Published as: *J Neurophysiol.* 2000 December ; 84(6): 2945–2960.

Changes in the Responses of Purkinje Cells in the Floccular Complex of Monkeys After Motor Learning in Smooth Pursuit Eye Movements

Maninder Kahlon and Stephen G. Lisberger

Howard Hughes Medical Institute, Department of Physiology, Neuroscience Graduate Program, and W. M. Keck Foundation Center for Integrative Neuroscience, University of California, San Francisco, California 94143

Abstract

We followed simple- and complex-spike firing of Purkinje cells (PCs) in the floccular complex of the cerebellum through learned modifications of the pursuit eye movements of two monkeys. Learning was induced by double steps of target speed in which initially stationary targets move at a “learning” speed for 100 ms and then change to either a higher or lower speed in the same direction. In randomly interleaved control trials, targets moved at the learning speed in the opposite direction. When the learning direction was the *ON* direction for simple-spike responses, learning was associated with statistically significant changes in simple-spike firing for 10 of 32 PCs. Of the 10 PCs that showed significant expressions of learning, 8 showed changes in simple-spike output in the expected direction: increased or decreased firing when eye acceleration increased or decreased through learning. There were no statistically significant changes in simple-spike responses or eye acceleration during pursuit in the control direction. When the learning direction was in the *OFF* direction for simple-spike responses, none of 15 PCs showed significant correlates of learning. Although changes in simple-spike firing were recorded in only a subset of PCs, analysis of the population response showed that the same relationship between population firing and eye acceleration obtained before and after learning. Thus learning is associated with changes that render the modified population response appropriate to drive the changed behavior. To analyze complex-spike firing during learning we correlated complex-spike firing in the second, third, and fourth 100 ms after the onset of target motion with the retinal image motion in the previous 100 ms. Data were largely consistent with previous evidence that image motion drives complex spikes with a direction selectivity opposite that for simple spikes. Comparison of complex-spike responses at different times after the onset of control and learning target motions in the learning direction implied that complex spikes could guide learning during decreases but not increases in eye acceleration. Learning caused increases or decreases in the sensitivity of complex spikes to image motion in parallel with changes in eye acceleration. Complex-spike responses were similar in all PCs, including many in which learning did not modify simple-spike responses. Our data do not disprove current theories of cerebellar learning but suggest that these theories would have to be modified to account for simple- and complex-spike firing of floccular Purkinje cells reported here.

Introduction

Smooth pursuit eye movements provide many advantages as a model system for understanding the neural mediation of motor learning in a complex cortically mediated movement. Smooth

pursuit allows primates to track small moving visual targets. Many papers have revealed critical details of the visual-motor performance of pursuit (e.g., Lisberger and Westbrook 1985; Rashbass 1961). The basic neural circuit for pursuit is known (for recent reviews, see Keller and Heinen 1991; Lisberger et al. 1987; Tusa and Ungerleider 1988), the discharge during pursuit has been recorded at many levels of this circuit, and a paradigm has been developed for eliciting reliable motor learning. The visual signals for pursuit originate in the primary visual cortex and are relayed to the middle temporal (MT) and medial superior temporal (MST) areas of the extrastriate visual cortex and to the frontal pursuit area (FPA) in the arcuate sulcus. All three of these cortical regions have been shown, through lesion studies, to be necessary for driving pursuit eye movements (Dursteler and Wurtz 1988; Dursteler et al. 1987; MacAvoy et al. 1991; Newsome et al. 1985; Segraves et al. 1987). Each projects to the dorsolateral pontine nucleus (DLPN) (Boussaoud et al. 1992; Glickstein et al. 1980; Leichnetz 1989; Ungerleider et al. 1984), which relays its inputs to two regions of the cerebellum—the caudal vermis and the floccular complex (Brodal 1979, 1982; Glickstein et al. 1994; Langer et al. 1985; Yamada and Noda 1987). The outputs of these two regions exit the cerebellar cortex through the fastigial and vestibular nuclei respectively, where signals related to pursuit have been recorded and quantified (Fuchs et al. 1994; Lisberger et al. 1994b).

