

Responses of Cerebellar Purkinje Cells to Slip of a Hand-Held Object

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SUMMARY AND CONCLUSIONS

1. Two monkeys were trained to grasp, lift, and hold a device between the thumb and forefinger for 1 s. The device was equipped with a position transducer and strain gauges that measured the horizontal grip force and the vertical lifting or load force. On selected blocks of 20–30 trials, a force-pulse perturbation was applied to the object during static holding to simulate object slip. The animals were required to resist this displacement by stiffening the joints of their wrists and fingers to obtain a fruit juice reward. Single cells in the hand representation area of the paravermal anterior lobe of the cerebellar cortex were recorded during perturbed and unperturbed holding. If conditions permitted, the cell discharge was also recorded during lifting of objects of various weights (15, 65, or 115 g) or different surface textures (sandpaper or polished metal), and when possible the cutaneous or proprioceptive fields of the neurons were characterized with the use of natural stimulation.

2. On perturbed trials, the force pulse was always applied to the manipulandum after it had been held stationary within the position window for 750 ms. The perturbation invariably elicited a reflexlike increase of electromyographic (EMG) activity in wrist and finger muscles, resulting in a time-locked increase in grip force that peaked at a latency between 50 and 100 ms.

3. The object-slip perturbation had a powerful effect on cerebellar cortical neurons at a mean latency of 45 ± 14 (SD) ms. Reflexlike increases or decreases in simple spike discharge occurred in 55% (53/97) of unidentified cells and 49% (21/43) of Purkinje cells recorded in the anterior paravermal and lateral cerebellar cortex.

4. The perturbation failed to evoke complex spike responses from any of the Purkinje cells examined. All the perturbation-evoked activity changes involved modulation of the simple spike discharge. The perturbations stimulated the simple-spike receptive field of most Purkinje cells recorded here, which suggests that the short-latency unit responses were triggered by afferent stimulation. Only one Purkinje cell was found with a distinct complex-spike receptive field on the thumb, but this neuron did not respond to the perturbation. It appears that simple- and complex-spike receptive fields are not always identical or even closely related.

5. The majority of Purkinje and unidentified neurons that responded to the perturbation had cutaneous receptive fields, although some had proprioceptive fields. Seventy-seven neurons were examined for peripheral receptive fields and were also tested with the perturbation. A significantly higher proportion of cells with cutaneous receptive fields (86%) than cells with proprioceptive receptive fields (40%) responded to the perturbation.

6. Preparatory increases in grip force preceding the perturbation appeared gradually and increased in strength with repeated disturbances. Also, the preparatory responses did not disappear immediately when the perturbations were withdrawn, but instead diminished progressively as unperturbed trials were repeated. Thirteen Purkinje cells and 22 unidentified neurons demonstrated changes in discharge associated with these preparatory grip force increases. Although a few cells ($n = 7$) exhibited preparatory re-

sponses without evidence of a reflexlike reaction to the perturbation, a considerably greater number ($n = 28$) displayed both the preparatory and reflex responses.

7. The strong responses of neurons in the cerebellar cortex to slip of a hand-held object indicate that the cerebellum may participate in corrective responses (e.g., cutaneous or proprioceptive long-loop reflexes). With predictable repetition, the perturbations elicited preparatory responses from many of the same neurons that responded at short latency. In general, as the cerebellar activity related to preparatory grip forces increased, the reflex responses in the same neurons decreased. The present study suggests that the information relayed by peripheral afferents about perturbations could contribute to establishing preparatory motor control strategies within the cerebellum.

INTRODUCTION

Since the observations of Flourens in the early 19th century, the cerebellum has been thought to play an important role in both the detection and correction of errors of movement. Both cutaneous and proprioceptive afferents provide powerful excitation of cerebellar Purkinje cells via the mossy fiber–parallel fiber afferents (Eccles et al. 1972; Ishikawa et al. 1972). In locomotion, simple spike activity is strongly modulated during the step cycle (Armstrong and Edgley 1984), whereas complex spike responses have been found only when limb perturbations or electrical stimulation were applied (Apps et al. 1990; Gellman et al. 1985; Kim et al. 1987; Lidieth and Apps 1990). It is not clear whether the same Purkinje cells with complex spike activity evoked by perturbations also have simple spike responses to the same stimulus or whether simple- and complex-spike receptive fields for the same Purkinje cell are largely different.

In humans and monkeys, the sensitive mechanoreceptors in the skin of the fingertips provide important signals about the potential slip of an object grasped in the hand, and these stimuli have been shown to be essential for appropriately adjusting prehensile force to the weight and coefficient of friction of hand-held objects (Johansson and Westling 1987, 1988a; Westling and Johansson 1987). In an earlier study, we observed that the spontaneous slip of an object held between the thumb and index finger produced an altered discharge of cerebellar Purkinje cells in the area of the anterior lobe receiving afferents from the hand (Espinoza and Smith 1990). However, our monkeys usually gripped the test object with a high safety margin (i.e., the extent to which the grip force exceeded the point of potential slip), such that spontaneous slips were relatively rare. Therefore, to study the reaction of Purkinje cells and other cerebellar cortical neurons to object slip, we applied force-pulse perturbations to the hand-held object during

stationary holding (Fig. 1). The objectives of the present study were to investigate the role of cerebellar cortical neurons, first, in detecting the slip of grasped objects and second, in altering and generating new adaptive preparatory motor strategies to compensate for the perturbations.

METHODS

Animals

Two adolescent female *Macaca fascicularis* monkeys weighing between 2.5 and 3.4 kg were used in these experiments.

