Response patterns and postspike effects of premotor neurons in cervical spinal cord of behaving monkeys[1]

E.E. Fetz, S.I. Perlmutter, M.A. Maier, D. Flament, and P.A. Fortier

Abstract: Most of our information about spinal neurons has been derived from experiments with anesthetized or surgically reduced preparations. To investigate these neurons under normal behavioral conditions, we recorded the activity of single afferent units in cervical dorsal root ganglia and of single interneurons in the cervical spinal cord of macaque monkeys, as they generated alternating flexion and extension torques about the wrist. Spike-triggered averages of rectified electromyographic activity were used to identify "premotor" (PreM) units associated with correlated postspike effects in active muscles. In addition to postspike effects, some spike-triggered averages showed early increases in average muscle activity, which were attributed to synchronous discharges in other PreM units. In recordings of peripheral afferents, 49% of the task-related dorsal root ganglia units produced postspike facilitation (PSF) of at least one forearm muscle, with a mean PSF latency of 5.8 ± 0.3 ms (SE). The PSF amplitude was measured as the mean percent increase (MPI): the average increase of the PSF as a percentage of the prespike baseline mean. PreM afferent units produced PSF with an average MPI of 4.6 ± 0.3%. In a study of cervical interneurons, about 13% (72/562) of the task-related cells showed postspike effects. These PreM interneurons had a mean PSF latency of 7.2 ± 0.3 ms and a mean MPI of 4.6 ± 0.2%. The MPI values for spinal neurons were similar to the MPIs reported for rubromotoneuronal and corticomotoneuronal cells. PreM neurons usually facilitated a subset of the coactivated muscles, called the unit's "muscle field." The PreM afferents facilitated an average of 46% of the synergistically coactivated muscles, while PreM interneurons facilitated an average of 37%. These are comparable with the percentage of muscles facilitated by corticomotoneuronal (40%) and rubromotoneuronal (50%) cells. During the step-tracking task the monkeys generated ramp-and-hold torques about the wrist. The PreM afferents typically became active during either flexion or extension of the wrist, although a few were bidirectionally active. The most common response pattern in PreM afferents was a tonic discharge, followed by phasic and phasic–tonic discharge. The most common patterns exhibited by PreM interneurons were tonic and phasic–tonic responses. PreM afferent units began to discharge on average 51 ± 13 ms before activation of their target muscle. This early onset supports our hypothesis that these PreM afferents arose from muscle spindles, which is also consistent with their short-latency PSF and their responses to perturbations that stretched their target muscles. The results reveal some salient differences between the discharge properties of dorsal root ganglia neurons, spinal interneurons, and supraspinal PreM cells in the motor cortex and red nucleus. All four PreM populations include tonic, phasic–tonic, and phasic cells, but in significantly different proportions. Most PreM afferents resembled corticomotoneuronal cells in being active only with their target muscles, unlike rubromotoneuronal cells and spinal PreM interneurons, which tended to exhibit more bidirectional discharges.

Key words: spinal interneurons, afferent fiber, spike-triggered average, premotor neurons.

Résumé : Notre information sur les neurones spinaux provient généralement d'expériences effectuées sur des sections chirurgicales ou des préparations anesthésiées. Pour examiner ces neurones dans des conditions normales, nous avons enregistré l'activité unitaire d'afférences provenant de ganglions de la racine dorsale (GRD) cervicale et d'interneurones provenant de la moëlle épinière cervicale de singes macaques pendant qu'ils exécutaient des mouvements de flexion–extension du poignet. Nous avons utilisé les moyennes post-stimulation (MPS) de l'activité électromyographique redressée pour identifier les unités «prémotrices» (PréM) associées aux effets post-stimulation correspondants dans les muscles actifs. Outre ces effets, certaines MPS ont montré des augmentations précoces de l'activité musculaire moyenne, qui ont été attribuées à des décharges synchrones dans d'autres unités PréM. Dans des enregistrements d'afférences périphériques, 49% des unités GRD liées à la tâche ont produit une facilitation post-stimulation (FPS) d'au moins un muscle de l'avant-bras, avec une latence de FPS moyenne de 5,8 ± 0,3 ms (ÉT). L'amplitude de la FPS a été exprimée en termes d'augmentation de la moyenne (AM) par rapport à la moyenne initiale de base pré-stimulation. Les unités afférentes PréM

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ont induit une FPS avec une AM moyenne de 4,6 ± 0,3%. Dans une étude d'interneurones cervicaux, environ 13% (72/562) des cellules liées à la tâche ont montré des effets de post-stimulation. Ces interneurones PréM ont eu une latence de FPS moyenne de 7,2 ± 0,3 ms et une AM moyenne de 4,6 ± 0,2%. Les valeurs de l'AM pour les neurones spinaux ont été similaires aux AM signalées pour les cellules cortico-motoneuronales (CM) et les cellules rubro-motoneuronales (RM). En règle générale, les neurones PréM ont facilité un sous-ensemble des muscles co-activés, appelé « champ musculaire » des unités. Les neurones PréM ont facilité en moyenne 46% des muscles co-activés synergiquement, et les interneurones PréM 37%, ce qui se compare au pourcentage de muscles facilités par les cellules CM (40%) et RM (50%). Durant la tâche de poursuite en escalier, les singes ont généré des couples rampe-maintien au niveau du poignet. Les afférences PréM se sont généralement activées uniquement durant la flexion ou l'extension du poignet, bien que certaines aient été actives dans les deux directions. Dans les afférences PréM, le profil de réponse le plus courant a été la décharge tonique, suivie de la décharge phasique et de la décharge phasique–tonique, alors que les décharges toniques et phasique–tonique ont dominé dans les interneurones PréM. Les unités afférentes PréM ont commencé à décharger 51 ± 13 ms en moyenne avant l'activation de leur muscle cible. La précocité de cette réponse confirme notre hypothèse que ces unités originent de faisceaux musculaires, ce qui explique aussi leur FPS de brève latence et leurs réponses aux perturbations ayant provoqué l'étirement de leurs muscles cibles. Les résultats révèlent des différences manifestes entre les propriétés de décharge des neurones des GRD, des interneurones spinaux et des cellules PréM supraspinales dans le cortex moteur et le noyau rouge. Les quatre populations PréM possèdent des cellules phasiques, phasiques–toniques et toniques, mais en proportions très différentes. La plupart des unités afférentes PréM, à l'instar des cellules CM, ont été actives seulement avec leurs muscles cibles, contrairement aux cellules RM et aux interneurones PréM spinaux qui ont présenté davantage de décharges bi-directionnelles.

Mots clés : interneurones spinaux, fibre afférente, moyenne déclenchée par une stimulation, neurones prémoteurs.
[Traduit par la Rédaction]

Introduction

Ironically, our current view of the functions that muscle afferents and spinal interneurons perform in generating voluntary movements is largely based on inferences from observations in anesthetized, immobilized animals. Such studies have yielded a wealth of information about the convergent inputs to different classes of interneurons from afferent fibers and from descending tracts (for reviews see Baldissera et al. 1981; Jankowska 1992). Nevertheless, little is known about two crucial properties that are essential to understanding the functions of segmental neurons in movements: their activity patterns during normal voluntary limb movements and their output effects on the agonist muscles. The combination of these two properties provides significant information about how the activity of spinal neurons contributes to muscle activity. Previous studies have elucidated the response patterns and the output effects of supraspinal premotor (PreM) neurons in the motor cortex and red nucleus (for reviews see Cheney et al. 1988; Fetz et al. 1990). This paper summarizes results of similar experiments with dorsal root afferent fibers (Flament et al. 1992) and interneurons in the cervical spinal cord.

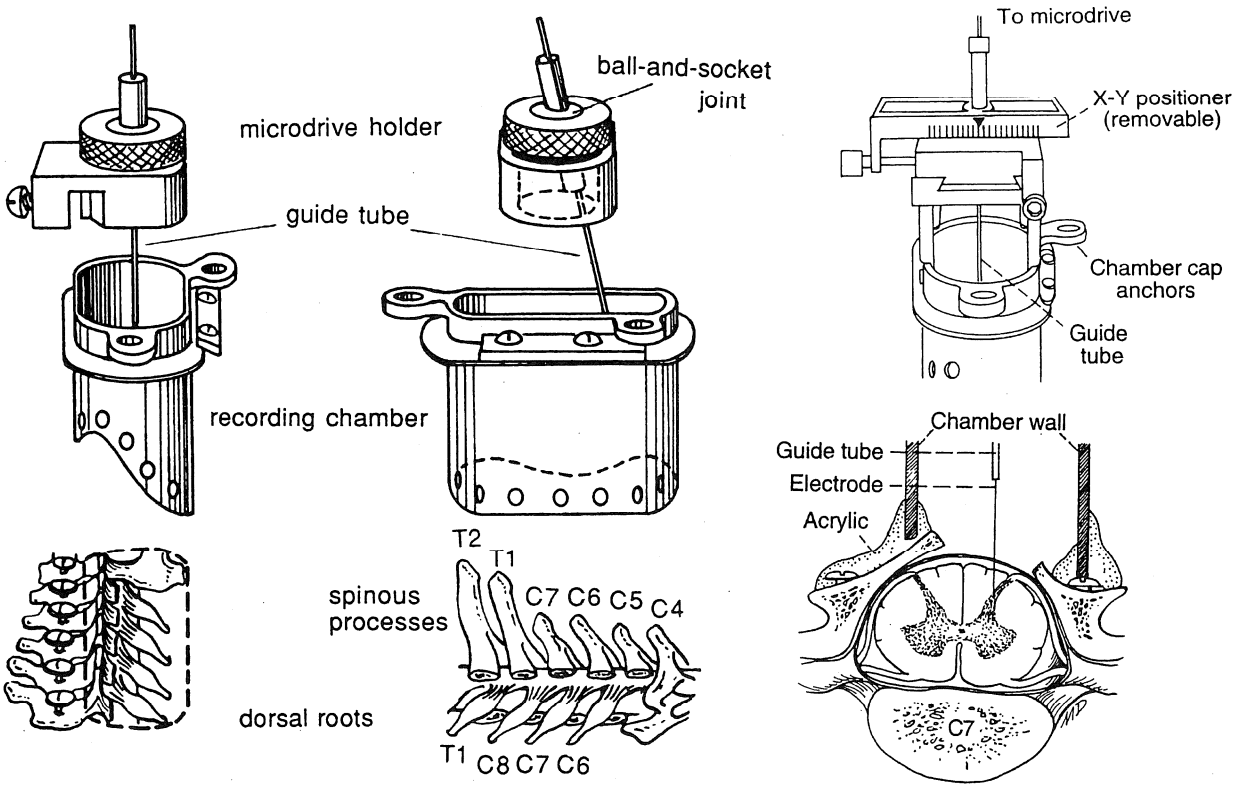
Indirect information about the possible roles of segmental interneurons in active movement has come from reflex testing in humans and neural recordings during rhythmic behaviors in paralyzed or reduced animals. H-reflex studies in man have suggested that inhibitory and excitatory interneurons contribute to voluntary hand and foot movements (e.g., Pierrot-Deseilligny 1989; Nielsen and Kagamihara 1992). The activity of some identified interneurons has been recorded directly during fictive locomotion (e.g., Terakado and Yamaguchi 1990; Jordan 1991) and scratching (Berkowitz and Stein 1994). Virtually nothing is known about the activity of these cells during normal voluntary movement in intact animals.