Several papers have demonstrated that learning induces appropriate increases or decreases in eye acceleration in the first 100 ms of pursuit when subjects track double steps of target speed in which initially stationary targets move at one speed for 100 ms and then change to either a higher or a lower speed (in monkeys: Kahlon and Lisberger 1996; in humans: Fukushima et al. 1996; Ogawa and Fujita 1997; other paradigms: Optican et al. 1985; van Donkelaar et al. 1994). Learning is acquired quickly and reaches an asymptote after 300 presentations of the double steps of target speed. The ability to induce learning in the first 100 ms of pursuit provides advantages for analyzing this system with behavioral and physiological approaches because the first 100 ms of pursuit is an open-loop response that probes the visuo-motor transformations in the brain before there has been time for visual feedback (Lisberger and Westbrook 1985).

Kahlon and Lisberger (1996) studied the generalization of pursuit learning to determine whether the site of learning might be localized in the sensory or motor side of the system. In one experiment, they probed learning by providing 100-ms perturbations of target velocity during different initial conditions. When learning had been induced for a given direction of pursuit, the responses to the perturbations gave evidence of learning only when perturbations in the learning direction were delivered during pursuit in the learning direction. They concluded that learning did not occur in either sensory or motor coordinates and, instead, suggested that the learned changes occur at a site or sites where both image motion and smooth eye motion were represented. Since there is no evidence for extra-retinal signals in MT related to eye movement (Ferrera and Lisberger 1997; Newsome et al. 1988), the most likely sites of learning include downstream areas such as MST, the FPA, the DLPN, the floccular complex of the cerebellum, and the pursuit pathway through the vermis and the fastigial nucleus.

In the present paper, we begin a survey of the candidate sites of pursuit learning by looking for a neuronal correlate in the floccular complex. In primates, neuronal correlates of motor learning have been found in the cerebellum for complex movements like hand flexion/ extensions against a load (Gilbert and Thach 1977) and hand tracking of visual targets (Ojakangas and Ebner 1991, 1992). However, the functional significance of these correlates was unclear because of the paucity of basic information about the neural circuitry for the behaviors. In contrast, the signal processing that creates the simple-spike (SS) and complex-spike (CS) responses of floccular Purkinje cells (PCs) during pursuit eye movements is well understood (Krauzlis and Lisberger 1996; Lisberger and Fuchs 1978; Miles and Fuller 1975; Miles et al. 1980; Noda and Suzuki 1979; Stone and Lisberger 1990a,b). At least for horizontal eye movements, there are two classes of PCs that respond during pursuit eye velocity: the

horizontal gaze velocity PCs (HGVPs) and the horizontal eye velocity PCs (HEVPs) (Lisberger and Fuchs 1978; Miles et al. 1980; Stone and Lisberger 1990a). One goal of the present paper was to ask how learning in pursuit alters the responses of these PCs and their putative homologs for vertical pursuit (Krauzlis and Lisberger 1996; Miles et al. 1980; Stone and Lisberger 1990a). We followed changes in the responses of PCs while monkeys underwent pursuit learning. Some PCs showed large and consistent SS correlates of pursuit learning while others did not even though they showed clear modulation of SS firing during the learning trials. Although the expression of learning varied among neurons, the mean population response underwent learning-induced changes in SS responses that were of the correct amplitude and direction to drive the concomitant behavioral learning.

In any neural learning system, the behavior is shaped by the interaction of many neurons at different anatomical sites, and learning may occur at several of those sites. At each individual site, learning is thought to be mediated by changes in properties such as synaptic strength. The control of properties such as synaptic strength depends on local electrical activity, often involving a comparison of presynaptic and/or postsynaptic neural activity across the synapses that are subject to change. Part of our motivation for looking first for expressions of learning in a cerebellar component of the pursuit system arises from theories of cerebellar learning. These theories hold that learning in the cerebellum involves control of synaptic weight by “long-term depression” of the synapses from parallel fibers onto PCs (Ito 1982). The depression would be controlled by local electrical signals in the form of afferent climbing fiber and parallel fiber activity (Albus 1971; Marr 1969).