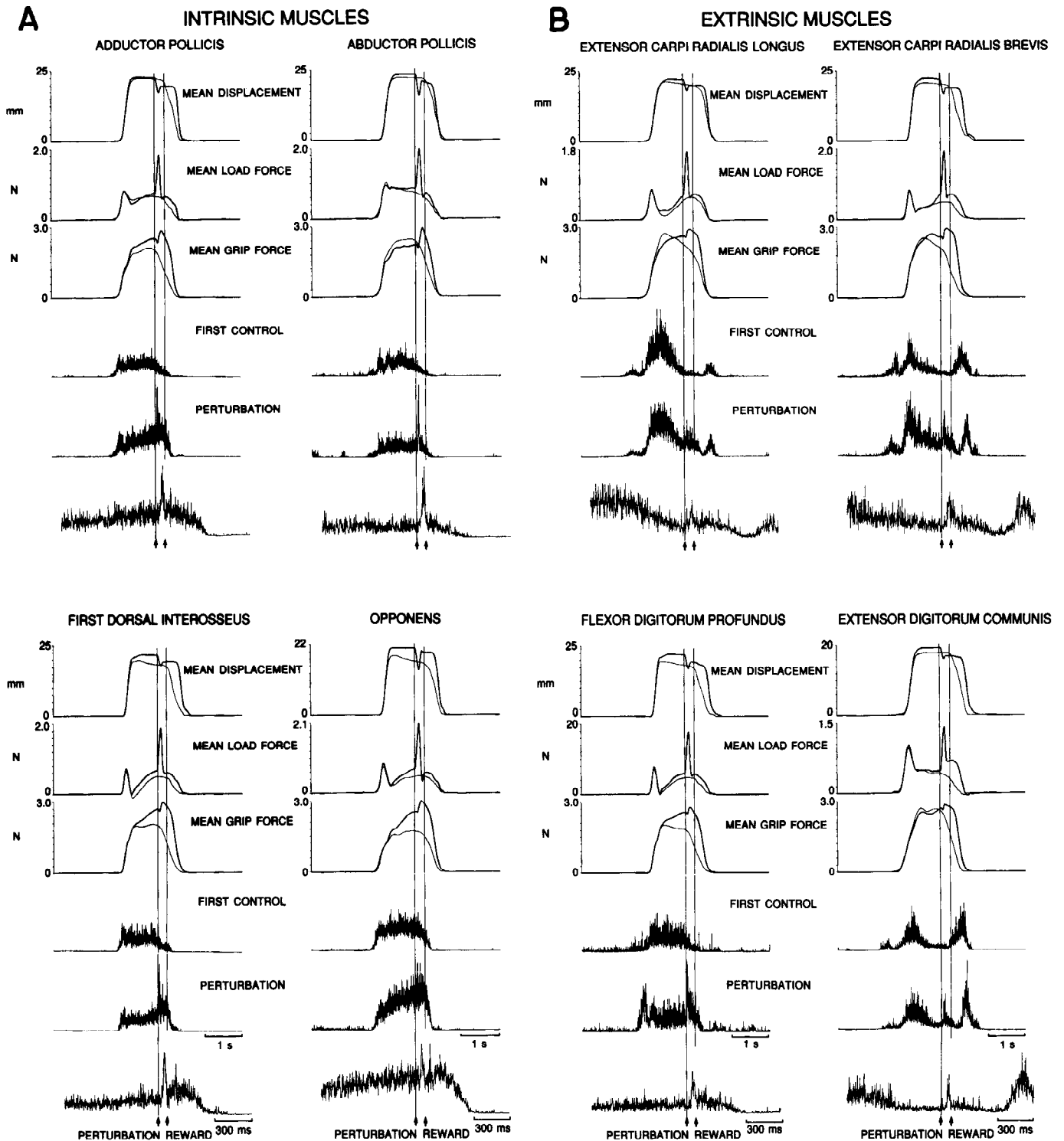


FIG. 1. Mean forces and rectified EMG traces of 4 intrinsic (A) and 4 extrinsic wrist and finger muscles (B) demonstrating clear reflex responses to the perturbation. At least 20 trials were averaged for both the control and perturbed conditions. Grip force, load force, and displacement traces for the 1st control (thin line) and the perturbed (medium line) condition have been superimposed. Mean perturbed EMGs have been shown a 2nd time at *bottom* indicate latencies of perturbed responses with an expanded time base. Gradual increase in the mean rectified EMG in some of the muscles before the perturbation is the preparatory response.

Apparatus and task requirements

The apparatus used to measure grasping and lifting forces was the same as that used by Espinoza and Smith (1990). As in the earlier study, the animals were required to pinch and lift the object a vertical distance of 1.0–2.0 cm above the resting surface and to maintain its position against gravity for 1 s. The object, which was mounted on roller bearings and attached to a vertical track to reduce friction, could be displaced 4 cm vertically from its resting position. Strain gauges measured both the horizontal grip force and the vertical lifting or load force exerted by the animals. A position transducer measured the movement of the object in the vertical axis. A solenoid mounted below the object and connected to it by a rigid rod enabled the application of a transient (100 ms) downward force of ~ 10 N to the object.

The training procedures are described in greater detail in an earlier study (Espinoza and Smith 1990). Essentially, the monkeys were required to pinch, lift, and hold the object in a fixed vertical position for 1 s. A 1-kHz tone served to alert the animals that the object was within the 1-cm vertical position window. After an error that consisted of either overshooting or undershooting the target position window, the animals were obliged to release the object for 2 s before a subsequent trial could be initiated. Once the animal had mastered the essentials of the task, changes in the weight and surface texture of the object were introduced. In the present study, fine sandpaper provided a rough surface and polished aluminum furnished a smooth surface. Object weights of 15, 65, and 115 g were used.

The final step in training was to introduce the 100-ms, 10-N force-pulse perturbation. Unless the animal actively resisted the perturbation, the object would be pulled from the position window and the opportunity for reward would be lost on that trial. The force pulse, once initiated, occurred on every trial after the object had been successfully maintained in the position window for 750 ms. The consistently predictable application of the perturbation gave the monkeys ample opportunity to anticipate its arrival.

For the first monkey tested, the perturbations were introduced only after all combinations of textures and weights had been presented to the animal. For the second monkey, however, testing began with a block of unperturbed control trials followed by a block of perturbed trials and then a second block of unperturbed control trials. The presentation of a second block of unperturbed trials revealed alterations in the motor program that outlasted the perturbation stimulus and also allowed the animal to readapt to the unperturbed condition. The rough surface and 65-g weight were used for all the control and perturbed conditions, although in retrospect the choice of a smooth surface might have been preferable because it would have elicited more slip. Lifting and holding the object at the specified height was repeated ~ 20 – 30 times for each condition of texture and weight. The animals were thoroughly familiar with all the experimental conditions before recording was initiated. During performance of the task, the voltages corresponding to grip and lifting forces and object position were digitally converted at 250 Hz.

Surgical preparation

At the end of the training period, the animals were anesthetized by pentobarbital sodium (30 mg/kg iv or ip) and prepared for chronic single-cell recording according to previously published surgical procedures (Evarts 1965). A circular chamber with an 18-mm ID was stereotaxically implanted over the cerebellum ipsilateral to the working hand at 5.0 mm posterior and 5.0 mm lateral.

Unit recording procedures

After a postoperative recovery period, recording sessions were conducted on a daily basis. Glass-insulated tungsten microelec-

trodes were advanced into the cerebellum with the use of a Trent-Wells microdrive attached to an X-Y micropositioner. Single cells recorded in the cerebellar cortex were identified as Purkinje cells if the presence of a clear climbing fiber discharge could be detected in addition to the presence of simple spikes. Purkinje cells with complex and simple spikes that were sufficiently distinct to trigger an amplitude window discriminator were computer digitized at a frequency of 100 kHz. Six digitized complex and six simple spikes were superimposed for each Purkinje cell; some examples will be illustrated later. Neurons without clearly identifiable complex and simple spikes were grouped together to form a heterogeneous group of unidentified cerebellar cortical units.

Electromyographic recording procedures

Electromyographic (EMG) activity was recorded from intrinsic and extrinsic hand muscles by inserting pairs of 50- μ m Teflon-insulated single-strand stainless steel wires through the skin, and the target muscles were identified by electrical stimulation.

Receptive field evaluation

Whenever possible, an attempt was made to determine the receptive field for each recorded cell. The examination consisted of imposing movements on the wrist and finger joints when the animal was quiescent, stroking the skin with a small camel-hair brush, and palpating and tapping the muscle mass of the ventral and dorsal forearm and the thenar eminence.

Histological preparation of the cerebellum

Before the conclusion of recording, a number of small lesions were made in the cerebellar cortex by passing current through the recording microelectrodes. The animals were anesthetized and perfused with saline followed by a Formalin solution. Each cerebellum was embedded in paraffin and sectioned in the parasagittal plane.

Statistical analysis

Several different statistical tests were applied to the digitized force traces and to the mean discharge frequency during the periods of dynamic and static force on each trial. These methods were described in an earlier paper (Espinoza and Smith 1990). An analysis of variance was used to determine whether the pinch force and discharge frequency of each neuron were significantly altered by the textures, weights, or perturbation. The Scheffé test (Ferguson 1966) was used to make multiple post hoc comparisons between pairs of groups found to contain differences with the analysis of variance. A simple *t* test was used to compare neuronal firing 100 ms before and after the perturbation to determine whether the activity significantly increased or decreased. All the effects of object texture and weight, as well as the reflex and preparatory responses to the perturbation, were statistically significant ($P < 0.001$).

RESULTS

Reflex changes in grip force due to the perturbation

The perturbation typically evoked two types of responses: a reflex reaction, which followed the perturbation, and, with repetition, a preparatory response, which preceded the perturbation. As shown in Fig. 1, the reflex reaction was a stereotyped increase in grip force of ~ 1.4 N, which peaked at a latency between 50 and 100 ms after the stimulus. This prehensile reflex decreased the slip between the fingers and helped stabilize the manipulandum within the position window. The prehensile reflex appeared on the

first presentation of the perturbation and was conspicuous on nearly all subsequent trials. That is, the reflex reactions were invariably locked to the perturbing stimulus and disappeared immediately once the perturbation had been discontinued.