The normal activity of peripheral afferents during movement has been elucidated in previous studies in humans (Edin and Vallbo 1990; Vallbo et al. 1979; Gandevia et al. 1986), cats (Prochazka 1981; Hulliger 1984; Loeb et al. 1985), and monkeys (Schieber and Thach 1985; Flament et al. 1992). Descriptions of the responses of muscle afferents have elucidated the nature of proprioceptive information reaching the central nervous system during isometric lengthening and shortening contractions of forearm and hind-limb muscles. These studies have shown that the activations of α- and γ-motoneurons are frequently dissociated.

Despite this information, the correlational links of afferents and interneurons with motoneurons have rarely been tested during nonreflex behaviors. Since Ia afferent fibers and last-order spinal interneurons have monosynaptic connections with many motoneurons (Mendell and Henneman 1971; Baldissera et al. 1981; Munsen 1990; Jankowska 1992) and these connections have probabilistic effects on the motoneuron firing (Cope et al. 1987), it seems possible to test the correlational links of afferents with motor units. In the cat, Prochazka (1981) found facilitation of gastrocnemius muscle activity in averages triggered from spikes of spindle primary afferent fibers.

To further elucidate the role of segmental neurons in voluntary movement, we applied the same techniques previously used to investigate supraspinal PreM cells. Spike-triggered averages (STAs) of muscle activity during active movement have revealed output connections of neurons in the motor cortex and red nucleus to α-motoneurons (Fetz and Cheney 1980; Lemon et al. 1986; Mewes and Cheney 1991). The activities of these corticomotoneuronal (CM) and rubromotoneuronal (RM) cells during the performance of a simple ramp-and-hold wrist response exhibit several characteristic discharge patterns that provide insight into the control of dynamic and static components of force (Cheney and Fetz 1980; Buys et al. 1986; Cheney et al. 1988; Fetz et al. 1990; Mewes and Cheney 1994). Here, we investigate primate peripheral afferents and segmental interneurons during a similar behavioral task. Information on both the discharge pattern and postspike effects of

Fig. 1. Schematic view of the spinal unit recording system. An elongated recording chamber was cemented to screws in the lower cervical and upper thoracic vertebrae with dental acrylic. The microdrive holder had a ball-and-socket joint for recordings from dorsal root ganglia (left). An X–Y adapter was used for recording interneurons (right). In both cases the electrode was advanced by a hydraulic microdrive under visual guidance.



PreM neurons provides a basis for comparing the relative control exerted by supraspinal and segmental PreM cells on voluntary muscle activity.

Methods

The activity of units in the cervical spinal cord was recorded extracellularly with glass-coated tungsten or Elgiloy (Rocky Mountain Orthodontics, Denver, Colo.) microelectrodes via a chamber attached to the vertebrae. With the monkeys under halothane anesthesia, we performed a unilateral laminectomy of vertebrae C5–T1 to expose three or four dorsal root ganglia (DRG) (two monkeys) or the dorsal surface of the spinal segments up to the midline (three monkeys). The recording chamber was cemented with dental acrylic to screws in the vertebrae (Fig. 1). For recording, a microdrive was mounted on the chamber with a ball-and-socket joint (three monkeys) or an X–Y stage, which allowed systematic exploration of spinal segments (two monkeys). Electrode penetrations were made under visual guidance through a dissecting microscope. The implants remained securely fixed to the vertebrae for 5–27 weeks.

During recording of unit activity, each monkey was seated in a primate chair with its head and upper back restrained via flexible threaded nylon rods. This restraint permitted the monkey to make small postural adjustments but maintained sufficient stability for recording unit activity. In addition, the arms were restrained by nylon straps. Because forceful movements generally resulted in loss of unit isolation, the limbs were not often released for sensory stimulation. Recording sessions lasted 3–5 h.

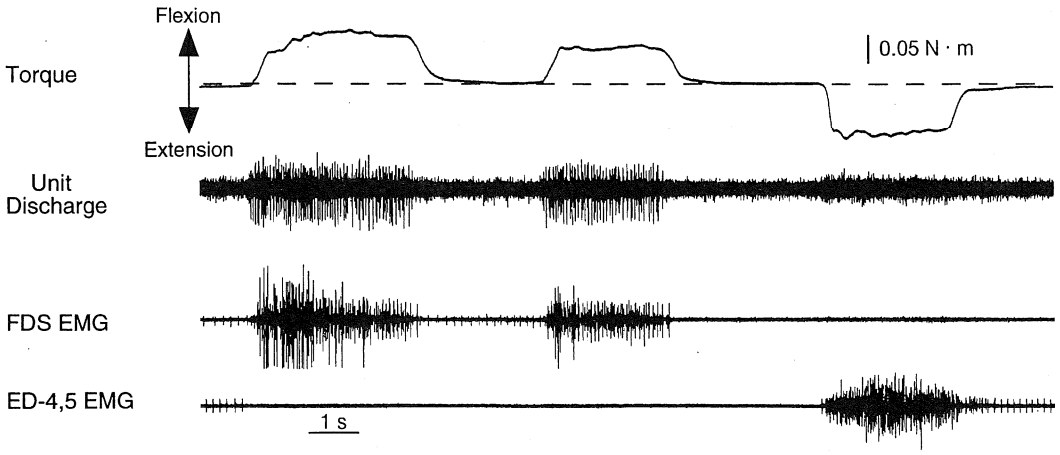
Electromyographic (EMG) activity from wrist and digit flexor and extensor muscles was recorded with stainless-steel multistranded wires implanted transcutaneously in four monkeys. With the monkey under ketamine anesthesia, forearm muscles were identified on the basis of the carpal, metacarpal, and phalangeal movements elicited by intramuscular electrical stimulation (400 Hz, 20-ms train, 0.2-ms pulses, 0.01–1.0 mA). Once implanted, the wires were taped to the arm and could be left in place for 2–3 weeks without loss of recording quality. When they returned to their cages, the monkeys wore a jacket to protect the implants. In one monkey the EMG electrodes were surgically implanted in muscles and connected via subcutaneous leads to a connector attached to the head.

The following muscles were implanted: flexor digitorum superficialis (FDS), flexor carpi ulnaris (FCU), brachioradialis (BR), flexor carpi radialis (FCR), palmaris longus (PL), flexor digitorum profundus (FDP), pronator teres (PT), supinator (SUP), extensor digitorum communis (EDC), extensor carpi radialis (ECR), adductor policis longus (APL), extensor digitorum 4–5 (ED-4,5), extensor digitorum 2–3 (ED-2,3), and extensor carpi ulnaris (ECU).

Near the conclusion of recording in each animal, the sites of a few recorded interneurons were marked with iron deposits from Elgiloy electrodes or electrolytic lesions from tungsten electrodes. Animals were euthanized with an overdose of sodium pentobarbital and perfused with Formalin or paraformaldehyde. The lower cervical and upper thoracic spinal cord were sectioned at 75 μm and stained with cresyl violet and Perls iron reaction when iron deposits were present. Pending detailed reconstruction of electrode tracks the location of the recorded interneurons was provisionally estimated from the electrode depth relative to the first recorded unit activity.

All procedures were approved by the University of Washington Animal Care and Use Committee, and the animals were cared for as directed in the *Guide for the Care and Use of Laboratory Animals* published by the National Institutes of Health, US Public Health Service.

Fig. 2. Recordings of a spinal premotor interneuron and EMG activity during isometric torque trajectories to visual targets. The sweep shows action potentials of a C8 interneuron, flexor and extensor EMGs, and the torque record (flexion upward) for three trials of step tracking.



Behavioral paradigm

The monkeys were trained to make ramp-and-hold torque responses of the wrist in an isometric step-tracking task (Fig. 2). Flexion and extension targets were displayed successively on a video monitor placed in front of the monkey. A cursor controlled by the monkey's wrist torque was displayed continuously on the screen. To receive a fruit-juice or applesauce reward, the monkey had to position the cursor in the target zones for a minimum hold time. Under isometric conditions, unit recordings remained stable for 10–40 min for afferents and up to 60 min for interneurons.

In some cases, when a unit was still isolated after STAs were sufficiently documented, we released and manipulated the monkey's arm to test for natural stimuli (touch, pressure, or displacement) that activated the unit. Because this procedure typically led to loss of unit isolation owing to the passive limb movements or the monkey's reaction, we obtained only limited information on unit responses to peripheral stimulation.

Measurements of postspike effects

STAs of full-wave rectified EMG activity (Fetz and Cheney 1980) were computed on-line to identify units for study, and off-line from tape recordings. A postspike effect was defined as a change in the level of the averaged EMG activity following the trigger spike, which began after a minimal latency of 3.5 ms (see Fig. 3). This latency criterion was chosen because 3.5 ms was the shortest delay observed for poststimulus effects evoked by single-pulse microstimuli delivered through the recording electrode in the DRG and in spinal cord. Triggering units could produce either postspike facilitation (PSF) or postspike suppression (PSS). In some cases, changes in the level of averaged EMG activity started before or near the time of the trigger point (Figs. 3C and 3D). These early changes could be due only to synchronous activity in other units that fired before the triggering neuron and modulated EMG activity.

The latencies of the onset and peak and the magnitude of the postspike effects were calculated after the effects of synchrony were excluded, as shown in Fig. 3 for three different afferent units. First, the mean baseline level of the STA was calculated for a 10- to 15-ms period before the trigger spike. Spike-related changes in EMG, identified as increases or decreases from baseline that persisted for several milliseconds, were characterized as either postspike or synchrony effects on the basis of onset times of more or less than 3.5 ms, respectively. PSF could appear alone (Fig. 3B) or on top of a broad synchrony facilitation (Fig. 3C). The magnitude of the PSF was quantified using the mean percent increase (MPI) of EMG activity, defined as

$$\text{MPI} = \frac{\text{mean bin amplitude of PSF above base}}{\text{mean baseline level}} \times 100$$

In the absence of synchrony facilitation the PSF base was defined as the baseline mean (Fig. 3B). In the presence of synchrony facilitation (Fig. 3C), the onset and offset of the PSF were visually identified as the times at which the STA rose sharply above the broad synchrony effect. The PSF base was taken as the mean of the STA level at these two times. A similar mean percent decrease (MPD) of EMG activity quantified the magnitude of PSS, which was seen alone or superimposed on synchrony suppression.

