ELSEVIER

Neuroscience Letters 310 (2001) 183–187

Neuroscience Letters

www.elsevier.com/locate/neulet

Quantitative evidence for multiple widespread representations of individual muscles in the cat motor cortex

Cyril Schneider[a], Daniel Zytnicki[b], Charles Capaday[a,*]

[a]Department of Anatomy and Physiology, Université Laval, Brain and Movement Laboratory, CRULRG, Hôpital Robert Giffard, 2601 de la Canardière, Québec City G1J 2G3, Canada

[b]Neurophysique et Physiologie du Système Moteur, CNRS-FRE 2361, Université René Descartes, Paris, France

Received 9 April 2001; received in revised form 15 June 2001; accepted 12 July 2001

Abstract

We sought to determine why a given muscle appears represented in widespread loci in the motor cortex (MCx). To this end, we microstimulated every 500 μm along medio-lateral rows and recorded the evoked electromyographic (EMG) responses of up to a dozen forelimb muscles of the cat. A consistent finding in all animals studied was that along a given row, distal muscle responses could be elicited from medially situated cortical loci and conversely, proximal muscles responses from laterally situated cortical loci. In many such cases, the evoked EMG responses were such that the largest responses from a distal muscle were obtained by stimulation at a medially situated point and those of a proximal muscle from a laterally situated point. A Spearman correlation analysis showed that there was no correlation between cortical position and where the peak response of a given muscle occurred. These quantitative results strongly support the view that in the forelimb area of the cat MCx there exists widespread 'colonies' of corticospinal neurons with common spinal cord targets.

Keywords: Motor cortex map; Microstimulation; Corticospinal neurons; Evoked motor potentials

In the classic organizational scheme of the motor cortex [15,19] each limb joint (e.g. wrist) is spatially segregated from nearby limb joints (e.g. the elbow). However, the more modern view based on microstimulation results is that single muscles, or joints, are represented many times over [1]. The sites from which a given muscle can be recruited appear non-contiguous and intermingled with sites that recruit other muscles, acting either at the same or at a different joint [1,5,6,10,12] (see Ref. [4] for a review). The reasons for these observations are not fully understood, but there are three possible explanations. They apply to all three methods that have been used to identify motor output: limb movement [13]; electromyographic (EMG) activity [2,5]; and intracellular potentials [11]. Firstly, they may be due to the spread of current from the tip of the microelectrode [1,7]. This seems unlikely, however, since current threshold increases with the square of the distance from the tip of the microelectrode [1,17]. A second possibility is that the results are due to the spread of impulse activity along lengthy intracortical axonal arbors [3,8,9,18] converging on a discrete zone in which a given muscle is truly represented. Lastly, it may be that widespread 'colonies' of corticospinal neurons with common spinal cord targets are indeed an intrinsic feature of motor cortical organization [1]. We sought to find which of these alternatives best explains results obtained with microstimulation. To this end, we microstimulated every 500 μm along medio-lateral rows and simultaneously recorded the evoked EMG responses of up to a dozen forelimb muscles of the cat. We reasoned that, if the repeated representations of single muscles across the expanse of the motor cortex are the result of intracortical connections that funnel into a focal zone in which a given muscle is truly represented [7] then, the size of the evoked EMG responses should decrease as the microelectrode moves away from the focal zone. Likewise, the latency of the responses should increase as the microelectrode moves away from the focal zone. The present investigation expands in a quantitative manner an approach first taken by Pappas & Strick [14].

The data reported herein were obtained from experiments on seven cats weighting between 2.5 and 4.0 kg. The methods were approved by the local ethics committee and conformed to the procedures outlined in the Guide for the

* Corresponding author. Tel.: +1-418-663-5747; fax: +1-418-663-8756.

E-mail address: charles.capaday@anm.ulaval.ca (C. Capaday).