CSs in the floccular complex respond to image motion (Fushiki et al. 1994; Kobayashi et al. 1998; Simpson and Alley 1974; Stone and Lisberger 1990b) and thus could encode an error signal for the pursuit system (Kitazawa et al. 1998; Stone and Lisberger 1990b). To test whether the cerebellar learning theory holds for pursuit learning, we analyzed the CS responses of PCs held through pursuit learning paradigms. We confirmed that CS firing is driven by image motion and revealed a signal with the potential to guide learning of decreased but not increased eye acceleration. However, there was no correlation between the strength of the CS “error” signal and the presence or size of the learning-related changes in SS firing. CS sensitivity to image motion itself changed during learning but was not consistently attenuated as might be predicted for a signal that would guide learning.

Methods

We recorded the activity of single units in the right floccular complex of two rhesus monkeys, weighing 6–8 kg. After initial behavioral training, the monkeys underwent surgery using sterile procedure under isoflurane anesthesia (for details, see Lisberger and Westbrook 1985). A device for head restraint was provided by using dental acrylic to attach a cylindrical receptacle to bolts implanted in the skull. To monitor eye movements with the magnetic search coil system, a coil of wire was implanted on one eye using the technique of Judge et al. (1980). After the monkeys recovered from surgery, they were trained to pursue a small spot of light and were tested on a variety of eye movement tasks, including pursuit learning, for experiments that have been reported in other publications (Kahlon and Lisberger 1996, 1999). In a second surgery, we trephined a hole in the skull with its mid-point 11 mm lateral to the midline and at the anterior-posterior level of the ear bars and implanted a recording cylinder that was tilted 26° back from the coronal plane. Monkeys received postsurgical analgesia for at least 48 h after each survival surgical procedure (0.01 mg/kg Buprenorphine HCL every 12 h).

Daily recording sessions ran for up to 4 h. For each session, the monkey was transferred from his home cage to a specially designed primate chair, and his head was held by attaching the implanted head-restraint receptacle to the ceiling of the chair. The chair was mounted on a

Contraves-Goertz turntable that provided rotation in the horizontal plane. A set of 18-in magnetic field coils was lowered and fastened to the chair. During each session we made one to three electrode tracks. At the end of each session, the recording cylinder was cleaned with saline, filled with antibiotic solution, and firmly capped. Once a week, we anesthetized the monkey with ketamine and removed the scar tissue from the dura while viewing through an operating microscope.

Visual stimuli

A 0.5° white pursuit target was created by reflecting the beam from an optical bench off an x-y pair of mirror galvanometers and projecting it onto the back of a translucent tangent screen that was 114 cm from both the monkey's eyes and the mirrors. The mirror galvanometers were controlled by a servo-system driven by commands from D/A converters in a PC computer. A 0.1° fixation spot was provided by a red LED that was imaged through a pinhole and aimed at the center of the tangent screen.

Single-unit recording

Recordings were made with glass-insulated platinum-iridium microelectrodes manufactured in our laboratory. Electrodes were positioned by an adjustable stage that attached to the recording cylinder and were driven with a hydraulic microdrive. Extracellular unit activity was passed through a standard head stage, amplified, filtered (band-pass 100 Hz to 10 kHz), and converted into trigger pulses either on-line with a hardware window discriminator (SSs) or off-line with a software discriminator (CSs).

Electrodes were introduced through the thinned dura and traveled 20–25 mm through the cerebral cortex before arriving at the cerebellum. Entry into the cerebellum was recognized by the sharp increase in background activity and by the sound of CSs over the audio monitor. Depending on our medial-lateral position, we entered into the floccular complex either directly or after traversing some cerebellar folia where units didn't respond to eye movement. Once entry into the floccular complex was confirmed by saccade-related activity in the background, we searched for eye- or gaze-velocity PCs with the behavioral paradigms described in *Experimental paradigm*. PCs were recognized by a combination of their broad negative-positive waveform, their high spontaneous SS firing rates, the moderate variability of their spontaneous discharge, and the presence of a CS detected either on the oscilloscope or the audio monitor. PCs were isolated by careful movement of the electrode and were held usually for less than 1 h but occasionally for up to 2 h. CSs were identified during recordings in 31 of our 47 PCs. The remaining 16 cells were classified as PCs because they shared all other characteristics with identified PCs. We were sometimes able to cause death of these cells at the end of a session, revealing large CSs and confirming that we probably had been recording from a PC.