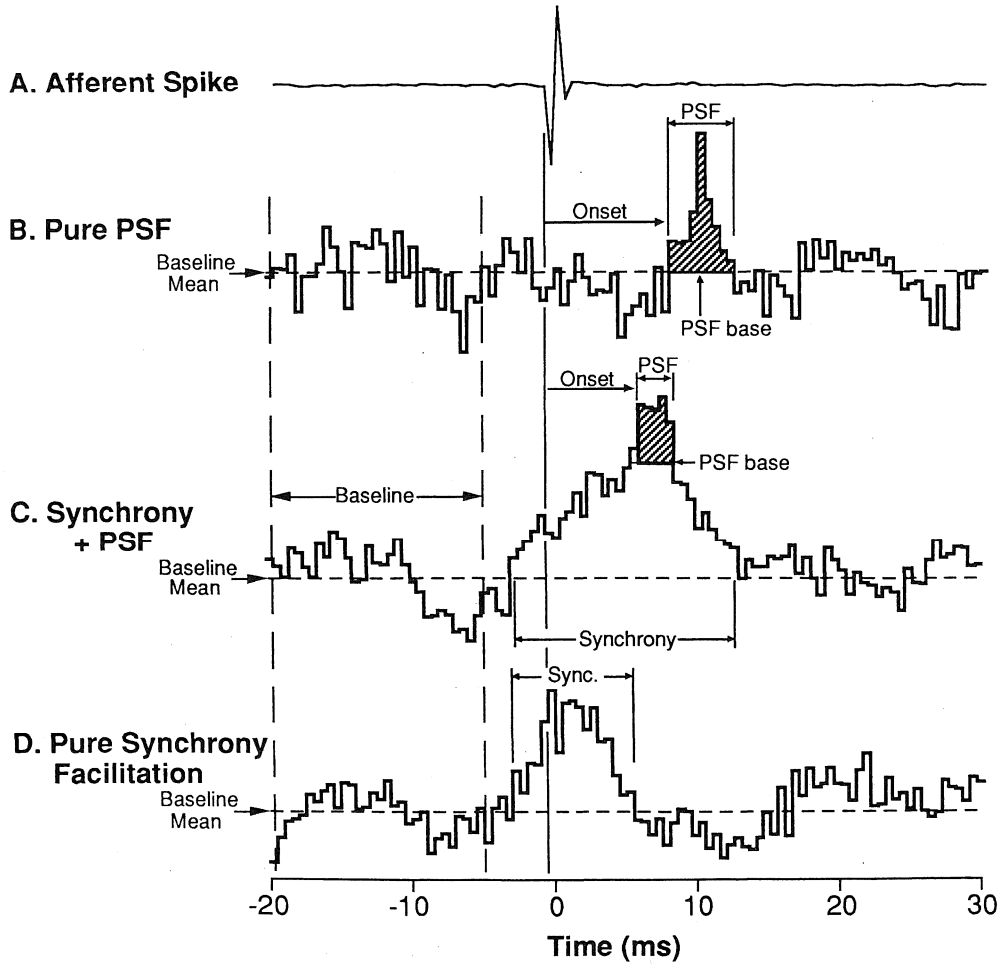
Previous experience with STAs from CM and RM cells had shown that postspike features may appear in averages compiled with substantially fewer than 2000 triggers, but that the postspike effect is more reliably reproduced in averages of 2000 events. Apparent postspike changes obtained with fewer than 1000 trigger events can fluctuate and disappear as more triggers are included. We therefore chose 2000 triggers as a minimal sample size for identifying and measuring postspike effects with reliability.

Redundant EMG recordings were identified by cross-correlating EMG activity off-line (Fetz and Cheney 1980; Lemon et al. 1986). EMG-triggered averages of activity in all muscles were compiled with triggers generated from each muscle in turn. Independent EMG records should produce flat, featureless averages when triggered from each muscle. Peaks of activity in EMG averages, occurring at or near the time of the trigger, can reflect two different mechanisms: redundant recording of the same motor units and (or) synchronous firing of different motor units produced by common synaptic input. To eliminate the possible redundant records from our analysis, we excluded EMG records that showed peaks in EMG averages triggered from other muscles that were >15% of the amplitude of the self-triggered average peaks. It should be noted that this criterion may have excluded some muscles with motor units that were strongly synchronized with those of the trigger muscle.

Response patterns of units

Response patterns of the units during ramp-and-hold torque responses were determined from averages aligned at onset of the torque ramps. These response averages included time histograms of the unit activity, averages of rectified EMG activity, and the isometric torque. The onset latencies of the units relative to the onset of activity in their target muscles were calculated from the response averages.

Fig. 3. Spike-related features in STAs of EMG obtained from three different afferent fibers. (A) The trigger spike. (B) A pure PSF. (C) A PSF superimposed on a synchrony facilitation. (D) A facilitation produced by synchrony (Sync.) alone. The PSF base is identical with the baseline mean in Fig. 3B and is estimated as the average of pre- and post-PSF bin levels in Fig. 3C. Onset latencies were measured relative to onset of the spike (vertical line). From Flament et al. 1992.



Results

DRG neurons producing PSF

Activity of DRG neurons was documented in two macaques, monkeys S and T, generating alternating flexion–extension torques (Flament et al. 1992). STAs of at least 2000 triggers were computed for 59 units. Of these, 29 showed postspike effects in at least one coactive forearm muscle and were classified as PreM afferents. Figure 4 illustrates the STAs and response averages for a neuron in the C8 dorsal root. The STA (Fig. 4, left) shows synchronous discharge in all muscles, in addition to clear PSF in FCU, FDS, FDP, PL, and FCR. The response average (Fig. 4, right) shows a phasic–tonic discharge pattern (histogram at top) in association with the flexion torques (bottom trace). Both the STAs and response averages of muscle activities showed larger modulations in one monkey (S) than in the other (T).

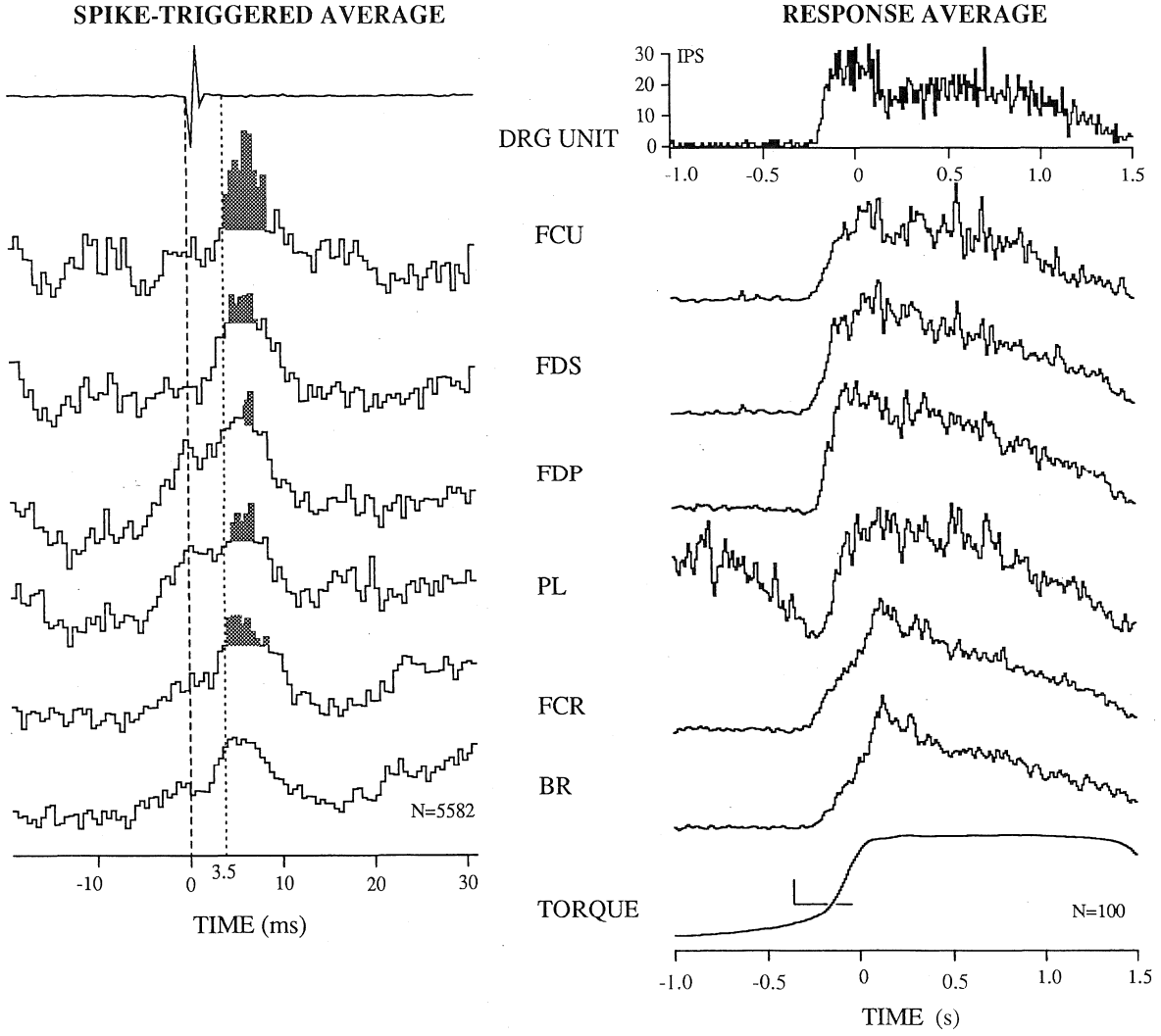
Parameters and distribution of PSF from DRG neurons

The only postspike effect obtained from PreM afferents was facilitation. The mean latency of PSF onset was 5.8 ± 0.3 ms (mean ± SE). The peaks (i.e., the maximal bin value) of the PSF occurred at a mean latency of 7.5 ± 0.3 ms. In contrast, the synchrony facilitation began at a mean latency of 0.2 ± 0.3 ms.

The average MPI of postspike effects for both monkeys, after synchrony was discounted, was 4.6 ± 0.3%. Including the synchrony facilitation with the PSF yielded mean MPIs of the combined facilitation of 9.7 ± 0.5% in monkey S and 4.2 ± 0.8% in monkey T. Synchrony enhancements were found in more STAs of monkey S (56/74 = 75%) than in monkey T (5/18 = 28%). Thus, monkey S showed more synchrony in afferent fiber discharge than did monkey T.

The 29 PreM afferents facilitated one to five muscles, with a decreasing probability distribution. Eleven units (38%) facilitated only one muscle. On average, 46% of all independent EMGs were facilitated. The proportion of independent EMG records showing PSF was similar for flexor muscles (42/92 = 45.6% of EMG records) and extensor muscles (31/67 = 46.3%). About half the units had PSF in flexor muscles, and half in extensors, but no PreM afferents facilitated EMG activity in both.

Fig. 4. STA and response average of a flexor-related phasic–tonic afferent unit recorded from C8 DRG in monkey S. The vertical dotted line indicates the latency that separates postspike from synchrony effects. This unit produced PSF (shaded) in FCU, FDS, FDP, PL, and FCR in addition to synchrony peaks in the flexors. To accurately represent their relative sizes, the STAs are shown in proportion to the MPI in activity relative to baseline (this is equivalent to equalizing their baseline levels). This afferent was coactivated with the flexor muscles during target tracking (right). *N*, number of trigger events. IPS, impulses per second. Verical torque bar = 0.1 Nm; horizontal = zero torque. From Flament et al. 1992.



Discharge patterns of afferent units during wrist activity

During the step-tracking task, 79% of the PreM afferents were active with only one direction of torque, increasing their discharge during either extension (13 units) or flexion (10 units). Six units were active during both flexion and extension, although they facilitated either flexor or extensor muscles, but not both. Five of these cells (17% of PreM afferents) had bidirectionally modulated activity, with a phasic increase in discharge rate during transitions between the static hold phases in both directions. Response patterns of PreM afferents fell into three general categories. The most common (52% of units) was a tonic pattern characterized by a steady increase in activity to a constant level that was maintained throughout the static hold period. The second (21% of units) was a phasic–tonic pattern characterized by an abrupt increase in activity during the dynamic phase of the trajectory, followed by a lower but constant level of activity during the hold period (Fig. 4). In both cases the tonic discharge was maintained despite a tendency of the target muscles to show decrementing levels of activity (Fig. 4). The third pattern (27% of units) was a phasic discharge characterized by a brief increase in activity with change of torque (Fig. 5).

