

The novelty of our study is that by using prisms we induced out-of-target movements which were found not to be associated to changes in the prime mover activation, as it might be expected, but only to changes in the APAs size.

In the following, we will first deal with the origin of the pointing error, then we will recall that accurate motion of a segment (e.g. the hand) require a proper coordination between distal (e.g. the upper-limb) and proximal (e.g. the trunk) body parts.

Origin of pointing error

Performing a goal-directed movement classically implies a "strategic" phase (what is my goal), a "tactic" phase (what I should do) and an "executive" phase (the ensuing movement). When a healthy subject points to the target cross, strategy, tactics and execution are perfectly tuned each other so as to achieve the goal, i.e., to reach the cross.

When wearing prisms, the goal is still to reach the cross, but the motor plan is biased by an illusory deviation of the target image, due to the shift of the binocular eye field. The ensuing movement, which is shaped by the new motor plan, now ends out-of-target; during the following adaptation to prisms, an iterative retuning of tactics on strategy occurs. In these conditions, the pointing error is the expression of the mismatch between strategy and tactics.

Pointing to a visually displayed target requires a neural transformation from a visual representation of target location to an appropriate pattern of arm muscles activity. According to Soechting and Flanders (1989a, b; see also 1992 for a review), errors in such a movement derive from errors in the sensorimotor transformation from the visual representation of the target to the kinematics representation of the planned trajectory. Indeed, such transformation is intrinsically non-linear, but subjects usually employ a linear approximation when they have to remember the target location and point to it. These Authors also showed that when subjects have to reach a position which has been previously appreciated kinaesthetically (thus after having empirically built up the exact transformation), pointing errors dramatically reduce. Several evidences were collected that the parietal cortex plays a critical role in integrating visual and somatic inputs for building up this sensorimotor transformation (see Kalaska et al. 1997 for a review).

In our study, pointing errors were apparently due to the changes in the sensorimotor transformation induced by prisms. In a few trials, thanks to the visual feedback, our subjects empirically solved the new sensorimotor transformation and succeeded in reaching the target. The same occurred (in the opposite direction) after doffing goggles. An increase in pointing error when requiring to change the sensorimotor transformation, for instance by asking to reach a target in an horizontal workspace while looking at the initial position of the hand and target on a vertical screen, was also observed by Messier and Kalaska (1997).

By extending the "strategy-tactics-execution" framework to pathology, it may be speculated that an ataxic patient, which regularly fails in reaching the target (wrong execution), is still able to identify the goal (good strategy) but pays for a mismatch between strategy and tactics, as well as between tactics and execution (see, for a review, Manto 2009).

Coordination between proximal and distal body segments

More and more papers show that the performance of dexterous motor tasks, such as pointing and reaching, relies on the exact coordination between proximal (e.g. trunk) and distal body segments (e.g. hand).

The crudest set of evidences comes from pathology. For example, Hsieh and colleagues (2002) have shown that trunk control at an early stage after stoke was one of the strongest predictors of upper-limb functional recovery in the Activities of Daily Living test. Similarly, Stoykov and collaborators (2005) reported that speed and movement accuracy of the ataxic upper-limb improved when a subject, who suffered brainstem stroke, was specifically trained to improve sitting balance and trunk control.

Several motor control studies (Ma & Feldman 1995, Archambault et al. 1999, Pigeon et al. 2000, Robertson & Roby-Brami 2011) give also evidence that motion of the trunk and the upper-limb are appropriately scaled each other to ensure the maximal accuracy when moving the hand toward a target. In their seminal paper, Hollerbach and Flash (1982) offer a model, which describes shoulder-elbow coordination in hand reaching movements. The same model also predicts the modifications of the distal segments trajectory and its final position when the force exerted at proximal joints is inadequate, a condition in which an out-of-target movement would result. Similarly, out-of-target movements are also expected when the Coriolis force acting on the arm during the simultaneous displacement of the upper-limb and torso is not compensated in a feed-forward manner (Bortolami et al. 2008).

Interestingly, Era and co-workers (1996) have reported that top-level rifle shooters stabilize their whole body balance better than naive shooters, particularly in the last seconds before the shot. This and other studies investigating the same topic (e.g. Aalto et al. 1990; Mononen et al. 2007) give evidence that shooting accuracy relies on the accurate trunk and lower-limb posture control, allowing coordination of these body segments with the focal trigger pull.