0304-3940/01/$ - see front matter
PII: S0304-3940(01)02105-X

Care and Use of Laboratory Animals, published by the Canadian Council For Animal Protection. Details on the electrophysiological methods, surgical procedures and homeostatic measures used in the present study can be found in a previous report from this laboratory [3]. Briefly, the animals were anaesthetized with an intramuscular injection of Ketamine (33 mg/kg) and Xylazine (1 mg/kg). The animal's temperature was maintained near 37°C by a heating blanket wrapped around the animal's trunk and by an overhead radiant heat lamp. The blood pressure was maintained at about 100 mm Hg. Once the surgical procedures were terminated, a perfusion pump was connected to a cannula in the femoral vein and a steady flow of anesthetic (10–30 mg/h Ketamine, depending on the animal) was

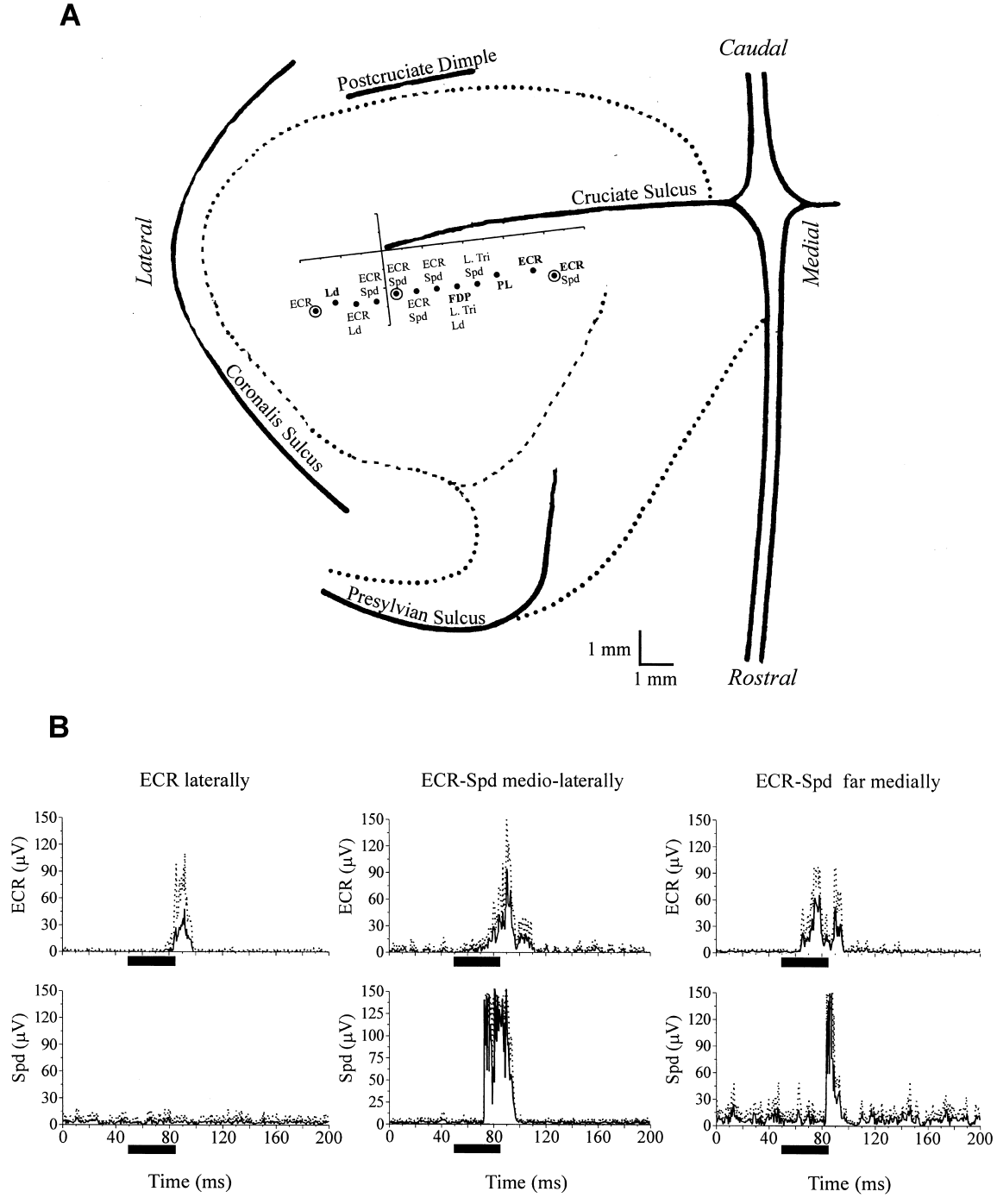

Fig. 1. Example of microstimulation-evoked responses obtained along a row is shown in (A). The stimulus intensity was 40 μA, throughout. The EMG responses of two muscles, the ECR and the Spd, obtained at three different points (circled in A) along the row, are shown in part (B). Each trace is the mean (plus one SD unit above the mean) of eight consecutive responses. The horizontal bars under the graphs in part (B) indicate the onset and duration of the stimulus train.

delivered throughout the experiment. Stainless steel microelectrodes ranging in impedance from 500 KΩ to 1 MΩ were used to microstimulate the motor cortex. Trains of stimuli (33 ms trains of 200 μs square pulses, at a rate of 333 pulses/s) were delivered by a constant current source every 1.5 s as the microelectrode was slowly advanced through the motor cortex to a depth of between 1200 and 1500 μm. This is approximately the depth of layer V of the cat motor cortex, where the current thresholds are lowest. The current intensities used ranged between 10 and 60 μA. The EMG response(s) elicited by microstimulation of a given cortical point was identified for threshold (T) as well as for suprathreshold stimuli, up to 4–6 × T. The EMG signals were recorded with intramuscularly implanted pairs of multi-stranded stainless steel electrodes. The signals were amplified, typically by factor of 1000, high-pass filtered at 20 Hz, rectified, and low-pass filtered at 1 kHz. Recordings were obtained from at least the following forelimb muscles which were exposed and separated by blunt dissection: the flexor digitorium profundus (FDP), the flexor carpi