Twelve PreM afferents were sufficiently recorded during passive manipulation of the monkey's arm and hand that their sensory inputs could be estimated. Of these, nine responded to passive displacement of the wrist or fingers and to deep pressure on the forearm tendons. During movements they also responded to perturbations that stretched their target muscles.

Onset latencies of afferent unit activity

An unexpected finding was the early onset of activity in PreM afferents relative to the onset of activity of their facilitated target muscles. Figure 5 illustrates a phasic unit that began to discharge well before its target muscles (asterisks) and before

all other recorded synergists. Figure 6 plots the relative onset latencies for the tonic, phasic, and phasic–tonic PreM afferents. Almost half of the units increased their activity before the onset of EMG activity in each of their target muscles, with a mean onset latency for all cells of –51 ± 13 ms (SE). The cells with a phasic component in their discharge pattern tended to discharge earlier than tonic cells (see symbols at top of Fig. 6). The earliest afferent units recruited were the phasic cells, with a latency of –150 ± 59 ms, followed by the phasic–tonic cells (–72 ± 13 ms) and even later by the tonic cells (–24 ± 17 ms).

Output effects from spinal interneurons

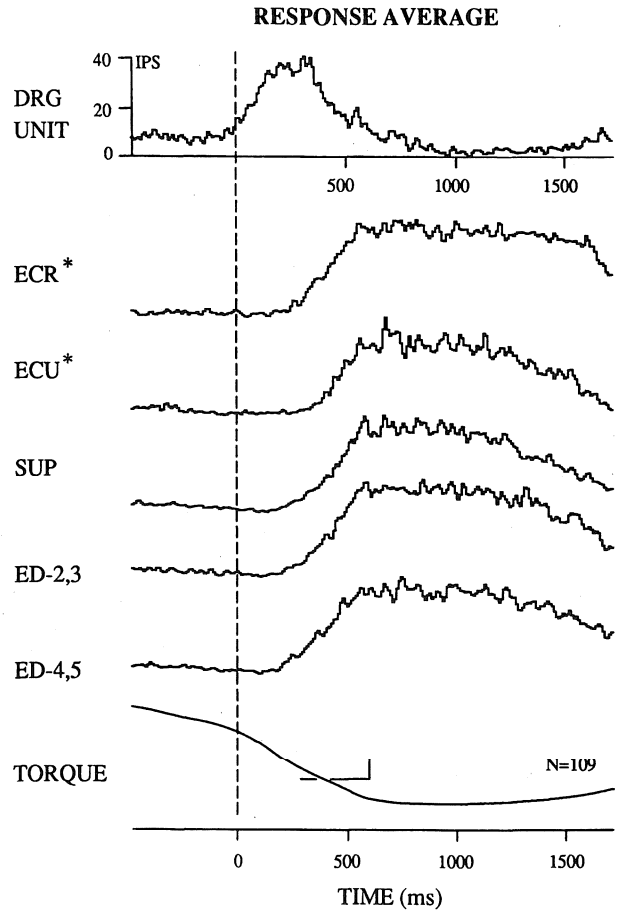
Spinal interneurons were recorded in the C6–T1 spinal segments in three macaques while they performed isometric flexion–extension torques about the wrist. STAs with at least 2000 triggers were calculated for 562 interneurons whose activity was modulated during the wrist task (Fig. 2). These cells were encountered throughout the gray matter, but most were in the intermediate and ventral laminae. In addition, 25 spinal neurons were identified as motoneurons, on the basis of low firing rates (less than 30 spikes/s), activity only during active force generation and only for force in one direction, and depth from the cord dorsum. STAs of unrectified EMGs from these motoneurons often revealed motor unit profiles in a single forearm muscle after a few hundred triggers. Their response patterns during this task resembled those of peripherally recorded motor units documented more extensively in a previous study (Palmer and Fetz 1985).

As with DRG neurons, STAs from spinal interneurons revealed two types of functional correlations: postspike effects and synchrony effects. These were distinguished by comparing their onset latencies with the minimum latency of effects evoked with intraspinal microstimuli, which in these monkeys was 3.5 ms. Seventy-two interneurons exhibited PSF or PSS in at least one muscle beginning at latencies of >3.5 ms and were classified as premotor interneurons (PreM-INs). In addition, 22 interneurons exhibited synchrony effects without identifiable postspike effects in any of the recorded muscles.

Figure 7 shows the STAs and response averages for an excitatory PreM-IN activated during wrist flexion. The neuron's location was estimated to be in the dorsal part of lamina VII medial to the FDS motor nucleus (Fig. 7). This neuron produced PSF in the FDS muscle (Fig. 7, left). The response average, triggered from flexion movements relative to a neutral hold (Fig. 7, right), shows a strong initial burst of activity followed by a slowly decrementing discharge; the neuron did not fire during extension torques. Of the 72 PreM-INs, 58 (81%) showed PSF of target muscles, either in isolation (as in Fig. 7) or superimposed on synchrony facilitation. The mean onset latency of the PSF relative to onset of the triggering spike was 7.2 ± 0.3 ms (mean ± SE, $n = 100$). The latencies of synchrony facilitations from spinal interneurons (mean of 0.4 ms) were similar to those from DRG neurons. The average MPI of PSF from PreM-INs, after synchrony was discounted, was 4.6 ± 0.2%.

An inhibitory PreM-IN estimated to be in a similar laminar location in segment C8 is shown in Fig. 8. The STAs reveal PSS in two of the coactivated flexor muscles, FCR and FCU. This cell, which was recorded while the monkey generated alternating flexion and extension torques, showed a higher level of activity during flexion hold than during extension. A

Fig. 5. Response average of a PreM afferent that became active before any of the recorded agonist muscles, including its two target muscles (ECR* and ECU*). The vertical broken line indicates the onset of unit activity. From Flament et al. 1992.

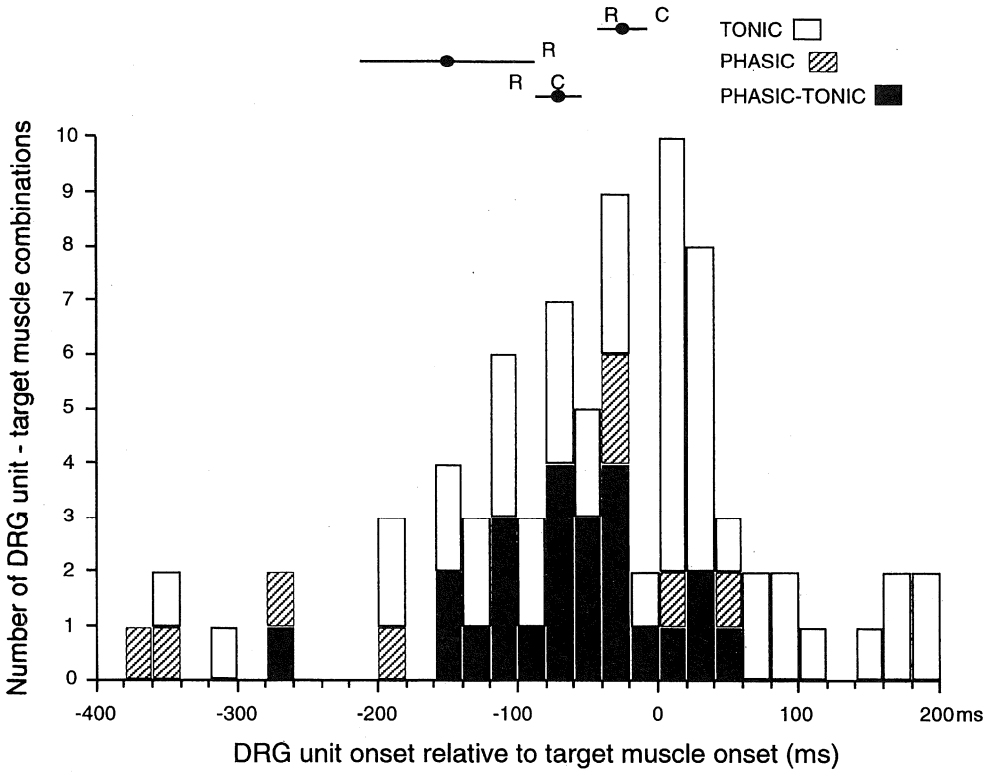


transient pause accompanied the dynamic transitions between extension and flexion holds. Fourteen (19%) of the PreM-INs showed PSS of target muscles, either in isolation (Fig. 8) or superimposed on synchrony suppression. The mean latency of the PSS was 8.1 ± 0.8 ms ($n = 23$) and the average MPD was 3.7 ± 0.3%.

Interneurons exhibited postspike and synchrony effects more often in flexor muscles than in extensors: 62% of PreM-INs had postspike effects in flexor muscles only, while 25% had effects in extensors alone and 13% had effects in both. The proportion was similar for both PSF and PSS. The magnitudes and timing of PSF and PSS were similar in both flexor and extensor muscles in all monkeys.

Thirty-three of the 72 PreM-INs (46%) had postspike effects in only one of the recorded muscles; the rest had larger muscle fields. On average, postspike effects were seen in 37% of the independent coactive muscles. For most of the PreM-INs with divergent postspike effects in more than one muscle (30/39 – 77%), these effects were restricted to either flexor muscles or extensor muscles. The nine neurons with effects in both extensor and flexor muscles included six neurons that cofacilitated the activity in antagonistic muscles, one that co-suppressed flexor and extensor activity, and two that had reciprocal effects.

Fig. 6. Onset latencies of PreM afferents relative to the onset of activity in the facilitated target muscles. Zero on abscissa is the onset of target muscle activity. The results are presented individually for the cells with tonic, phasic, and phasic–tonic response patterns. The histogram displays the latencies of all afferent–muscle pairs; dots and horizontal bars at the top designate the mean ± SE of these distributions. The mean onset latencies of the CM and RM cells, obtained from Cheney and Fetz (1980) and Mewes and Cheney (1991), are also presented for each response type by locations of C and R. From Flament et al. 1992.