Reflex changes in EMG activity due to the perturbation

The introduction of the perturbation produced changes in the activity of the thumb and index finger muscles that could be related to both the reflex reaction and the preparatory responses. The mean rectified EMG traces of four intrinsic hand muscles and four extrinsic muscles of the wrist and fingers are shown in Fig. 1, *A* and *B*, for blocks of perturbed and unperturbed trials. In addition, the EMGs on perturbed trials are illustrated with an expanded time base to illustrate the latencies of the reflexlike responses, which varied from ~30 to 50 ms. It can be seen that, although the reflex reactions were present in all the muscles shown, these reactions were strongest in the finger muscles contributing most directly to the grip force. It seems reasonable to suppose that the increased EMG activity in these finger muscles contributes directly to the reflex increase in grip force. In contrast, extensor carpi radialis brevis and longus, the prime movers of the wrist involved in lifting the object, were stretched by the perturbation but showed reflex responses that were clearly smaller.

Preparatory changes in grip force due to the perturbation

In contrast to the reflexes, the preparatory response consisted of grip force increases occurring at various times before the perturbation during stationary holding. These responses served to reduce the displacement of the hand due to the impact of the perturbation. The preparatory responses did not appear immediately with the introduction of the perturbation; instead, they emerged over the initial four or five trials and increased in strength with repetition. We have called these responses preparatory to draw attention to the simultaneous presence of both the behavioral manifestation of expectancy shown by the increased grip force and the concomitant preparatory neuronal discharge (to be presented later). Preparatory activity might be distinguished from other forms of set-related neuronal activity for which there is no immediately associated behavior and the anticipatory nature of which is inferred from the subsequent action of the animal (see, for instance, Mauritz and Wise 1986; Wise et al. 1986).

A variety of other preparatory responses could also be seen in the mean force and displacement traces, although they were not analyzed in detail. For example, increases in the peak rate of force change (dF/dt) and peak movement velocity were sometimes observed, and slight increases in the height at which the object was maintained within the position window were also seen on some perturbed trials.

In this study, we confined our analysis to changes occurring during the period of holding without movement. The preparatory response analyzed here consisted of a gradual increase in static grip force before the perturbation. The divergence of mean grip force traces between control and perturbed conditions occurred an average of 450 ms before the perturbation. Collectively, these preparatory responses

stiffened the wrist and fingers before the onset of the perturbation and prevented the hand from being displaced from the position window by the force pulse. The preparatory responses appeared to be learned adaptations that were usually weaker at the outset but became stronger as the monkeys came to expect the perturbation with greater certainty. However, the responses varied in strength and were not necessarily present on every trial. The preparatory responses also persisted for some time after the perturbations had ceased and in general were slower to disappear than they were to appear initially.

Preparatory changes in EMG activity due to the perturbation

Figure 1, *A* and *B*, also shows that preparatory increases in EMG activity are present in some, but not all, of the muscles showing reflex responses. For example, the preparatory responses were conspicuous in the agonist muscles of thumb-forefinger pinching, such as opponens and adductor pollicis, but absent in the coactivated antagonists, such as abductor pollicis brevis and extensor digitorum communis.

Identification and location of recorded neurons

From the histological examination of the lesions and the evidence of electrode tracks in the cortex, it appeared that all the neurons were recorded within the hand representation area 3–9 mm lateral to the vermis in lobules IV–VI and extending ≥ 2 mm in the rostrocaudal plane. The approximate area explored was shown in Espinoza and Smith (1990). Purkinje cells were identified by the presence of the characteristic climbing fiber discharge known as the complex spike (Thach 1967). Neurons not demonstrating complex spikes were grouped together as unidentified cells and may have included both inhibitory interneurons and some mossy fiber terminals. For selected Purkinje cells, the effect of the perturbations as well as of object texture and weight could be analyzed for the complex and simple spikes separately because the potentials could be reliably sorted with a duration-amplitude window discriminator, as will be shown later.

Activity changes related to object texture and weight

The responses to variations in object texture and weight in the present study were very similar to those seen in an earlier study (Espinoza and Smith 1990). The activity of 72 cells was examined in relation to changes in surface texture (smooth metal or fine sandpaper) and three object weights (15, 65, and 115 g). The analysis of variance with post hoc comparisons was used to evaluate the effects of texture, weight, and perturbation.

The Venn diagram in Fig. 2*B* shows the distribution of 72 neurons showing significant ($P < 0.001$) activity modulation with the surface texture, object weight, or perturbation. In this group, about one-half of the neurons responded to the perturbation (43/72 or 60%). All the cells with activity changes related to either surface texture or object weight also responded to the object slip induced by the perturbation. That is, no neurons responded to the perturbation that did not also respond to changes in either object weight or texture as well. Conversely, ~40% (29/72) responded to

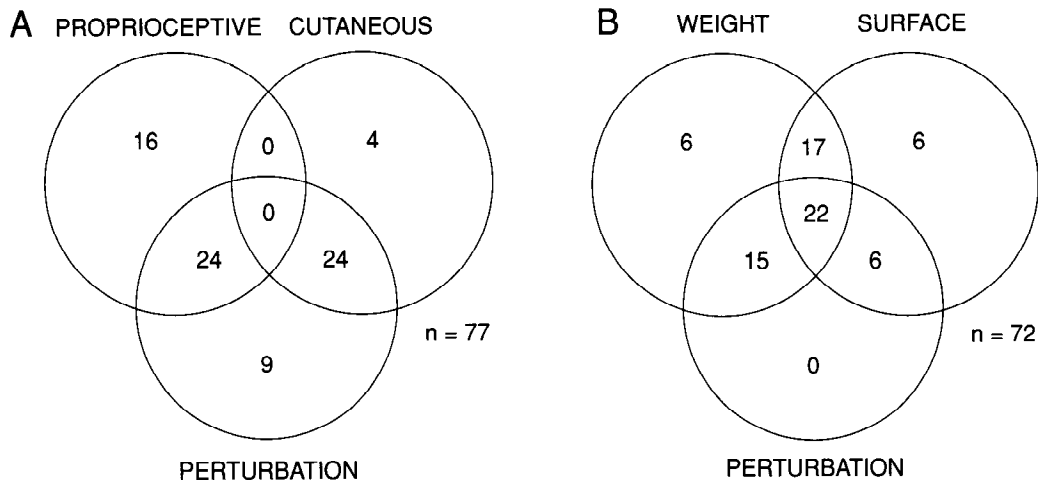


FIG. 2. Venn diagrams showing how many Purkinje and unidentified cells with cutaneous or proprioceptive receptive fields responded to the perturbation (*A*) and how many responses to the perturbation were invariably associated with responses to either texture or weight (*B*).

the changes in texture and weight without responding to the perturbation. For the Purkinje cells, all the activity changes reflected changes in simple spike activity, and no modulation of complex spike frequency was associated with changes in object texture or weight.

The analysis displayed in the Venn diagram in Fig. 2*A* compares 77 cells tested for cutaneous or proprioceptive receptive fields with significant ($P < 0.001$) responses to the perturbation as well. This sample contained no cells with combined proprioceptive and cutaneous receptive fields, which had previously been found to be relatively rare (Espinoza and Smith 1990). As might be expected, the majority (62%) of cells responding to the perturbation had either cutaneous or proprioceptive receptive fields. The Venn diagram in Fig. 2 also shows that a higher proportion of cutaneous cells than proprioceptive cells responded to the perturbation (64 vs. 49%).

Activity changes related to the perturbation

As shown in Table 1, slightly more than one-half of the 140 cerebellar cortical neurons (including 21 Purkinje cells) active in the task had statistically significant responses ($P < 0.001$) to the perturbation within 100 ms. Among these were six cells that responded to the perturbation with decreased discharge frequency. Twenty percent of the cells exhibited both preparatory and reflex responses. Forty-six neurons, or 33%, demonstrated a reflex reaction without any evidence of a preparatory response, and only 7/140, or 5%, of the neurons showed a preparatory response without a reflex reaction.