ulnaris and palmaris longus (PL), extensor carpi radialis longus and brevis (ECRl, ECRb), the lateral head of the triceps, the brachialis (Br), the medial head of the biceps, the teres major, the latissimus dorsi (Ld) and the spinodeltoid (Spd). Selected EMG signals were monitored on two four-channel oscilloscopes and the stimulating current on a separate oscilloscope. Responses were digitized at 2 kHz and averaged ($n = 8$) in real-time. During online sampling the stimulus train was delivered at random every 1.5–4 s.

The stimulating microelectrode was initially positioned either far medially, or far laterally in the peri-cruciate area, at random. At this point the response threshold was recorded and the stimulus increased to 1.25 T. The microelectrode was then moved in steps of 500 μm along a row, avoiding blood vessels when necessary. Along each row the same stimulus intensity was used throughout, 1.25 T at the first point. We stopped exploring a row when responses could no longer be evoked. An example of responses recorded along one such row is shown in Fig. 1A. The row spanned 7 mm along the medio-lateral extent of the peri-cruciate area. There are several salient and commonly observed response features along this row. Firstly, the ECR muscle is highly represented, either on its own or in synergy with other muscles such as the Spd or Ld. Secondly, the ECR is highly represented along the row at both lateral and medial sites. One can also observe that the FDP (toe flexor) and the PL (wrist flexion) are represented more medially than what is reported in classical maps (e.g. [13,19]). Similarly, one can observe shoulder muscles such as the Spd and the Ld represented far laterally, again contrary to what is reported in classical maps. Thus, responses in proximal and distal muscles can be elicited from stimulation of both medial and lateral sites, with near equal probability (Table 1). The novel and important aspects of the present study are the quantitative measurements of response size and latency. In Fig. 1B we show the responses of a distal (ECR) and a proximal forelimb muscle (Spd) obtained from microstimulation at three different points along the row shown in Fig. 1A. The size of the ECR response along this row is in fact smallest at the most lateral stimulation point where, according to classical maps, it ought to be the largest. The response of the Spd is largest near the middle of the row, rather than far medially as would be expected from classical maps.