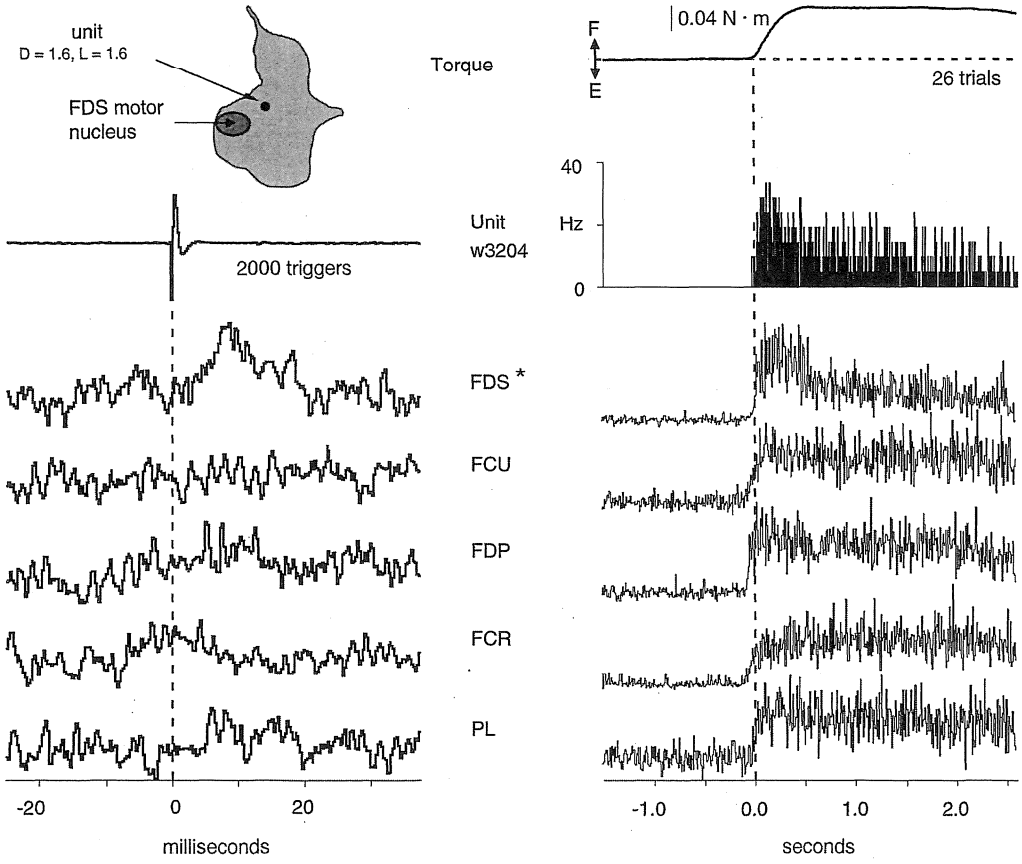
Response patterns and onset latencies of spinal interneurons

Most PreM-INs exhibited the same response patterns observed for PreM afferents, CM cells, and RM cells during the step-tracking task: tonic, phasic–tonic, and phasic patterns. More than half of the PreM-INs had tonic or phasic–tonic activity associated with the ramp-and-hold torque trajectory. Other PreM-INs exhibited patterns not observed for PreM afferents. Figure 9 summarizes the responses of facilitatory PreM-INs, PreM afferents, CM and RM cells, and of single motor units under the same behavioral conditions. A few PreM-INs had a decrementing pattern (e.g., Fig. 7) that is common for forearm muscle motor units (Palmer and Fetz 1985). A phasic-suppression response pattern (e.g., Fig. 8) was identified for several PreM-INs. This pattern is characterized by a significant decrease in discharge rate during the dynamic phase of torque production, superimposed on another firing pattern associated with the static hold phase. Figure 9 also documents the response patterns of 468 spinal interneurons that produced no spike-correlated effects in forearm muscles and are therefore called unidentified interneurons. The relative proportions of response patterns were generally similar in both groups of interneurons; however, a higher percentage of unidentified interneurons exhibited a pure phasic firing pattern.

In contrast with the PreM afferents, only 18 PreM-INs (25%) were active for only one direction of movement. Sixteen PreM-INs (22%) exhibited bidirectional phasic increases in firing during torque transitions, including 4/7 PreM-INs that cofacilitated or cosuppressed flexor and extensor muscles. Most of the remaining PreM-INs were activated during movements in one direction and had some tonic firing during the static hold in the opposite direction.

A basic functional question concerns the relation between the postspike output effects of a PreM cell and its response patterns relative to its target muscles. PreM-INs had relationships that could be classified as "simple" or "complex." The simple category comprises cells whose postspike effects and activation patterns are functionally consistent with simple reciprocal control of flexion–extension movements. These include cells that were activated for one direction of movement and facilitated coactive muscles, suppressed reciprocally active muscles, or did both. Almost half of the PreM-INs (34/72 = 47%) had such simple relations. Of these, most (29/34) fired with facilitated target muscles; three cells fired reciprocally with muscles showing PSS, and two had reciprocal postspike effects, facilitating muscles active in one direction of movement and suppressing antagonist muscles active in the other direction. The remaining PreM-INs had more "complex" relations, in that they were activated at times when their postspike effects would be functionally inconsistent with simple alternating movements. Some cells had totally paradoxical relations. For example, among the interneurons that increased their activity during flexion torques were two cells that produced PSF in extensor muscles and four cells that produced PSS in flexor muscles. A second group of neurons had a steady discharge rate that was not modulated during

Fig. 7. STA and response average of a flexor-related excitatory PreM-IN. This neuron produced PSF in the FDS muscle, in the STA of rectified but not unrectified EMG activity. Its location was estimated on the basis of microelectrode coordinates (inset; D, depth from first neuron recorded in the electrode track; L, distance from the midline). For the locations of cervical motoneuron pools, see Jenny and Inukai (1983). The response average (right) shows activity during flexion (F) from a center hold (broken horizontal line); the neuron did not respond during extension (E).



alternating flexion and extension torques. Each of these cells produced PSF in either flexors or extensors, and maintained a tonic firing rate even when their target muscles were silent. Finally, some reciprocally active neurons cofacilitated or co-suppressed antagonist muscles.

Since most spinal interneurons were not silenced for one direction of movement, onset latencies were measured to the first modulation in activity associated with torque transitions. The mean onset latency, calculated for 44 of the PreM-INs, was –5 ± 63 ms (SD) relative to EMG onset in their target muscles. As with PreM afferents, latencies were widely distributed, from as early as 120 ms before to as late as 120 ms after target muscle onset. Interneurons with phasic increases in activity during torque transitions were modulated earliest in the task, with a mean onset time of –14.5 ms. Cells with response averages exhibiting only tonic components started later, with a mean onset latency of +13 ms relative to target muscles.

Discussion

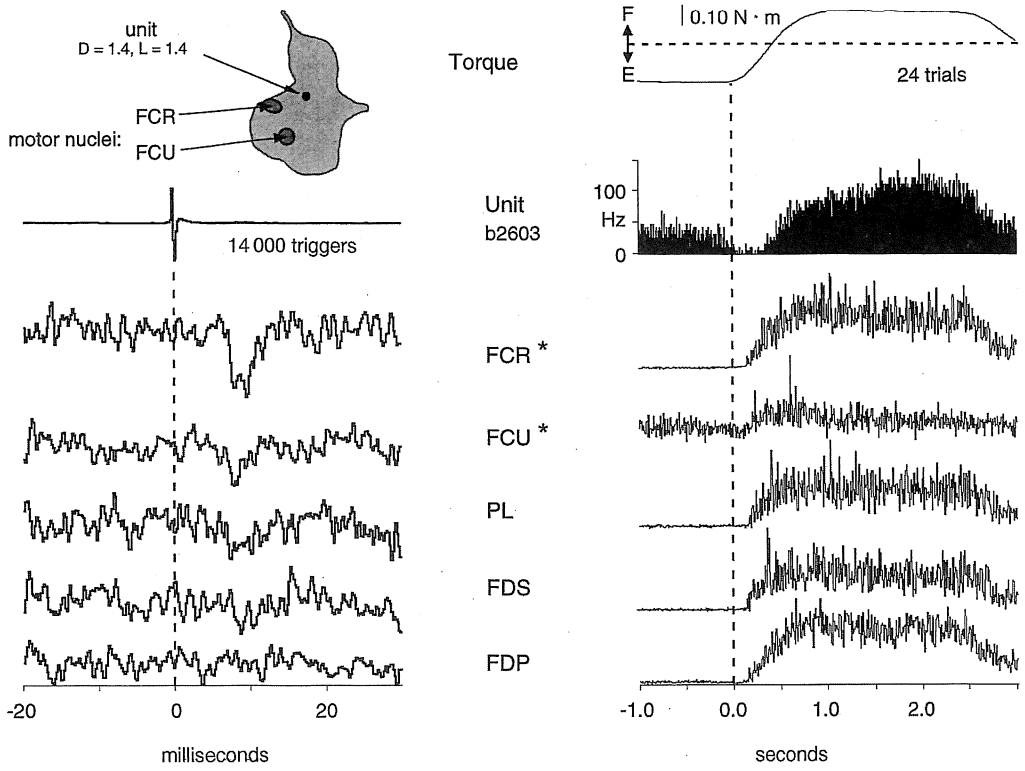
These studies have provided new information about the response properties and output effects of segmental neurons during voluntary motor responses in the primate. At the same time they also raise questions about the identity of these cells, as conventionally defined by criteria used in acute experiments. Unfortunately, our ability to apply similar tests in awake reactive monkeys was limited by our desire to maintain unit isolation and to avoid aversive stimulation. Although the identity of the spinal interneurons remains speculative, there are good reasons to think that the PreM afferents recorded in dorsal root ganglia were Ia afferents.

Identification of dorsal root PreM afferents

Our DRG units were recorded from C8 and T1, which contain fibers from cutaneous, muscle, and joint receptors in the forearm. The sensory receptors of the afferents could not be determined in detail, but of the tested units most (75%) responded to joint movement and deep pressure, both of which are compatible with a spindle origin for these afferents.

Although we could not prove that the PreM afferents arise from spindles, several observations indirectly support this possibility. First, they produced PSF, indicating that they had excitatory correlational links to motoneurons of the facilitated target muscles. Clear PSFs were often obtained with fewer than 2000 trigger events and never required more than 4000, suggesting relatively strong synaptic linkages. While some of these PSFs may have been mediated by disynaptic links, it seems reasonable to assume that the afferents producing

Fig. 8. STA and response average of a flexor-related inhibitory PreM-IN. Asterisks indicate independent muscles with significant postspike suppression. Weak suppressions can also be seen in PL and FDS, but are not statistically significant. Same format as in Fig. 7.



strongest PSF probably contacted α-motoneurons monosynaptically. These afferents would represent group Ia and group II spindle afferents (Kirkwood and Sears 1974). Group Ib muscle afferents (Watt et al. 1976) and cutaneous afferents (Jenner and Stephens 1982) can also provide excitatory drive to motoneurons, but are at least disynaptically linked and would therefore be expected to produce weaker PSF, if any.

Second, the short latency of PSF of most of the afferents is consistent with monosynaptic connections to target motoneurons. A polysynaptic route to the motoneurons would add synaptic delays to the PSF latency. In fact, the PSF of DRG units had a shorter mean onset latency than did PSF of PreM-INs within C7–T1, suggesting that many of the afferents were fast-conducting, probably group I, fibers. Third, 49% of the afferent units for which sufficient data were recorded to calculate STAs with more than 2000 triggers had postspike effects. This high percentage of units facilitating coactive muscles may reflect a sampling bias toward recording the largest afferents, which include the large fibers with direct inputs to motoneurons.

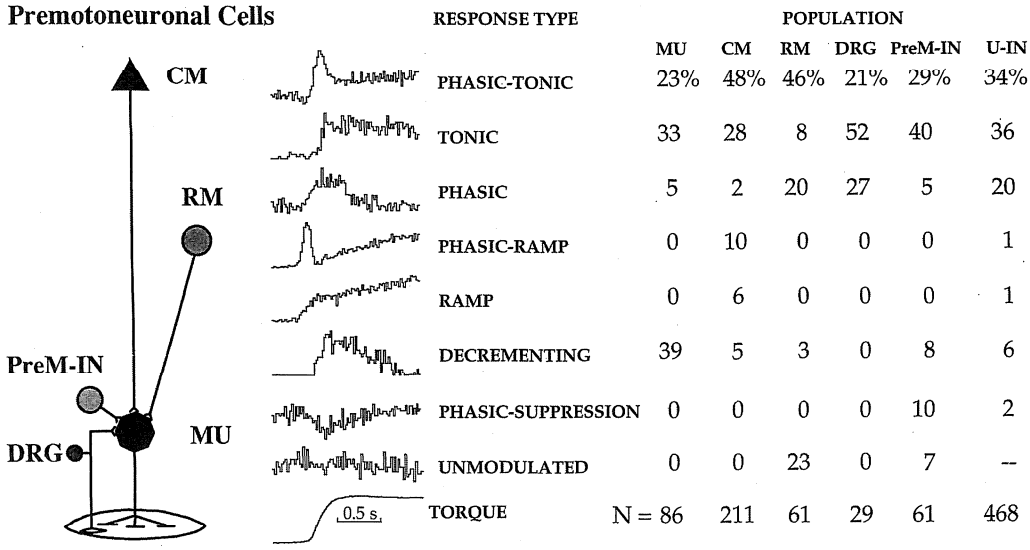
Finally, the PreM afferents that were active prior to any detectable EMG activity in their target muscles were probably spindle afferents. Fusimotor drive to the muscle spindles is the most likely mechanism that could account for increases in afferent activity before EMG onset and in the absence of movement.