TABLE 1. Responses of cerebellar cortical neurons to the perturbation

Type of Reaction	<i>n</i>	%
Reflex only	46	33
Reflex and preparatory	28	20
Preparatory only	7	5
None	59	42

n, number of neurons responding (of 140 tested).

Reflex reactions

The neuronal responses to the perturbations were relatively fixed-latency reflexlike increases or decreases in activity that disappeared immediately once the perturbations ceased. A *t* test was used to compare the firing frequency 100 ms before with that 100 ms after the perturbation to determine if the force pulse produced significant changes in activity. Seventy-four units had statistically significant responses to the perturbation and can be found in Table 1. The onset of responses to the perturbation was sufficiently abrupt in 66/74 units to calculate a latency measured from when the manipulandum first changed position. The distribution of cerebellar cortical response latencies, shown in Fig. 3, is unimodal about a mean of 45 ± 14 (SD) ms. There was no significant difference between unidentified neurons and Purkinje cells in the latencies of the reflexes. Also, in both groups of cells, most of the responses to the perturbation were increases in activity, although six cells responded with decreased discharge (e.g., Fig. 4, right).

The magnitude of the reflexlike neuronal responses to the perturbation appeared to be related to either the

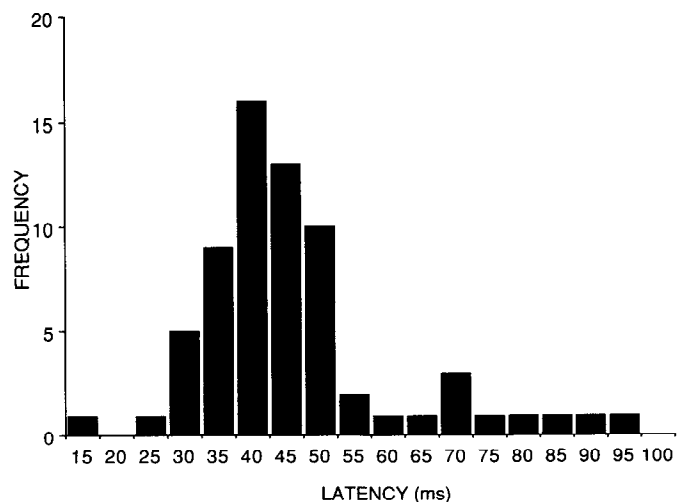


FIG. 3. Latency distribution of 66 cerebellar cortical neurons showing reflex responses to the perturbation.

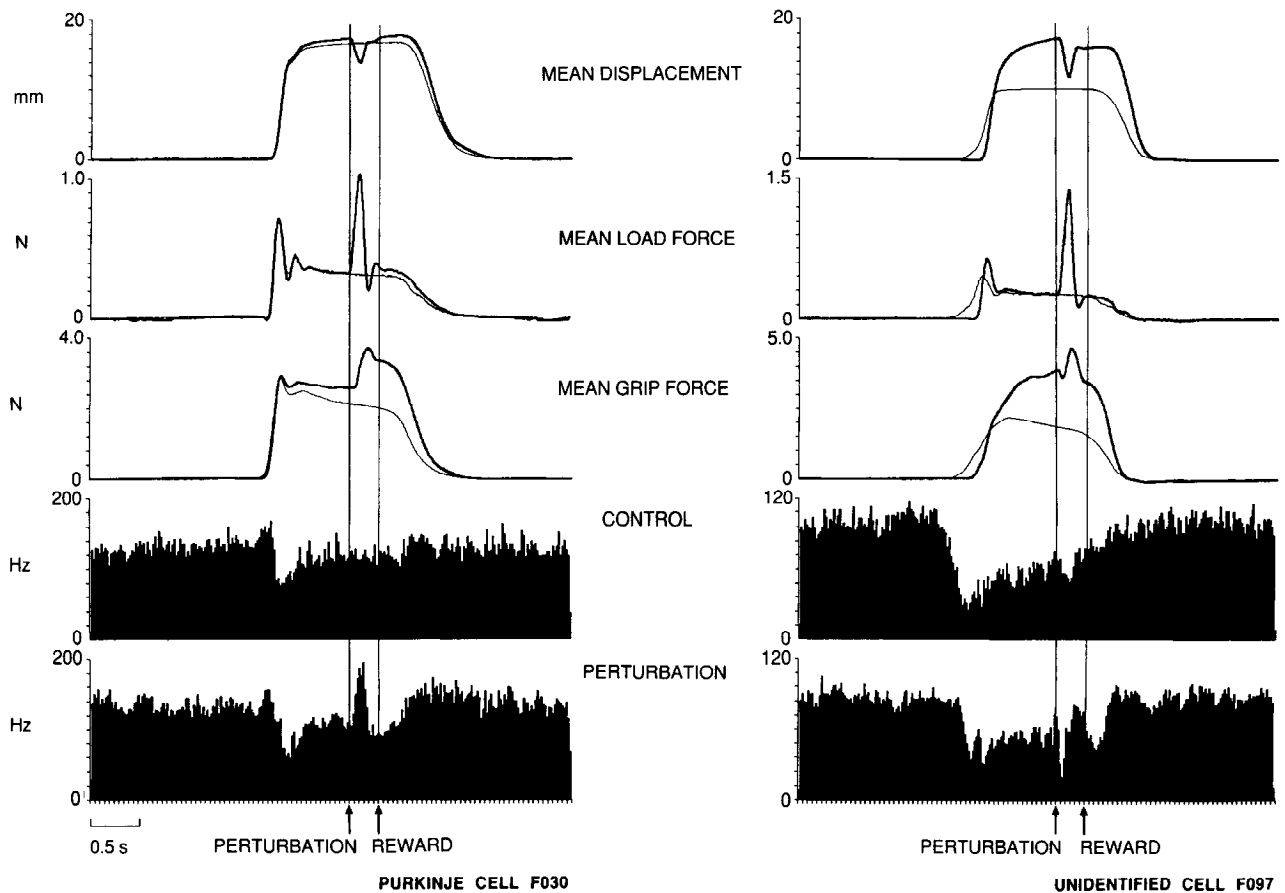


FIG. 4. *Left*: Purkinje cell with an excitatory reflex reaction. *Right*: unidentified neuron with an inhibitory reflex reaction. Forces traces have been superimposed as in Fig. 1.

amount of wrist displacement or the degree of slip against the fingers. Consequently, the greatest reflex responses occurred in those trials when the monkey failed to maintain the object within the 1-cm position window (i.e., an error), and, conversely, the smallest reflex responses occurred when the wrist position was well maintained and minimal slip occurred at the fingers. Although not expressly quantified, it appeared that as the preparatory responses increased the reflexlike response diminished because of decreased slip and reduced displacement of the wrist. That is, the reflexlike neuronal responses and the preparatory neuronal responses appeared to be somewhat inversely related.

Figure 4 illustrates a Purkinje cell and an unidentified neuron, one showing the more commonly encountered reflex increase in activity in response to the perturbation and the other showing a decrease. Because both cells in Fig. 4 exhibited decreased activity during the grasping and lifting movement, it appears that the perturbation can increase or decrease cerebellar cortical cell discharge even when the level of excitation is diminished during grasping. Unfortunately, the Purkinje cell shown on the *left* was lost before a receptive field could be determined, and no clear receptive field was found for the unit shown on the *right*.