A more detailed example of response size and latency along a row obtained from a different animal is shown in Fig. 2. In the graphs shown in Fig. 2B the response integral and latency of four muscles are plotted as a function of the medio-lateral distance. In the examples shown in Fig. 2B, the largest response of the two most distal muscles (FDP and ECR) was obtained by microstimulation at the most medial points. Conversely, the largest response of the Ld, a proximal muscle, was obtained from points situated more laterally relative to the points yielding the largest distal muscle responses. Results such as those shown in Fig. 2B do not depend on stimulus strength. Increasing the stimulus strength simply increases all responses proportionately. For the five most represented muscles, the average distance between points yielding the maximum and minimum response is given in Table 1. The latencies of the responses were, on the whole, inversely related to response amplitude. However, response latency decreases markedly when there is tonic activity in the muscle. This is why there are instances (marked by an asterisk in Fig. 2B) where a small response may occur at a shorter latency than a larger response. Taking this caveat into consideration, response latency was, nonetheless, closely related to response size (i.e. inversely) and not to the position of the stimulated point. It is also important to note that the largest responses were not associated with increased spontaneous background activity. Furthermore, by repeatedly going back to the first and other points of stimulation and measuring the response threshold, we could be confident that the general level of excitability of the corticospinal system – taken as a whole – was comparable during the course of mapping the row. For the two rows (total = 21) where the response threshold varied by more than ±25%, we stopped mapping that row and proceeded to a new one.

Results such as those shown in Fig. 2 were repeatedly

Table 1
Mean distance and SD between points yielding the maximum and minimum response and probability of obtaining a given response either laterally or medially[a]

Muscle	Mean distance (mm)	Probability*
FDP	2 mm ($n = 5$, SD = 0.3 mm)	0.50 lat, 0.50 med
ECR	3.11mm ($n = 19$, SD = 1.3 mm)	0.52 lat, 0.48 med
Br	3.45 mm ($n = 15$, SD = 1.37 mm)	0.35 lat, 0.65 med
Spd	3.36 mm ($n = 13$, SD = 0.74 mm)	0.46 lat, 0.54 med
Ld	2.86 mm ($n = 16$, 0.68 mm)	0.53 lat, 0.47 med

[a] The edge of the cruciate sulcus was used to demarcate medial (med) and lateral (lat) sites.