Synchrony facilitation and PSF latencies

STAs from afferents and interneurons often revealed a spike-related increase in average EMG activity that began before or near the afferent trigger spike. Increases beginning with a latency of <3.5 ms, the minimal time for electrical stimulation in the cord and DRG to produce a muscle response, could not have been produced by the trigger cell. A likely source of the earlier facilitation is synchronous firing in other PreM cells that discharged before the recorded unit. All our PSF parameters were measured after the presumed synchrony component of facilitation was subtracted. The resulting mean onset latency of the PSF after the trigger spike was 5.8 ms for DRG neurons and 7.2 ms for interneurons. This can be compared with the mean onset latencies reported previously for PSF of RM cells (5.7 ms; Mewes and Cheney 1991) and CM cells (6.7 ms; Fetz and Cheney 1980). The latter would be expected to be longer, since the intraspinal distance to the motoneuron (12–18 mm) is appreciably less than the distance from supraspinal neurons to the same motoneuron (several centimetres).

Synchrony facilitation may explain this apparent inconsistency in latencies. The effects of synchrony in STAs of CM and RM cells were less obvious and were considered negligible in the previous studies. However, recording pairs of CM cells simultaneously, Smith and Fetz (1989) measured synchrony directly and estimated its contribution to the PSF calculated from a CM cell. The effect produced by one synchronous CM cell was minute, although the effects of the entire population of CM cells could be significant. Taking the latency of poststimulus facilitations elicited by microstimulation of the cortex and red nucleus as a measure of conduction time (plus utilization time) would suggest that the previously reported PSFs of CM and RM cells may have included effects due to synchrony (Cheney and Fetz 1985; Mewes and Cheney 1991). Since the PSFs of RM and CM cells were not corrected

Fig 9. Summary of response patterns of different populations of neurons. Examples of each pattern are illustrated in response averages. Proportions are given for motor units (MU; data from Palmer and Fetz 1985), corticomotoneuronal (CM; Fetz et al. 1990) and rubromotoneuronal (RM; Fetz et al. 1990) cells, premotor afferents in dorsal root ganglia (DRG; Flament et al. 1992), facilitatory premotor spinal interneurons (PreM-IN), and unidentified interneurons (U-IN), which did not produce postspike effects. U-INs with bidirectional responses were assigned a response pattern based on the firing profile in the direction of movement that produced the strongest modulation.



RESPONSE TYPE	POPULATION					
	MU	CM	RM	DRG	PreM-IN	U-IN
PHASIC-TONIC	23%	48%	46%	21%	29%	34%
TONIC	33	28	8	52	40	36
PHASIC	5	2	20	27	5	20
PHASIC-RAMP	0	10	0	0	0	1
RAMP	0	6	0	0	0	1
DECREMENTING	39	5	3	0	8	6
PHASIC-SUPPRESSION	0	0	0	0	10	2
UNMODULATED	0	0	23	0	7	--
TORQUE	N = 86	211	61	29	61	468

for synchrony effects, some of the short-latency PSFs could have included an early component of synchrony facilitation.

Magnitude of PSF

The magnitude of the PSF was quantified in terms of the MPI, which measures the size of the PSF above its base as a percentage of the pretrigger baseline EMG activity. PreM afferents and facilitatory PreM-INs had average MPIs of 4.6% after synchrony effects were subtracted. This is about the same as the average MPI of CM cells (4.4%, Kasser and Cheney 1985) and slightly larger than the average MPI of RM cells (2.1%, Mewes and Cheney 1991). The latter may have been overestimated as a result of an undetermined contribution of synchrony facilitation.

The magnitude of the PSF is determined by several mediating factors. One major factor is the size of the excitatory postsynaptic potentials (EPSPs) produced by the trigger cell in the facilitated motoneurons, since motoneuron firing probability is raised in proportion to the amplitude of the EPSP. In motoneurons of anesthetized cats, single-fiber Ia EPSPs produced postspike peaks in the spike-triggered histograms whose counts above baseline were proportional to EPSP amplitudes (Cope et al. 1987). In cats, the sizes of unitary Ia EPSPs range from 10 to 200 μV (Cope et al. 1987; Mendell and Henneman 1971; Watt et al. 1976; Munsen 1990). These would produce cross-correlogram peaks with a MPI of 3–60%. Similar amplitudes have been measured for unitary inhibitory postsynaptic potentials (IPSPs) in hind-limb motoneurons from Ia inhibitory interneurons (Jankowska and Roberts 1972*b*). In cat forelimb motoneurons, Hongo et al. (1989) reported EPSPs of 20 μV amplitude from interneurons in cutaneous reflex pathways.

A second factor affecting the size of PSF is the proportion of motor units facilitated by the trigger cell, since the nonfacilitated motor units would contribute only to baseline and reduce the MPI. Thus, PreM cells that distribute terminals to more motoneurons of the pool would produce a larger PSF, as confirmed for simulations (Fortier 1994). Mendell and Henneman (1971) found that Ia afferents produce EPSPs in virtually all motoneurons of their homonymous muscle. Cross-correlation studies suggest that CM cells facilitate about 72% of the motor units of their target muscles (Mantel and Lemon 1987; Fortier et al. 1989). Since smaller EPSPs may produce no statistically significant correlogram peaks (Cope et al. 1987), the correlational data may underestimate the extent of divergent connections. Information on the divergence of individual last-order interneurons within motor nuclei is available only for a few interneuron types in the lumbar segments of the cord. Jankowska and Roberts (1972*b*) estimated that Ia inhibitory interneurons produce IPSPs in 20% of the motoneurons of the posterior biceps – semitendinosus muscle, based on simultaneous intracellular recordings from motoneurons. Axon morphologies of Ia inhibitory interneurons (Jankowska and Roberts 1972*a*), Ib pathway interneurons (Czarkowska et al. 1976), and midlumbar interneurons with group II input (Bras et al. 1989) show extensive branching within an individual motor nucleus, as well as in multiple motor nuclei.

Finally, the contribution of a facilitated motor unit is proportional to the size of its peripheral muscle unit action potential. In cats the Ia afferent fibers produce larger EPSPs in small motor units (Munsen 1990), which probably constitute the bulk of our EMG records.

Distribution of postspike effects

The number of muscles facilitated by our PreM afferents (an average of 2.6) is compatible with the number found in previous studies of afferent inputs to motoneurons. Recordings of motoneuron EPSPs evoked by stimulation of forearm Ia afferents in baboons revealed convergence from two to five muscle nerves (Clough et al. 1968). A similar experiment in cats

showed convergence from four to six forearm muscles (Fritz et al. 1989). The distribution of the muscle field sizes of our PreM afferents was similar to that reported for CM and RM cells (Fetz and Cheney 1980; Mewes and Cheney 1991).

Multiple target muscles were observed more frequently than single target muscles for both dorsal root afferents and interneurons. For PreM afferents, the facilitated muscles were always agonists contributing to either the flexion or extension torques. Postspike effects on antagonist muscles could not be revealed with the STA technique when DRG units were active only during flexion or extension; however, six cells were bidirectionally active and failed to show postspike effects on antagonist muscles. In contrast, several PreM-INs, like many RM cells (Mewes and Cheney 1991), were active for both directions of movement and showed cofacilitation of antagonistic muscles.

The frequency with which PreM afferents and PreM-INs facilitated flexor and extensor muscles was different. Flexors and extensors were facilitated by nearly equal numbers of DRG neurons, but output effects from interneurons were much more common in flexor muscles. RM cells have the opposite distribution pattern, with effects seen more often in extensor muscles than in flexors (Mewes and Cheney 1991). Similarly, the largest PSFs of CM cells are in extensor muscles (although the frequency with which CM cells facilitate flexor and extensor muscles is similar (Fetz and Cheney 1980)). This suggests that certain muscles are preferentially controlled from supraspinal inputs and others from segmental inputs, a result consistent with the intracellular data of Clough et al. (1968).

Discharge characteristics of premotor neurons and temporal control of motoneuron activity

While PSF identifies the target muscles affected by PreM neurons, the neuron's effect on target motoneurons during active movement is proportional to its discharge rate. The discharge patterns of different populations of PreM neurons and forearm muscle motor units have been studied during similar isometric ramp-and-hold torque trajectories about the wrist (Fig. 9). The distributions of response patterns for PreM afferents and PreM-INs share some features with those for CM and RM cells and motor units.

In some respects the firing patterns of PreM-INs (more than the PreM afferents, CM or RM cells) resemble those of the motor units. The proportion of tonic, phasic–tonic, and phasic units is similar in these two populations. In addition, several interneurons had a decrementing response profile, which was commonly observed for motor units (Palmer and Fetz 1985). In our spinal recording experiments, averages of multiunit EMG typically showed a decrementing profile.

Other characteristics of interneuron firing, however, did not reflect motoneuron properties. Seventy-one percent of PreM-INs maintained at least some background activity during movements in both directions. In the isometric flexion–extension task used here, the only muscles that were ever active for torque production in both directions were the pronator teres, abductor policis longus, brachioradialis, and occasionally the extensor digitorum 2–3. Thus, many interneurons that produced PSFs in primary flexor or extensor muscles continued to fire when their target motor units became silent.

Discharge activity in both directions of movement was also seen for many RM cells (Mewes and Cheney 1994), which shared other firing properties with spinal interneurons. First, bidirectional phasic activity was observed frequently for RM cells and PreM-INs, but only infrequently for PreM afferents and rarely for CM cells. Second, only spinal interneuron and rubral populations included large numbers of neurons with postspike effects that were not correlated with the cell's firing pattern during movement ("complex relationship"). This included some PreM-INs (7%) and many RM cells (23%) with unmodulated firing rates during the flexion–extension task. Third, neurons with output effects on both flexor and extensor muscles have been observed in the red nucleus and spinal cord, but not in the motor cortex or DRG. Cofacilitation of antagonist muscles was seen for 26% of the RM population and cofacilitation or cosuppression for 10% of PreM-INs. Finally, inverse relations between postspike effects and response direction were almost never seen for CM and PreM afferents, but were present for 19% of PreM-INs and 8% of RM cells (Mewes and Cheney 1994).

PreM afferents, like CM cells and motor units, were essentially silent for the direction of torque in which their target muscles were inactive. In contrast with these predominantly reciprocal activities observed in the present study, Schieber and Thach (1985) observed bidirectional spindle afferent activities during precisely controlled wrist movements. One crucial feature of their task was the requirement that the monkey track a slowly moving target. They postulated that the low velocity of the movements required finer control of the wrist; in this task, the central nervous system may adjust the fusimotor drive to permit continuous Ia afferent feedback of muscle length during both flexion and extension of the wrist.