Receptive fields were found for a total of 77 cerebellar neurons, of which 40 had proprioceptive receptive fields and 28 had cutaneous receptive fields. There were no significant differences in the latencies of the reflex reactions of neurons with proprioceptive or cutaneous receptive fields. Figure 5 illustrates the task-related simple spike activity of

two Purkinje cells, one with a proprioceptive receptive field (Fig. 5A) and the other with a cutaneous receptive field (Fig. 5B). The cell receiving proprioceptive input was best activated by a passive supination of the wrist, suggesting input from stretch receptors in either pronator teres or quadratus. The cutaneous cell responded to stimulation of the tips of all four fingers, but especially on digits 2–4. The overall activity pattern displayed during grasping and holding appeared to be identical in both Purkinje cells, and the responses to the perturbations were also nearly identical.

Purkinje cell simple and complex spikes

In 19 Purkinje cells, the complex spikes were sufficiently distinct either in polarity or in amplitude to be detected separately by a window discriminator. None of these Purkinje cells consistently emitted complex spikes in response to the perturbation. There was no evidence that the initial perturbation elicited a complex spike that habituated when the force pulse was repeated. All of these same cells, however, had clear simple spike responses to the perturbation. An additional 10 Purkinje cells had complex spikes that could not be sorted automatically but could be displayed and recognized visually with the use of an electrostatic printer (Gould, ES 1000). None of these complex spikes appeared to be related in any way to the perturbation.

Preparatory responses

Although preparatory changes in force and movement were sometimes seen during the dynamic period, we ana-

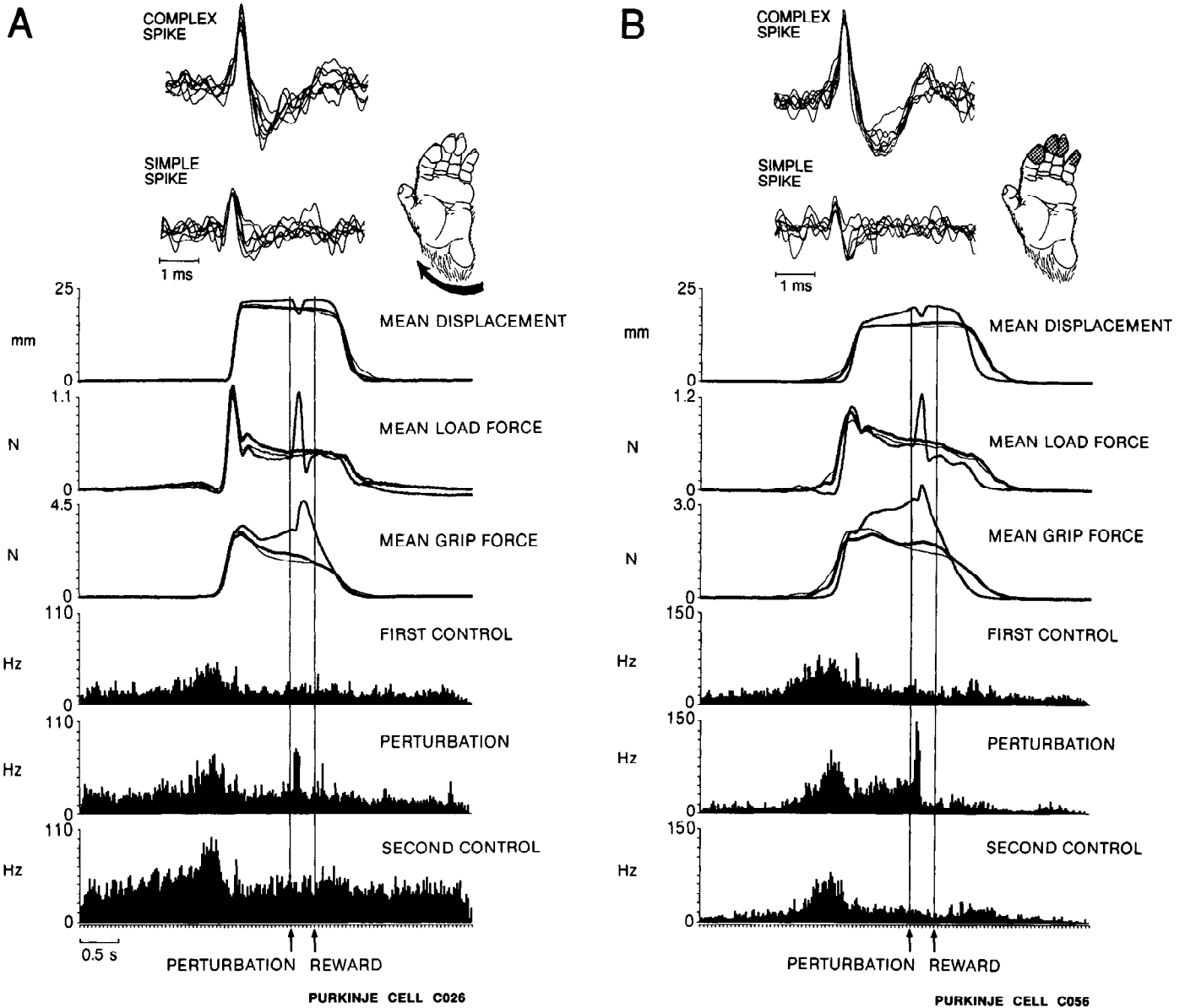


FIG. 5. *Left*: Purkinje cell with a proprioceptive field; *right*: Purkinje cell with a cutaneous receptive field. Average grip force, load force, and displacement traces for the 1st control (thin line), the perturbed condition (medium line), and the 2nd control (thick line) have been superimposed. Receptive field of each cell is shown *above* each panel. Proprioceptive cell was best driven by supinating the hand and stretching the pronator muscles. Cutaneous cell responded best to light touch of the fingertips. The 6 superimposed sweeps show the amplitude and form of the simple and complex spikes for each Purkinje cell.

lyzed only the mean forces and associated activity changes during static holding (i.e., after movement had ceased). By comparing the average grip force traces of perturbed trials with the average grip force on trials before the introduction of the perturbation, it was possible to determine the point at which the mean force traces diverged because of an anticipation of the perturbation. On average, the two force traces began to diverge 450 ms before the onset of the perturbation (Fig. 6). Because all the preparatory responses consisted of increases in discharge frequency, it was possible to estimate the time of activity change with respect to the preparatory force increases for 35 cerebellar cortical neurons. For each cell, the mean firing frequency and standard deviation were calculated for the 1 s of static holding during the first control period before the perturbation was introduced. The mean activity during static holding with the

perturbation was sequentially examined every 20 ms to find the point at which the discharge exceeded the control frequency by two standard deviations. This point was taken as the onset of preparatory activity for the cell. An example is illustrated in Fig. 6. According to this criterion, 13 Purkinje cells and 22 unidentified neurons changed their firing frequencies during the holding period before the perturbation.

A question we wished to address with this analysis was whether the cells with preparatory changes in discharge were responding to altered motor strategy or whether feedback from increased finger pressure and muscle contraction could adequately explain the increases in activity. From the distribution of the onset times of preparatory activity changes in cerebellar cortical neurons shown in Fig. 7, it can be seen that about as many cells increased activity before the grip-force change as after it. The data do not pro-