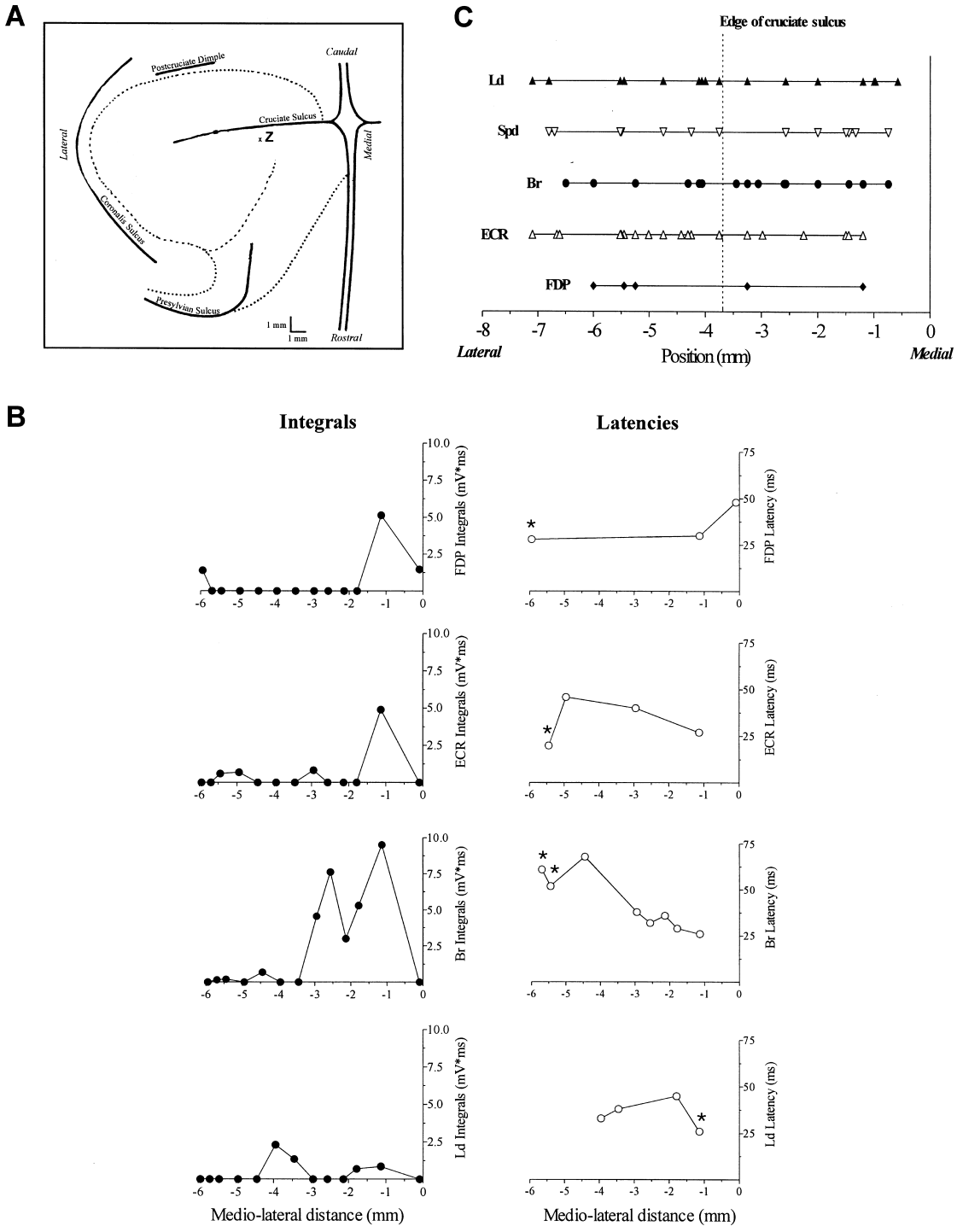

Fig. 2. The position of the zero coordinates of the x-axes of the graphs is indicated by the letter Z in the inset diagram (A) at the top of the figure. Examples of response integrals and latencies of four muscles along a row of microstimulated points (B). Each point on the graphs is the average of eight consecutive responses. Points in the latency graphs marked by asterisks indicate the occurrence of tonic spontaneous activity at the time the response was elicited. For the nineteen rows investigated, the position at which the maximum response of the five most represented muscles was obtained is shown in part (C).

observed in the 19 rows (178 cortical sites and 238 recorded responses) systematically examined in this study and as ancillary observations in our previous study [3]. For the data set as a whole, a Spearman correlation analysis revealed no correlation between cortical position and where the maximum response of a given muscle occurred ($r = 0.15$, $P = 0.22$), as can be seen in Fig. 2C. In other words, the cortical position at which a given muscle would yield its maximum response could not be predicted. We conclude that corticospinal neurons influencing common

spinal cord targets are widely distributed across the forelimb representation area. The results presented herein extend and add support to the conclusions of Phillips and colleagues [1] on the intrinsic organization of the hand area of the simian motor cortex. There is also preliminary neuroanatomical evidence in support of this idea. Intramuscular injections of retrograde transneuronal viruses in the extensor digiti communis muscle have revealed widely distributed clusters of corticospinal neurons in the monkey motor cortex [16]. It remains to be elucidated why the corticospinal neuron colonies of a given muscle are arranged in this manner and how they are used during motor activity.

This work was supported by the NSERC of Canada and in part by the CIHR. Charles Capaday is a senior scholar of the FRSQ. Cyril Schneider was supported by a fellowship of the Canadian Neurotrauma Research Program. We thank Dr Allan Smith for his comments and suggestions on a draft of this manuscript.