On these premises, APAs may represent the earliest part of the motor command necessary for proximal and distal body segments coordination, as also supported by our results.

APAs contribution to movement accuracy

Only in recent years, some Authors suggested that APAs function is not limited to ensure the whole-body balance, but might also encompass the ability to provide the most appropriate conditions to guarantee an accurate movement execution.

APAs decrease in size as the accuracy demand increases (i.e. when pointing smaller and smaller targets), a feature which has been shown both in the upper-limb (Bonnetblanc et al. 2004) and in the lower-limb (Bertucco & Cesari 2010). Lower limb pointing was also investigated by Duarte and Latash (2007), who have shown that the faster the movement is, the larger the APAs variability is. It is also well described the relation between movement speed and scattering of the final position around a target (Fernandez & Bootsma 2004, Schmidt et al. 1979). In the other way round, all these observations suggest that small and less variable APAs should accompany slow, but precise movements. Finally, Berrigan and colleagues (2006) reported that when pointing is performed towards small targets (i.e. under high accuracy constraints) from an "unstable" position (i.e. standing vs. sitting), slowing movement speed actually represent a strategy to reduce the equilibrium disturbance i.e. the associate APAs.

Thus, when the accuracy demand increases, peak movement velocity decreases, as already described by the Fitts' law (1954). However, since APAs amplitude is known to be proportional to focal movement velocity (Shiratori & Aruin 2007, Lee et al. 1987), its reduction might be not directly related to the increased accuracy demand, but to the associated reduction in movement speed. With respect to the above studies, our work is novel because it proves the relationship between APAs and movement accuracy (i.e. the pointing error), rather than the accuracy constraint (i.e. the target size), by ruling out the

effect of movement velocity. The latter was indeed constant among sessions thanks to the fact that prismatic lenses did not affect the target size.

Further considerations on present results

As pointed out above, a crucial aspect in APAs modulation is movement velocity. However, our experiments show that when movement velocity remains constant among sessions (i.e. within each BLOCK) prism-induced pointing errors are still associated to changes in APA amplitude, thus supporting a linkage between APAs and movement accuracy.

When moving fast, stronger prime mover activation is associated to stronger (Shiratori & Aruin 2007, Lee et al. 1987), and more anticipated APAs (Zattara & Bouisset 1988, Horak et al. 1984). This is in agreement with our results which show that APAs and prime mover activation in the 1st BLOCK were larger and started earlier than those recorded in the 2nd BLOCK, when movements were slower.

Intriguingly, the relationship between movement speed / prime mover contraction and APAs changes was limited to Q and TA of both sides (although not significant in left-Q), with no modifications in H and SOL, a pattern which suggests that different muscles of the APAs chain could have different roles in equilibrium stabilisation. APAs are known to secure the equilibrium in a twofold way (Massion 1992): i) by counterbalancing the segmental equilibrium disturbance due to the reaction forces developing with the ongoing movement, (which grow when movement speed increases) and ii) by preventing the whole body CoM displacement produced by the new configuration of the body (regardless the speed with which that configuration has been reached). Considering that 1st and 2nd BLOCK trials have different movement speed, but similar movement amplitude on the sagittal plane, it can be proposed that anterior lower-limb muscles (Q and TA) may neutralize the time-changing perturbation due to prime mover contraction, while posterior muscles (H and SOL) may neutralize the CoM changes as the movement develops.

Prisms selectively modulate those APAs sensitive to modification in movement speed. When wearing prisms, APAs amplitude was significantly increased in right Q and TA muscles (1st BLOCK – DURING vs. 1st BLOCK – BEFORE); no changes were instead found in APA latency. Thus APAs timing and amplitude seems to be independently controllable, as also suggested by Nana-Ibrahim and colleagues (2008).

Finally, it is worth noting that the invariance of the shoulder voluntary movement in both vertical and horizontal directions is supported by the observed invariance of the AD and BB activity. Indeed, AD is known to play a double role of shoulder flexor and abductor, and BB

exerts the same two actions on that joint (see Kapandji 1982). Thus, any horizontal or vertical change in the upper limb movement should have been accompanied by a change in AD and BB recruitment. Since none of such modifications occurred (see Fig. 2) we are confident to propose that movement in shoulder and elbow joint angles was unmodified. The change in absolute fingertip trajectory when committing pointing errors should then be mainly ascribed to a torso rotation, as revealed by the observed increase in right Q and TA APAs.