By far the most common response pattern observed for PreM afferents (52%) was a tonic discharge for movement directions that activated their target muscles. The remainder of the DRG units had a phasic increase in activity associated with the dynamic phase of the torque trajectory. Among these, 27% of the population had pure phasic activity during the ramp-and-hold wrist task. This most clearly distinguishes the PreM afferent and CM populations. Relatively few CM cells showed pure phasic discharge (Cheney and Fetz 1980), although a larger proportion showed a transient response pattern in a precision grip task (Lemon et al. 1986). Interestingly, none of the PreM afferents had a decrementing response pattern, even though the associated multiunit EMG often showed a decrementing profile. Thus, the steady level of afferent activity did not reflect the decreasing activity of extrafusal muscle fibers. This is consistent with the α–γ dissociation reported by others (Vallbo 1971; Prochazka et al. 1985; Schieber and Thach 1985).

The combined inputs from all PreM populations to forearm muscle motoneurons during the wrist task were dominated by tonic and phasic response components. The tonic discharge pattern most accurately reflects the torque trajectory itself and the level of tonic discharge has been proportional to the static torque level for the CM, RM, and PreM-IN populations (Cheney and Fetz 1980; Mewes and Cheney 1994; Maier et al. 1994). Phasic discharges prior to the onset of movement in PreM cells could contribute to rapid activation of motoneurons. Other patterns of activity were also observed during step tracking, which could provide additional types of control over the motoneurons. PreM neurons with steady, unmodulated bidirectional discharge apparently provide an excitatory bias during both flexion and extension phases of the task. Some

CM cells show a ramp or a phasic-ramp pattern of activity, which has a steadily increasing discharge rate during the hold period. This would provide a steadily increasing excitatory input to the motoneurons and potentially compensate for their tendency to adapt. The response averages of a small number of spinal interneurons with unidentified outputs also showed ramp components.

Some PreM-INs were transiently suppressed during the dynamic phase of the torque tracking task. Most of these cells also showed increased firing rate during the static hold generated by their facilitated target muscles. Such phasic suppressions have not been described in other PreM populations, and may reflect spinal mechanisms that modulate the supraspinal control of movement initiation. During wrist movements the earliest inputs to forearm motoneurons may be involved in production of the force needed to overcome the inertia of the hand. This requirement may be reflected in the high proportion (nearly 50%) of CM cells with phasic–tonic activity patterns. Inertial forces vary significantly depending on the exact kinematics of the hand at the beginning of the movement. It is possible that supraspinal structures do not specify this force precisely, but that the appropriate motoneuron activations are fine tuned by segmental sensory input at last-order interneurons. The isometric nature of our task could require some downregulation of phasic excitatory inputs to motoneurons to control dynamic force production appropriately. PreM-INs with phasic suppression response components could reflect this element of control. This hypothesis can be tested by comparing the responses of these neurons during auxotonic and isometric torques.

Onset latencies of premotor neurons

Perhaps the most surprising finding about PreM afferents was their early onset of discharge. We expected that early onsets would occur preferentially in supraspinal neurons initiating movement, and that later discharges would occur in peripheral afferents responding to stimulated receptors. Instead, most of the PreM afferents were also part of the population of PreM cells contributing early inputs to motoneurons prior to their activation.

The activities of DRG neurons observed during isometric step tracking could have arisen from a number of peripheral sources. Small displacements of the wrist within the restraints, or activation of cutaneous receptors on the surfaces of the hand, could lead to modulated sensory activity following the EMG activity. A more potent sensory input could arise from fusimotor activation of muscle spindles. Descending motor commands can activate both α- and γ-motoneurons (Hunt and Kuffler 1951; Prochazka 1981; Hulliger 1984). Fusimotor drive to intrafusal fibers during isometric contraction would cause shortening in the poles of the spindle but stretch in the equatorial region, leading to increased spindle afferent activity.

Spindle afferent activities have been studied during normal alternating movements, such as walking (Prochazka et al. 1977; Loeb et al. 1985) and chewing (Taylor and Cody 1974; Goodwin and Luschei 1975), and during wrist flexion and extension movements (Schieber and Thach 1985). Particularly pertinent to the issue of onset time is the work of Vallbo, who examined the activity of spindle afferents during isometric contractions of human forearm muscles (Vallbo 1971; Edin and Vallbo 1990). Most of their spindle afferents began to discharge 0–150 ms after onset of the EMG activity; unit onsets preceded onset of EMG activity by 100 ms in only about 1% of the cases. In contrast, we commonly observed earlier afferent onsets. This apparent discrepancy is probably related to the difference in response conditions. Our monkeys made more forceful and rapid contractions than the controlled isometric forces generated by Vallbo's (1971) subjects; moreover, the monkeys made alternating flexion and extension responses rather than increasing force unidirectionally. Under our conditions most DRG neurons discharged 150 ms before to 100 ms after the onset of muscle activity (mean = –51 ms) and 14% of the unit – PSF muscle combinations discharged more than 150 ms before muscle activity (Fig. 6). The afferent discharge preceding the onset of muscle activity is probably Ia afferent discharge produced by γ-motoneuron contraction of muscle spindles. Since the muscle of origin of spindle afferents could not be identified in our study, it is significant that 12/29 PreM afferents increased their activity before each one of their target muscles.

Spindle afferent activity occurring before muscle activation has been suggested to subserve a follow-up length servo mechanism (Eldred et al. 1953). This hypothesis suggests that the central motor commands control desired muscle length by activating γ-motoneurons, thus generating appropriate afferent activity to produce the desired change in extrafusal muscle length. However, the excitatory inputs to motoneurons from spindle afferents cannot be acting alone to produce muscle contraction because excitatory inputs from supraspinal PreM cells appear well before the onset of EMG activity and movement. Cell onset occurs before activation of its target muscle in 78% of the cases for CM cells (mean = –82 ms; Cheney and Fetz 1980) and in 95% of the cases for RM cells (mean = –89 ms; Mewes and Cheney 1994). Consequently, the combined inputs from supraspinal and segmental PreM cells discharging before the onset of EMG activity contribute to activation of motoneurons. This suggests some degree of α,γ-motoneuron coactivation in the generation of the highly trained single-joint movements of this experiment.

Many spinal interneurons also had early latencies relative to EMG activity in their target muscles. Such responses may reflect inputs from spindle afferents activated by γ-motoneurons prior to movement onset, or from descending fibers, which may include branches of CM and RM cells. Ia afferents and cortico- and rubro-spinal fibers converge on many species of spinal interneurons (Baldissera et al. 1981).

However, the mean onset latency of all PreM-INs, –5 ms relative to target muscle activation, was significantly later than that of PreM afferents, CM cells, and RM cells. These differences appear greater than would be expected for last-order interneurons in a di- or tri-synaptic pathway from supraspinal or dorsal root neurons to motoneurons. Our population of interneurons probably included cells primarily driven by peripheral input from cutaneous or proprioceptive receptors stimulated during the flexion or extension torque, which would be activated at latencies following EMG onset. In contrast, the PreM afferents had earlier onsets because they were probably fusimotor activated spindle afferents. A more comprehensive study of all DRG neurons should reveal many afferents without postspike effects on muscles that have later onsets. Such results would suggest that there are two types of

PreM-INs: those with early onsets driven by networks that initiate movement, and those with later onsets driven by sensory feedback during movement execution. Thus the resulting mean onset latency near EMG onset may reflect a combination of last-order interneurons involved in feedforward and feedback functions.

Interestingly, interneurons and afferents with a phasic component in their discharge pattern tended to be recruited earlier than tonic units. A similar trend was observed in RM and CM cell populations (Cheney and Fetz 1980; Mewes and Cheney 1994). This is consistent with the greater functional efficacy of phasic activity in bringing the motoneurons to threshold and initiating discharge.

Spatial control of motoneuron activity

Some spinal interneurons produced postspike suppressions of EMG activity without reciprocal effects on antagonist muscles. These neurons were probably last-order inhibitory interneurons. In contrast, PSSs observed from CM and RM cells were accompanied by shorter latency PSF of antagonist muscles (Kasser and Cheney 1985; Mewes and Cheney 1991), suggesting that the PSSs reflected reciprocal inhibition mediated by an interposed inhibitory interneuron. The PSSs from spinal interneurons had larger amplitudes (mean MPD = 3.7%) and earlier onset latencies (mean = 7.4 ms) than those for CM (2.9%, 10.1 ms) and RM (2.0%, 9.2 ms) cells, consistent with a more direct synaptic connection.

Spinal interneurons with facilitatory postspike effects outnumbered those that produced PSSs by almost five to one in this study. Synchrony facilitation was also much more common than synchrony suppression. Our data probably underestimate the true proportion of inhibitory PreM-INs as a result of a sampling bias favoring the recording of excitatory interneurons. Although our recording tracks were well distributed throughout the C6–T1 gray matter, inhibitory PreM-INs may be localized in an undersampled region. Differences in cell soma size between excitatory and inhibitory interneurons could also contribute to a sampling bias. However, a more likely explanation is that the STA method could miss many inhibitory postspike effects. STAs of EMG activity will reveal functional connections only when the trigger unit and the target muscle are coactivated. However, many inhibitory interneurons are probably activated reciprocally with their target muscles. This could well explain the relatively small number of PSSs we observed. It could also explain why postspike suppressions were not observed in STAs from DRG neurons, most of which were active only during movements in one direction. Such an activity pattern is consistent with a strict reciprocal organization of flexor and extensor muscle control, where activation of agonists is coupled with inhibition of antagonists.

The activity patterns of other PreM-INs suggest a more complex control of flexor and extensor motoneuron activity. Only half of our PreM-IN population exhibited movement-related activity consistent with straightforward reciprocal control of target muscles. For many other neurons, activity associated with torque directions was more complex than would be predicted from their postspike effects. This included PreM-INs that were activated most strongly for movements opposite those involving their facilitated target muscles, neurons that were activated for both directions of movement but facilitated only flexors or extensors, and neurons with an unmodulated tonic discharge that was not correlated with flexion–extension torque at the wrist. Most of these nonreciprocally organized PreM-INs produced PSFs in forearm muscles. Their excitatory synaptic inputs to α-motoneurons could partially offset strong inhibitory input and keep motoneuron membrane potentials close to threshold. Such excitatory inputs provide more of a general bias than a modulating control, and would enable motoneurons to respond more readily to sensory feedback during phases of the movement when they would otherwise be deeply inhibited. The general absence of these nonreciprocal patterns in PreM afferents suggests that these interneurons, and rubral cells with similar responses, do not exert compelling effects on γ-motoneurons.

Alternatively, the cells with complex relations between postspike effects and firing properties may have synaptic connections whose primary functions are better expressed during other tasks. For example, neurons that cofacilitate flexor and extensor muscles may be in pathways whose main function is to elicit cocontraction during movements in which these muscles are synergists. These neurons may be activated more strongly during different behavioral tasks, such as the power grip. The postspike effects mediated by inappropriate components of the activity in nonreciprocal neurons must be overwhelmed by other inputs to the motoneurons. This could be accomplished with significant numbers of purely reciprocally active inhibitory inputs to motoneurons. As discussed above, our data are consistent with the presence of many such inhibitory interneurons in the C6–T1 segments.