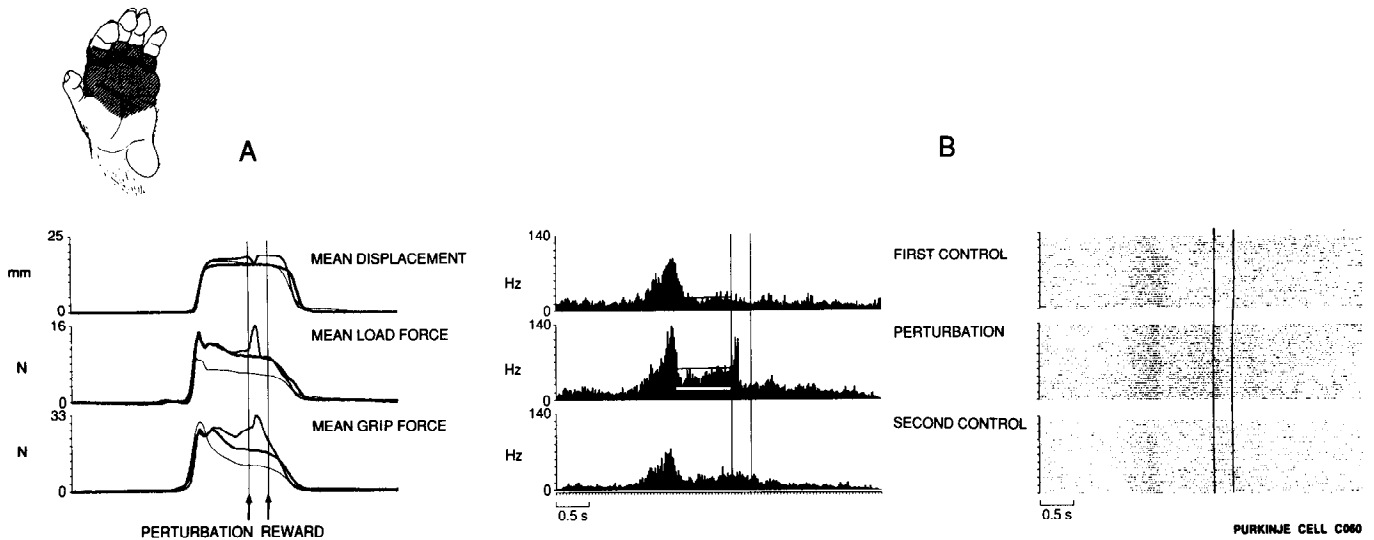


FIG. 6. A cell showing a preparatory and reflex discharge in response to the perturbation. *A*: point at which the preparatory response diverges from control traces can be seen from superimposed grip force traces. *Middle*: mean activity histograms for both control and perturbed trials. Horizontal line in the 1st control histogram indicates the mean static discharge frequency; the 2 horizontal lines in the perturbed histogram illustrate the frequencies corresponding to 2 SD above and below the mean control static frequency and indicate the point at which the mean activity exceeded the mean control frequency. *B*: trial-by-trial raster of cell's activity.

vide any clear answer as to whether the preparatory responses reflect changes in motor control strategy or changes in peripheral feedback due to changes in motor strategy.

The preparatory responses observed in all 13 Purkinje cells were composed entirely of changes in simple spike firing. No changes in complex spike activity or firing probability were found that could be associated with either the expectancy of the perturbation or the reflex reaction to the perturbation itself.

Figure 8 illustrates the mean discharge and grip-force profiles of a Purkinje cell that demonstrated both a strong preparatory discharge and a vigorous reflex reaction to the perturbation. This cell had a proprioceptive receptive field and responded to lateral displacement of the third finger. The mean activity and associated average grip force, load force, and displacement are shown for successive 35-trial blocks involving a block of unperturbed control trials followed by a block of perturbed trials and finally a second

block of unperturbed control trials. From the *middle* histogram, it can be seen that the preparatory discharge diverged from the control ~500 ms before the onset of the perturbation and that the cell discharge increased 50 ms before that.

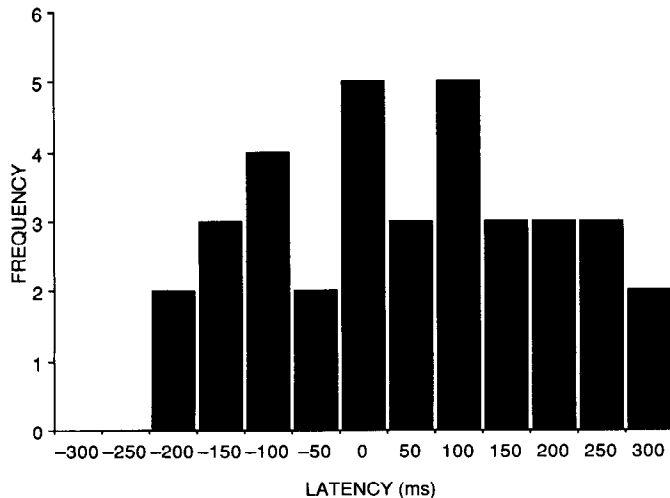


FIG. 7. Histogram of onset times for 13 Purkinje and 22 unidentified neurons with respect to the anticipatory changes in prehensile force.

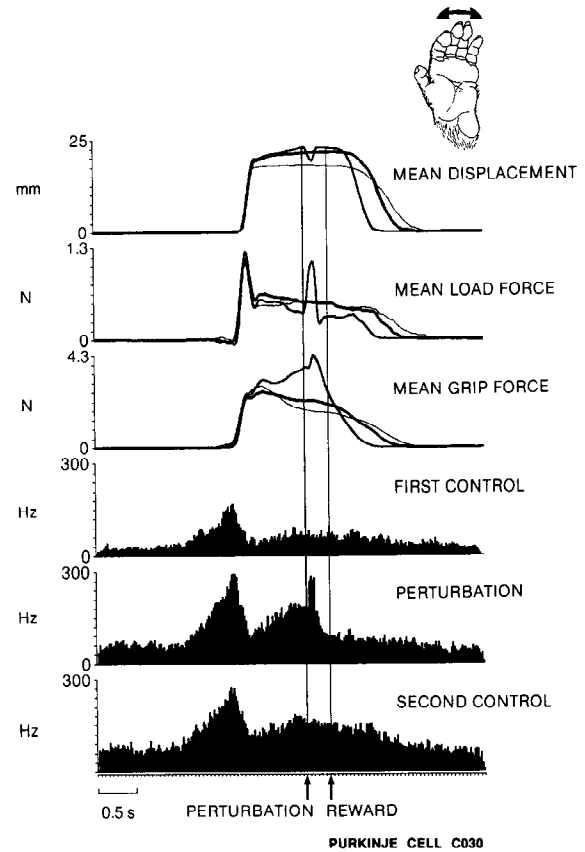


FIG. 8. Superimposed mean force and displacement traces (using the same conventions as in Fig. 5) and the mean discharge of a Purkinje cell demonstrating a preparatory response and a reflex reaction to the perturbation. This proprioceptive cell responded to lateral displacement of the 3rd finger.