Experiment 2

To the best of our knowledge this is the first experiment that shows that a short term immobilization interferes with the control of the postural muscles. In the past Moisello et al. (2008) have demonstrated that short term immobilization effectively modifies the focal movement concerning the sole kinematic variables. Our results, instead, do not show a modification to the focal movement kinematics (controlled with a goniometer), simply because subjects were explicitly asked to perform finger flexion in a way that the movement were mechanically comparable before and after immobilization.

After wrist immobilization, inhibitory APA in BB and AD muscles were increased, while facilitation of the TB muscle was decreased; this was also paralleled by an increase of the elbow excursion. The data presented here are in agreement with those reported by Moisello et al. (2008) in which immobilization was shown to produce changes in the coordination of adjacent joints leading to a decline in the accuracy of the trajectory, and ultimately to a scarce precision of the voluntary movement, similar to that already described in patients with proprioceptive de-afferentation (Ghez et al. 1995; Gordon et al. 1995; Sainburg et al. 1995). However our results disclosed that the trajectory impairment has to be attributed to focal as well as postural components of the movement. Few days of immobilization seem to decrease the somato-sensory cortex excitability (Facchini et al. 2002), but according to Huber et al. (2006) even 12 hours of immobilization were already effective in decreasing the amplitude of both the somato-sensory and motor evoked potentials, suggesting that a synaptic depression has developed in the cortex. According to Huber, the short-term immobilization lessens the amplitude of the P45-wave in median somato-sensory evoked potentials, which represents the processing of the proprioceptive information in the sensory-motor areas (Allison et al. 1992). Moreover, the P45 reduction was closely related with the inaccuracy of voluntary movement. As a whole, these authors suggested that short periods of immobilization trigger a synaptic depression upon the

primary sensory-motor cortical representations of the immobilized segment. We propose that in a situation of synaptic depression of the above sited areas, to obtain a quantitatively similar voluntary movement before and after immobilization (as we got in the present study) an increase in the central voluntary command is needed.

Since APAs are scaled to prime mover activation (Aruin and Shiratori 2004), a parallel change should then occur in APAs too, but this would lead to a significant increase of both excitatory APAs in TB and inhibitory APAs in BB. The unbalanced effect showed in the antagonistic muscles BB and TB, suggest, in agreement with the results of Facchini et al. (2002), that immobilization triggers a tonic enhancement of inhibitory drive involving not only the cortical representation of the immobilized joint but also the adjacent areas. In BB, in which a tonic voluntary drive maintains the elbow at the requested 90°, the tonic enhancement of inhibition that might decrease the EMG background level and affect the elbow angle is instead compensated by a comparable increase of the tonic voluntary drive. In this condition, the increase in central commands for the focal movement and for the associated APAs (required for overcoming the depression of the FDS representation) would simply lead to an increased inhibitory APA in BB. On the other side in TB, which is at rest before the movement, the enhancement of inhibition would not affect the background EMG level but would offset the increase in the central commands, thus affecting the excitatory APA. This tonic enhancement of inhibition may stem from an increased intracortical inhibition, as observed after immobilization by Clark et al. (2010), but may as well come from an increased cerebellar activity.

Our results deserve some further brief comments. First, the reciprocal effect on biceps (more inhibition) and triceps (less excitation) may be due to the general increase of descending inhibition distributing upon the two motoneuronal pools, but may as well as be the expression of a change in spinal reciprocal inhibitory circuits (Katz et al. 1991), which are known to be fed by cortico-spinal projections (Cavallari et al. 1984; Kudina et al. 1993). However, the first hypothesis seems to be preferred, taking into account that AD activity is also more inhibited and spinal reflexes seems unmodulated during the whole period of motor preparation (Caronni and Cavallari 2009b). Second, a series of control measurements allows us to exclude that other factors may have influenced our results. In particular, the constancy of M_{max} in the PRE and POST sessions revealed a substantial stability of the recording apparatus all along the 12 our period, as observed also by Facchini et al. (2002) after 3 days of immobilization. Moreover, since movement of amplitude and duration may affect the precision of the movement itself as well as the characteristics of APAs (Lee et al. 1987; Aruin and Shiratori 2004) it has also been also

verified that the kinematics of index-finger flexion did not change in the two experimental conditions. Last, the level of the background EMG activity in BB and AD was found to be analogous in the two sessions, the sensitivity of the pool to facilitation or to inhibition being thus comparable in the two situations (Crone et al. 1990).