Interestingly, the activity of CM cells during simple single-joint movements is almost exclusively unidirectional, showing no discharge for the opposite direction of torque (Cheney and Fetz 1980; Kasser and Cheney 1985). This suggests that corticospinal activity directly conveys to α-motoneurons the spatial activation patterns seen in effector muscles during the movement. PreM afferents also exhibit almost exclusively unidirectional activity. Since many of these DRG neurons have onset latencies prior to motoneuron activation, it appears that γ-motoneurons are activated by signals that strongly resemble those carried by CM cells. This suggests that central commands exert similar control via the fusimotor system and direct corticospinal inputs to motoneurons. In contrast, many PreM-INs and RM cells showed significant activity during both flexion and extension. This would suggest that these latter populations are involved in a general biasing function on which the reciprocal movements are generated.

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References

Baldissera, F., Hultborn, H., and Illert, M. 1981. Integration in spinal neuronal systems. Handb. Physiol. Sect. 1, Nerv. Syst. (Revis. Ed.), **2**: 509–595.

Berkowitz, A., and Stein, P.S.G. 1994. Activity of descending propriospinal axons in the turtle hindlimb enlargement during two

forms of fictive scratching: broad tuning to regions of the body surface. J. Neurosci. **14**: 5089–5104.

Bras, H., Cavallari, P., Jankowska, E., and Kubin, L. 1989. Morphology of midlumbar interneurones relaying information from group II muscle afferents in the cat spinal cord. J. Comp. Neurol. **290**: 1–15.

Buys, E.J., Lemon, R.N., Mantel, G.W.H., and Muir, R.B. 1986. Selective facilitation of different hand muscles by single corticospinal neurones in the conscious monkey. J. Physiol. (London), **381**: 529–549.

Cheney, P.D., and Fetz, E.E. 1980. Functional classes of primate corticomotoneuronal cells and their relation to active force. J. Neurophysiol. **44**: 773–791.

Cheney, P.D., and Fetz, E.E. 1985. Comparable patterns of muscle facilitation evoked by individual corticomotoneuronal (CM) cells and by single intracortical microstimuli in primates: evidence for functional groups of CM cells. J. Neurophysiol. **53**: 786–804.

Cheney, P.D., Mewes, K., and Fetz, E.E. 1988. Encoding of motor parameters by corticomotoneuronal (CM) and rubromotoneuronal (RM) cells producing postspike facilitation of forelimb muscles in the behaving monkey. Behav. Brain Res. **28**: 181–191.

Clough, J.F.M., Kernell, D., and Phillips, C.G. 1968. The distribution of monosynaptic excitation from the pyramidal tract and from primary spindle afferents to motoneurons of the baboon's hand and forearm. J. Physiol. (London), **198**: 145–166.

Cope, T.C., Fetz, E.E., and Matsumura, M. 1987. Cross-correlation assessment of synaptic strength of single Ia fibre connections with triceps surae motoneurons in cats. J. Physiol. (London), **390**: 161–188.

Czarkowska, J., Jankowska, E., and Sybirska, E. 1976. Axonal projections of spinal interneurones excited by group I afferents in the cat, revealed by intracellular staining with horseradish peroxidase. Brain Res. **118**: 115–118.

Edin, B.B., and Vallbo, A.B. 1990. Muscle afferent responses to isometric contractions and relaxations in humans. J. Neurophysiol. **63**: 1307–1313.

Eldred, E., Granit, R., and Merton, P.A. 1953. Supraspinal control of the muscle spindles and its significance. J. Physiol. (London), **122**: 498–523.

Fetz, E.E., and Cheney, P.D. 1980. Postspike facilitation of forelimb muscle activity by primate corticomotoneuronal cells. J. Neurophysiol. **44**: 751–772.

Fetz, E.E., Cheney, P.D., Mewes, K., and Palmer, S. 1990. Control of forelimb muscle activity by populations of corticomotoneuronal and rubromotoneuronal cells. Prog. Brain Res. **80**: 437–449.

Flament, D., Fortier, P.A., and Fetz, E.E. 1992. Response patterns and post-spike effects of peripheral afferents in dorsal root ganglia of behaving monkeys. J. Neurophysiol. **67**: 875–889.

Fortier, P.A. 1994. Use of spike-triggered averaging of muscle activity to quantify inputs to motoneuron pools. J. Neurophysiol. **72**: 248–265.

Fortier, P.A., Flament, D., and Fetz, E.E. 1989. Relationships between primate corticomotoneuronal cells and their target forearm motor units during torque tracking. Soc. Neurosci. Abstr. **15**: 788.

Fritz, N., Illert, M., Motte, S., Reeh, P., and Saggau, P. 1989. Pattern of monosynaptic Ia connections in the cat forelimb. J. Physiol. (London). **419**: 321–352.

Gandevia, S.C., Burke, D., and McKeon, B. 1986. Coupling between human muscle spindle endings and motor units assessed using spike-triggered averaging. Neurosci. Lett. **71**: 181–186.

Goodwin, G.M., and Luschei, E.S. 1975. Discharge of spindle afferents from jaw-closing muscles during chewing in alert monkeys. J. Neurophysiol. **38**: 560–571.

Hongo, T., Kitazawa, S., Ohki, Y., and Xi, M.C. 1989. Functional identification of last-order interneurones of skin reflex pathways in the cat forelimb segments. Brain Res. **505**: 167–170.

Hulliger, M. 1984. The mammalian muscle spindle and its central control. Rev. Physiol. Biochem. Pharmacol. **101**: 1–110.

Hunt, C.C., and Kuffler, S.W. 1951. Further study of efferent small-nerve fibers to mammalian muscle spindles. Multiple spindle innervation and activity during contraction. J. Physiol. (London), **113**: 283–297.

Jankowska, E. 1992. Interneuronal relay in spinal pathways from proprioceptors. Prog. Neurobiol. **38**: 335–378.

Jankowska, E., and Roberts, W.J. 1972*a*. An electrophysiological demonstration of the axonal projections of single spinal interneurones in the cat. J. Physiol. (London), **222**: 597–622.

Jankowska, E., and Roberts, W.J. 1972*b*. Synaptic actions of single interneurones mediating reciprocal Ia inhibition of motoneurones. J. Physiol. (London), **222**: 623–642.

Jenner, J.R., and Stephens, J.A. 1982. Cutaneous reflex responses and their central nervous pathways studied in man. J. Physiol. (London), **333**: 405–419.

Jenny, A.B., and Inukai, J. 1983. Principles of motor organization of the monkey cervical spinal cord. J. Neurosci. **3**: 567–575.

Jordan, L. 1991. Brainstem and spinal cord mechanisms for the initiation of locomotion. *In* Neurobiological basis of human locomotion. *Edited by* M. Shimamura, S. Grillner, and V.R. Edgerton. Japan Scientific Societies Press, Tokyo. pp. 3–20.

Kasser, R.J., and Cheney, P.D. 1985. Characteristics of corticomotoneuronal postspike facilitation and reciprocal suppression of EMG activity in the monkey. J. Neurophysiol. **53**: 959–978.

Kirkwood, P.A., and Sears, T.A. 1974. Monosynaptic excitation of motoneurones from secondary endings of muscle spindles. Nature (London), **252**: 243–244.

Lemon, R.N., Mantel, G.W., and Muir, R.B. 1986. Corticospinal facilitation of hand muscles during voluntary movement in the conscious monkey. J. Physiol. (London), **381**: 497–527.

Loeb, G.E., Hoffer, J.A., and Pratt, C.A. 1985. Activity of spindle afferents from cat anterior thigh muscles. I. Identification of patterns during normal locomotion. J. Neurophysiol. **54**: 549–564.

Maier, M.A., Perlmutter, S.I., and Fetz, E.E. 1994. Primate spinal interneurons: relation to wrist movement parameters. Soc. Neurosci. Abstr. **20**: 789.

Mantel, G.W.H., and Lemon, R.N. 1987. Cross-correlation reveals facilitation of single motor units in thenar muscles by single corticospinal neurons in the conscious monkey. Neurosci. Lett. **77**: 113–118.

Mendell, L.M., and Henneman, E. 1971. Terminals of single Ia fibers: location, density and distribution within a pool of 300 homonymous motoneurons. J. Neurophysiol. **34**: 171–187.

Mewes, K., and Cheney, P.D. 1991. Facilitation and suppression of wrist and digit muscles from single rubromotoneuronal cells in the awake monkey. J. Neurophysiol. **66**: 1965–1977.

Mewes, K., and Cheney, P.D. 1994. Primate rubromotoneuronal cells: parametric relations and contribution to wrist movement. J. Neurophysiol. **72**: 14–30.

Munsen, J.B. 1990. Synaptic inputs to type-defined motor units. *In* The segmental motor system. *Edited by* M.D. Binder and L.M. Mendell. Oxford University Press, New York. pp. 291–307.

Nielsen, J., and Kagamihara, Y. 1992. The regulation of disynaptic reciprocal Ia inhibition during co-contraction of antagonistic muscles in man. J. Physiol. (London), **456**: 373–391.

Palmer, S.S., and Fetz, E.E. 1985. Discharge properties of primate forearm motor units during isometric muscle activity. J. Neurophysiol. **54**: 1178–1193.

Pierrot-Deseilligny, E. 1989. Peripheral and descending control of neurones mediating non-monosynaptic Ia excitation to motoneurons: a presumed propriospinal system in man. Prog. Brain Res. **80**: 305–314.

Prochazka, A. 1981. Muscle spindle function during normal movement. Int. Rev. Physiol. **25**: 47–90.

Prochazka, A., Westerman, R.A., and Ziccone, S.P. 1977. Ia afferent activity during a variety of voluntary movements in the cat. J. Physiol. (London), **268**: 423–448.

Prochazka, A., Hulliger, M., Zangger, P., and Appenteng, K. 1985. "Fusimotor set": new evidence for alpha-independent control of gamma-motoneurons during movement in the awake cat. Brain Res. **339**: 136–140.

Schieber, M.H., and Thach, W.T. 1985. Trained slow tracking. II. Bidirectional discharge patterns of cerebellar nuclear, motor cortex, and spindle afferent neurons. J. Neurophysiol. **54**: 1228–1270.

Smith, W.S., and Fetz, E.E. 1989. Effects of synchrony between primate corticomotoneuronal cells on post-spike facilitation of muscles and motor units. Neurosci. Lett. **96**: 76–81.

Taylor, A., and Cody, F.W.J. 1974. Jaw muscle spindle activity in the cat during normal movements of eating and drinking. Brain Res. **71**: 523–530.

Terakado, Y., and Yamaguchi, T. 1990. Last-order interneurones controlling activity of elbow flexor motoneurones during forelimb fictive locomotion in the cat. Neurosci. Lett. **111**: 292–296.

Vallbo, A.B. 1971. Muscle spindle response at the onset of isometric voluntary contractions in man. Time difference between fusimotor and skeletomotor effects. J. Physiol. (London), **218**: 405–431.

Vallbo, A.B., Hagbarth, K.-E., Torebjork, H.E., and Wallin, B.G. 1979. Somatosensory, proprioceptive and sympathetic activity in human peripheral nerves. Physiol. Rev. **59**: 919–957.

Watt, D.G.D., Stauffer, E.K., Taylor, A., Reinking, R.M., and Stuart, D.G. 1976. Analysis of muscle receptor connections by spike-triggered averaging. I. Spindle primary and tendon organ afferents. J. Neurophysiol. **39**: 1375–1392.