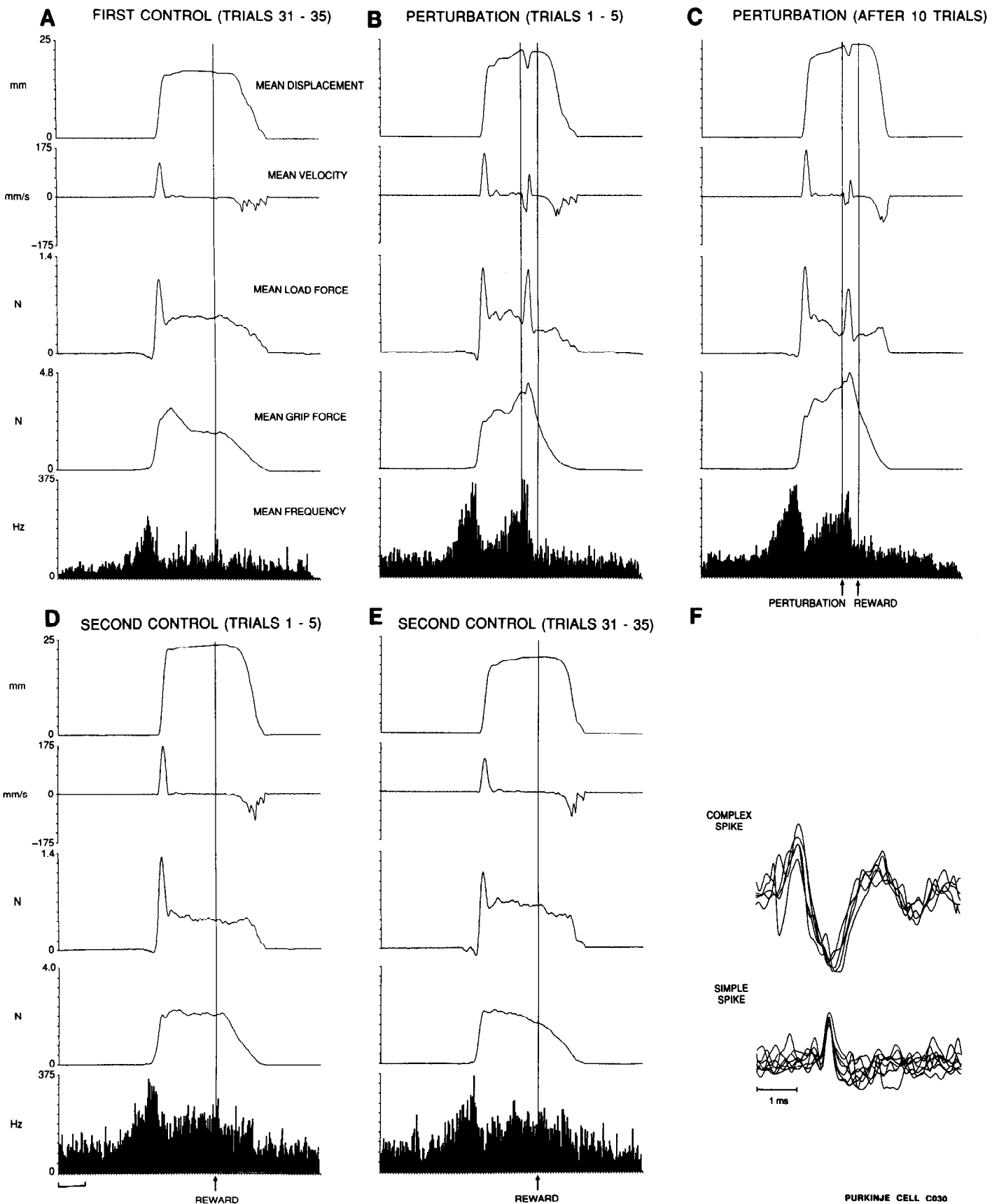


FIG. 9. Acquisition of the anticipatory response in a Purkinje cell to the perturbation and its relation to stabilizing the position of the hand. Gradual extinction of the anticipatory response after the perturbation was withdrawn is also shown. The 6 superimposed sweeps show the amplitude and form of the simple and complex spikes for this Purkinje cell.

Both the grip force and cell discharge increased monotonically up to the perturbation onset. An increase in discharge was also seen during the dynamic lifting phase of the task, but this was not included in the analysis. After the perturbation was withdrawn, the mean activity profile during the second control period diminished but did not return to its initial control level.

The gradual appearance of the preparatory responses within the block of perturbed trials is illustrated in Fig. 9 for the same cell shown in Fig. 8. The average grip force in the last five control trials of the initial unperturbed movements (Fig. 9A) is compared with that in the initial five perturbed trials (Fig. 9B) and with five subsequent trials showing minimal wrist displacement in response to the perturbation (Fig. 9C). By comparing Fig. 9, B and C, it can be seen that the reflex response was, if anything, decreased by the diminished object slip, whereas the preparatory response increased and emerged earlier on the later trials. This earlier and stronger preparatory increase in grip force probably added stiffness to the wrist and fingers, as shown by the reduced wrist displacement caused by the perturbation. Comparing Fig. 9, C and D, reveals that the reflex response was clearly locked to the stimulus, because it was immediately abolished after the perturbation ceased. In contrast to the reflex reaction, the preparatory reaction persisted temporarily in the absence of the perturbing stimulus. Fig. 9, D and E, depicts this gradual disappearance of the preparatory reaction once the perturbation had been withdrawn and shows that the modification in discharge had not completely dissipated even after 35 unperturbed trials. Although the preparatory responses rarely outlasted the perturbation to this degree, these responses usually did persist for several trials after the perturbations had ceased.

DISCUSSION

Changes in grip force and EMG activity due to the perturbation

The perturbations applied to the hand-held object in this study elicited two types of responses in the EMGs of wrist and finger muscles and in the grip force. The first we have called a reflex reaction because of its similarity to the responses seen in human subjects when perturbations were applied to a hand-held object (Cole and Abbs 1988; Johansson and Westling 1988b; Lacquaniti and Maioli 1989a,b). In agreement with these studies, an increase in grip force was observed from 50 to 100 ms after the increase in load force was applied to the fingers. These reflex reactions are at approximately the same latencies as the long-latency stretch reflexes but almost certainly involve cutaneous as well as proprioceptive afferents (Wiesendanger and Miles 1982).

In the present study, the perturbations, once introduced, were consistently repeated over a block of ≥ 20 trials. We chose to analyze the preparatory increase in grip force during the holding period when the object was stationary, because it was the most consistent adaptive response and because the difference compared with unperturbed grip forces was most obvious during the period of stable posture without movement. It is likely, however, that the monkeys were

“anticipating” the perturbation even before they grasped the manipulandum. This general expectancy or motor set produced a variety of other adaptive responses, which included an increase in the rate of grip force application, an increased lifting velocity, and a lifting to a greater height.

The onset of the preparatory response during stationary holding preceded the perturbation by an average of 450 ms. This was considerably earlier than the 150 ms reported by Johansson and Westling (1988b) or the 100 ms described by Lacquaniti and Maioli (1989b) for preparatory responses. However, it should be remembered that, in these studies, the subjects either initiated their own perturbation or they actually saw the perturbation approaching rapidly. In the present investigation, by contrast, the perturbation was applied by the experimenter and the animal was required to estimate its time of arrival 750 ms after the onset of the 1-kHz position tone (see task description). Under these somewhat less certain conditions, it is not surprising that the monkeys adopted a strategy of early preparation.

EMG changes associated with self-induced perturbations compared with unanticipated movement obstructions have been well studied (see Hugon et al. 1982; Dufossé et al. 1985). Less is known, however, about how the preparatory responses of uncued but predictable perturbations are organized. Al-Falahe and Vallbo (1988) and Lacquaniti and Maioli (1989a,b) found that predictable perturbations of the fingers produced both preparatory and reflex coactivation of antagonist muscles. These cocontractions were presumably organized by supraspinal centers to appropriately increase joint stiffness to reduce the limb displacement caused by the perturbation. The observations of Al-Falahe et al. (1990) are particularly important in this regard, because they found no evidence for enhanced fusimotor modulation in responses to anticipated perturbations despite clearly improved motor performance through learning.

Reflex reactions

The latency distribution of reflex reactions among cerebellar neurons is perhaps longer than one might expect from electrical stimulation of peripheral nerve cells (Eccles et al. 1971a,b). Recordings from the median nerve indicate that the recruitment of peripheral afferents is virtually coincident with the onset of the perturbation (Milner et al. 1991). Therefore the longer latency may be due to a conservative estimate of when the cerebellar cells changed discharge. It appears that afferent feedback activated by slip of a hand-held object has a maximal impact on cerebellar cortical neurons after a latency of ~ 40 ms. A conduction latency of 3–5 ms from the cerebellar nuclei to the motor cortex (Sasaki et al. 1976), plus a conduction time of 11 ms between the motor cortex and the intrinsic hand muscles (Lemon et al. 1987), would allow sufficient time for these neurons to participate in reflexlike reactions at 50–100 ms. These reflex latencies are similar to responses to arm perturbations recorded by Strick (1983) in the dentate and interpositus nuclei and suggest that both the cerebellar cortex and nuclei are activated nearly simultaneously by perturbations of limb segments.