[1] Andersen, P., Hagan, P.J., Phillips, C.G. and Powell, T.P.S., Mapping by microstimulation of overlapping projections from area 4 to motor units of the baboon's hand, Proc. R. Soc. Lond. B., 188 (1975) 31–60.
[2] Armstrong, D.M. and Drew, T., Electromyographic responses evoked in muscles of the forelimb by intracortical stimulation in the cat, J. Physiol (Lond.), 367 (1985) 309–326.
[3] Capaday, C., Devanne, H., Bertrand, L. and Lavoie, B.A., Intracortical connections between motor cortical zones controlling antagonistic muscles in the cat: a combined anatomical and physiological study, Exp. Brain Res., 120 (1998) 223–232.
[4] Cheney, P.D., Electrophysiological methods for mapping brain motor and sensory circuits, In A.W. Toga and J.C. Mazziotta (Eds.), Brain Mapping: The methods, Academic Press, New York, 1996, pp. 277–309.
[5] Donoghue, J.P., Leibovic, S. and Sanes, J.N., Organization of the forelimb area in squirrel monkey motor cortex: representation of digit, wrist, and elbow muscles, Exp. Brain Res., 89 (1992) 1–19.
[6] Gould, H.J., Cusick, C.G., Pons, T.P. and Kaas, J.H., The relationship of corpus callosum connections to electrical stimulation maps of motor, supplementary motor, and the frontal eye fields in owl monkeys, J. Comp. Neurol., 247 (1986) 297–325.
[7] Humphrey, D.R., Representation of movements and muscles within the primate precentral motor cortex: historical and current perspectives, Fed. Proc., 45 (1986) 2687–2699.
[8] Huntley, G.W. and Jones, E.G., Relationship of intrinsic connections to forelimb movement representations in monkey motor cortex: a correlative anatomic and physiological study, J. Neurophysiol., 66 (1991) 390–413.
[9] Keller, A., Intrinsic connections between representation zones in the cat motor cortex, NeuroReport, 4 (1993) 515–518.
[10] Kwan, H.C., MacKay, W.A., Murphy, J.T. and Wong, Y.C., Spatial organization of precentral cortex in awake primates. II. Motor outputs, J. Neurophysiol., 41 (1978) 1120–1131.
[11] Landgren, S., Phillips, C.G. and Porter, R., Minimal synaptic actions of pyramidal impulses on some alpha motoneurones of the baboon's hand and forearm, J. Physiol. (Lond.), 161 (1962) 91–111.
[12] Lemon, R.N., The output map of the primate motor cortex, Trends Neurosci., 11 (1988) 501–506.
[13] Nieoullon, A. and Rispal-Padel, L., Somatotopic localization in cat motor cortex, Brain Res., 105 (1976) 405–422.
[14] Pappas, C.L. and Strick, P.L., Physiological demonstration of multiple representation in the forelimb region of the cat motor cortex, J. Comp. Neurol., 200 (1981) 481–490.
[15] Penfield, W. and Rasmussen, T., The Cerebral Cortex of Man: a Clinical Study of Localization of Function, McMillan, New York, 1950.
[16] Rathelot, J.-A. and Strick, P.L., Transneuronal transport of rabies virus from a forelimb muscle to red nucleus and cerebral cortex, Soc. Neurosci. Abst., 25 (1999) 110.
[17] Stoney, S.D., Thompson, W.D. and Asanuma, H., Excitation of pyramidal tract cells by intracortical microstimulation: Effective extent of stimulating current, J. Neurophysiol., 31 (1968) 659–669.
[18] Tokuno, H. and Tanji, J., Input organization of distal and proximal forelimb areas in the monkey primary motor cortex: a retrograde double labeling study, J. Comp. Neurol., 333 (1993) 199–209.
[19] Woolsey, C.N., Settlage, P.H., Meyer, D.R., Spencer, W. and Hamury, T.P., Patterns of localization in precentral and "supplementary" motor areas and their relation to the concept of a premotor area, Res. Publ. Assoc. Res. Nerv. Ment. Dis., 30 (1952) 238–264.