Seki et al. (2001a; 2001b) reported that after immobilization the properties of the muscle underwent changes in the ratio between fast and slow muscle fibres, but these changes took place only after 3-4 weeks. In animal studies, researchers have also shown that 12 hours of decreased muscle activity produced some disturbances in intracellular levels of messenger RNAs for members of the aspects of metabolism and muscle structure, but these changes needed several weeks to produce changes at the level of the muscle fibber (Bey et al. 2003). The changes in motor performance found in our study occur, instead, too early to be related to changes in muscle structure.

Experiment 3

In the last experiment we described a new property of the phenomenon of the "motor resonance", in fact during movement observation not only the cortical area of the prime mover is subject to a rise in excitability but this happen also for the postural muscles.

We know that the motor resonance is mediated via the abundant connections (Cerri, Shimazu, Maier, & Lemon, 2003; Geyer, Matelli, Luppino, & Zilles, 2000; Shimazu, Maier, Cerri, Kirkwood, Lemon, 2004) between the premotor and the primary motor cortex, M1 results activated during both actual and observed movements   (Baldissera, Cavallari, Craighero, & Fadiga, 2001; Borroni et al., 2005; Fadiga, Fogassi, Pavesi, & Rizzolatti, 1995; Montagna et al., 2005) and we also know that the APAs (at least the inhibitory ones) are mediated by M1 circuitries (Caronni & Cavallari 20009b).

Here we wanted verify if motor resonance affect only on the prime mover circuitry or even on the postural circuitry. In the first case it would mean that the rise in excitability due to the "motor resonance" is strictly dependent on the movement of a particular body segment and that affect only the prime mover muscle, in the second case it would mean that the motor resonance "hit" also the postural muscles.

Proving that APAs "resonate"would sustain the hypothesis that mirror neurons encode movements, not actions despite what suggested in some studies.

Rizzolatti showed that   when observing hand grasping some mirror neurons in the monkey F5 area fire during both finger flexion or extension (Rizzolatti et al., 1988). They interpreted this finding as a proof that the motor resonance phenomenon is not linked to a specific movement but to the whole action. This point of view did not consider that the way in which

a motor act is organized is extremely complex and that every movements (even the smaller, see Caronni & Cavallari 2009a) is associated to activation of fixative muscles.

Since the feedforward postural control is an essential components of the voluntary movement we wanted verify if the motor resonance responds to the observation by activating the same motor program that the subject would need in order to perform the same movement.

In this light, the present finding that motor resonance involves postural muscles suggests that the 'mirror imitation' is a subliminal replica not only of the explicit actions on which observation is focused but of the whole activation pattern utilized during execution, including not directly observable muscular synergies and anticipatory postural adjustments occurring in other limbs.

CONCLUSION

The results of the first experiment reinforce the hypothesis that a successful on-target pointing movement relies upon a specific tuning between APAs and prime mover activation, as that obtained at the end of the adaptation phase.

The most important result of the second experiment is that, although the prime mover activation remains unchanged after the immobilization, the trajectory described by the index finger is most likely changed between the two sessions due to the modification in the postural control that led to a less effective stabilization of the proximal joint, as was suggested by the mechanical model designed by Caronni and Cavallari (2009a).

In the last experiment we demonstrated that the resonant response in resting subjects replicates, under threshold, both the primary movement and postural activity. The precocious increase in excitability observed in BB may be the expression of the anticipatory activation observed during the execution of the movement. Given that MR reflects aspects that are intrinsic to motor programming also this result strongly support the idea that primary movement and the postural command are essential components of the same neural process.

The APAs give an important contribution to the organization of a complex and accurate movement, they are affected by immobilization giving an "impaired" motr output they are involved in the activation of motor pathways induced by movement observation.

In conclusion, differently from other papers (Brown and Frank 1987), our works strengthens the argument that APAs, are organized similarly to voluntary movement.

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