Object slip exerted a powerful excitatory action on cerebellar cortical neurons. Cells with either proprioceptive or

cutaneous receptive fields were both excited by the perturbation at the same latency. Only a few (6) cells responded with inhibition. Also, regardless of whether a neuron received cutaneous or proprioceptive input, the task-related discharge was often very similar. This similarity suggests that, although the cutaneous and proprioceptive afferents do not, as a rule, converge on individual Purkinje cells, the output from both types of cells might ultimately be the same.

Potential cerebellar contribution to long-latency reflexes

How are the reflexes of the present study related to the 40- to 100-ms-latency reflexes thought to be transcortical in origin? Marsden et al. (1983) argued that the EMG response to limb perturbation was a long-latency proprioceptive reflex elicited by rapid direct activation of the motor cortex. There is additional evidence to suggest that part of the long-loop reflex is mediated by the corticomotoneuronal cells of the primate motor cortex (Cheney and Fetz 1984). However, it also seems likely that the cutaneous afferents of the hand can also give rise to reflexes with similar latencies (Cole and Abbs 1988; Garnett and Stephens 1980; Johansson and Westling 1988b; Lacquaniti and Maioli 1989a,b).

The responses to the perturbations studied here have ostensibly the same range of latencies as the transcortical reflexes. Moreover, because direct spinal afferents and the cerebello-thalamocortical afferents both terminate within the motor cortex, the cerebellum may also mediate some portion of the long-loop reflexes. Vilis and Hore (1977) reported that cooling the cerebellar nuclei in monkeys produced no effect on reflex EMG responses to limb perturbation in the 35- to 100-ms range, but the cooling may not have inactivated the entire cerebellum. In contrast, Friedmann and collaborators (1987) found that the 50- to 100-ms response to stretch of the first dorsal interosseus was increased in patients with cerebellar anterior lobe atrophy, presumably because of Purkinje cell loss, whereas these same reflexes were absent in patients with Freidreich's ataxia and associated degeneration of spinocerebellar pathways.

Although the data from the present experiment suggest that the long-loop reflexes may be partially transcerebellar as well as transcortical, it is not clear how increased Purkinje cell discharge leads to increased EMG activity. Recordings from the nuclear cell targets of the Purkinje neurons recorded here are needed to determine whether the cerebellum actually triggers the reflex response to object slip or whether it plays only a modulatory role in regulating reflex excitability.

The present study provided no evidence that the long-loop reflex could be changed by previous experience. As mentioned earlier, the neuronal responses to the perturbation were greatest on trials in which the preparatory grip force was least. By itself, the reflex increase in grip force was probably not great enough to arrest the object slip. However, feedback from the reflex may have provided an essential stimulus to the CNS needed to organize preparatory grip force increases on subsequent trials. These preparatory responses ultimately reduced the amount of slip on the fingers and decreased the stretch of the wrist muscles.

Simple and complex spikes in Purkinje cells

Both Purkinje cells and unidentified neurons within the hand representation area of the intermediate zone of the cerebellar cortex demonstrated reflex reactions and preparatory responses that were similar to one another. For the Purkinje cells, all of the responses involved modulation of the simple spike discharge, and no complex spike activity was ever seen either in preparation for or in response to the perturbation. The absence of complex spike responses was unexpected, because several studies have shown increased complex spike activity in response to a variety of perturbing conditions. Climbing fiber activity has been reported in response to perturbations or intense stimuli. For example, unexpected increases in inertial load applied to the wrist (Gilbert and Thach 1977), perturbed locomotion (Kim et al. 1987), or changes in the gain of the vestibuloocular reflex induced by retinal slip (Stone and Lisberger 1990a,b) are associated with increased complex spike activity. Increased complex spike activity has also been reported in association with the onset of visually guided tracking movements (Mano et al. 1986) or redirecting reaching movements (Wang et al. 1987).

Perhaps the perturbations applied in the present study were either not sufficiently intense (i.e., not adequate) or were too familiar because the animals had been exposed to the perturbing stimulus many times. Would complex spikes have appeared if the perturbations had been presented unexpectedly or at random amplitudes? We believe this is unlikely, because there was no evidence that complex spikes occurred more frequently on the initial perturbed trial and habituated thereafter. Moreover, random or unexpected perturbations prevent all but the most global adaptive responses and would therefore have precluded the development of a specifically adapted preparatory strategy. Another possibility, suggested by Mano et al. (1986) and Llinas (1991), is that, although complex spikes occur before movement in ensembles of Purkinje cells, the correlation between the complex spike generated by any single Purkinje cell of the ensemble and the movement or perturbation might not be obvious.

As mentioned previously, an additional number of Purkinje cells were examined in which the complex spikes could not be reliably discriminated automatically but could be detected by visual inspection. None of these further Purkinje cells appeared to have climbing fiber responses linked to the perturbation in any way.

The Purkinje cells recorded in this study all demonstrated both simple and complex spikes throughout the duration of recording. That is, no Purkinje cells that discharged exclusively complex spikes were included in this study. A second potentially important feature of the Purkinje cells recorded in the present study was the fact that all but one of the receptive field responses involved simple spike responses. In our sample, only one Purkinje cell demonstrated a climbing fiber receptive field, which was a proprioceptive response to flexing the distal phalanx of the thumb. However, no reliable complex spike discharge occurred during any phase of task performance for this cell. The absence of complex spike activity in response to slip of a grasped object in a region where the majority of Purkinje

cells had vigorous simple spike responses to the same stimulus may be an important observation. It suggests that the receptive fields for these two Purkinje cell afferents are commonly quite different.

Preparatory responses

Approximately one-quarter of the cells demonstrated an increase in activity associated with the preparatory grip force increases. Although one might expect comparable changes in any motor structure related to the hand, these responses were rare in the rostral precentral sulcus of the motor cortex (Smith et al. 1990). An approximately equal number of cerebellar cortical cells showed activity increases before and after the onset of preparatory grip force increases before the perturbation. Whether these neuronal activity changes are the result of peripheral input from the skin and muscle, or whether instead they represent anticipatory activity initiated by changes in motor strategy, remains equivocal. From the results of Strick (1983), one might have expected that Purkinje cells located in the more lateral hemisphere projecting to the dentate nucleus would show greater anticipatory responses (i.e., the influence of motor preparation) and that the more medial cells projecting to interpositus might show a greater response to peripheral stimulation. Our data did not reveal any clear distinction in this regard.

There were, however, both general and specific activity changes associated with the perturbation. As noticed in an earlier study (Espinoza and Smith 1990), marked increases in spontaneous activity were sometimes associated with an increased force or effort requirement in the ensuing grasping and lifting sequence. The perturbation induced similar increases in the spontaneous activity of the cells shown in Fig. 5 (left) and Fig. 8, which sometimes carried over into the second control period. Frequently, the preparatory activity emerged more quickly in response to the perturbation than it extinguished after the perturbation had been terminated. However, nonspecific changes in excitability could not account for all the preparatory responses. The neuron shown in Fig. 6, for example, had a specific preparatory response related to the ramplike increase in grip force before the perturbation, without any change in spontaneous activity.

Although the acquisition and extinction of preparatory increases in EMG activity and parallel changes in cerebellar cortical activity are correlated, the present study cannot determine whether the cerebellar discharge causes the preparatory increase in grip force or is a result of feedback from the increased grip force. However, there is no reason to suppose that this cerebellar activity and its associated behavior are totally unrelated, either, and it is likely that the preparatory neuronal discharge contains elements of both command and peripheral feedback signals. One of the challenges for future research will be to break down this complex signal into its constituent factors.

In spite of the uncertainty about causality, it is tempting to view the present results as supporting the role of the cerebellum in motor learning. However, to imagine that all neuronal changes related to motor learning are confined to a single region of the brain such as the cerebellum would be overly simplistic. Most current evidence suggests that mo-

tor learning involves many changes throughout the nervous system, and the cerebellum may be only one important part of a distributed process (Bloedel et al. 1992).

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