Experimental paradigm

Once a unit was isolated, we characterized it with behavioral paradigms that have been used in previous studies of the floccular complex. First, the monkey tracked sinusoidal motion of the pursuit target (0.5 Hz, $\pm 10^\circ$) along the horizontal and vertical axes. A PC was selected for further analysis if its firing was modulated in phase with horizontal or vertical eye velocity. If SS firing rate was modulated preferentially for pursuit along the horizontal axis, we also tested the PC during cancellation of the VOR by moving the turntable (and monkey) exactly in phase with the sinusoidal motion of the pursuit target. If the firing rate of the PC was modulated in the same phase relative to gaze motion during horizontal smooth pursuit and cancellation of the VOR with at least 50% as large a sensitivity to head velocity as eye velocity, then it was classified as a horizontal gaze velocity PC (HGVP). PCs that lacked modulation of firing during cancellation of the VOR were termed horizontal eye velocity PCs (HEVP). Vertical eye

velocity PCs (VEVP) could include vertical gaze velocity PCs (VGVPs) because they were not tested during cancellation of the vertical VOR. Of 53 PCs held through statistically significant learning-induced changes in smooth pursuit eye movements, 6 were discarded after off-line analysis revealed modulation of SS firing of less than 10 spikes/s during sinusoidal pursuit ($n = 2$) or imperfect isolation ($n = 4$). This left a sample of 47 PCs studied during learning.

Experiments to study pursuit learning were conducted as a series of trials with the basic structure illustrated in Fig. 1A. The monkey initiated each trial by fixating a target at straight-ahead gaze. After a random interval of 300–700 ms, the fixation spot was extinguished, and a tracking target appeared at an eccentricity of 3° and moved toward the fixation point. In test trials (Fig. 1A), the target moved at a single, constant speed for at least 600 ms. In learning trials designed to decrease the eye acceleration at the initiation of pursuit (Fig. 1B), the target moved at $25^\circ/\text{s}$ for 100 ms and then changed to $5^\circ/\text{s}$ for the rest of the trial. In learning trials designed to increase eye acceleration (not shown), the target moved at $10^\circ/\text{s}$ for 100 ms and then changed to $30^\circ/\text{s}$. The “learning” speed was defined as the target speed in the initial 100 ms of target motion because it is the initial pursuit response to this speed that undergoes learning.

After each PC had been characterized with sinusoidal target motion, its responses were recorded during a sequence of blocks of target motions termed “full-test” and learning blocks. Each block contained several different target motions, which were presented as different types of trials. Full-test blocks provided target motion at 5, 10, 20, and $30^\circ/\text{s}$ in two directions along the horizontal or vertical axis, depending on the preferred-direction of the PC under study (Fig. 1C). Each full-test block consisted of 15 repetitions of all eight target motions. Learning blocks provided up to 200 learning trials designed to increase or decrease eye acceleration in one direction along the preferred axis of the PC, and 200 test trials in the opposite, “control” direction (Fig. 1C). Within each learning and full-test block, the order of presentation of target motions was randomized by repeatedly shuffling the full list of trials and then requiring the monkey to complete each trial successfully. If a trial was not completed successfully, then it was presented again at the end of the list. Monkeys normally completed 90–95% of trials on the first presentation.

Each PC was first studied during a full-test block to determine the control relationship between firing rate and target velocity in both directions along the preferred axis. In our sample, 47 PCs were followed long enough to cause statistically significant changes in eye acceleration during a learning block that increased or decreased eye acceleration in one direction along the preferred axis of the PC under study. Twenty PCs were held through 200 learning trials, while 27 PCs were lost or died late in the first learning block. For the remaining 20 PCs, we attempted to increase the amount of learning by starting another block of 200 learning trials with the same learning direction. In the four PCs that remained well isolated after 400 learning trials, a final block of full-test trials was run. Finally, two PCs remained isolated at the end of this long sequence and were recorded during a learning block in the opposite direction. Therefore we report data from 47 PCs but 49 learning paradigms.

Data acquisition and analysis

The experiments were run by a computer system that consisted of a Unix workstation (DEC Alpha) and two Pentium computers. The Unix machine provided a user interface and a file server for the data. The Pentium computers operated as “data servers” that communicated with the Unix workstation over the local area network. One of the Pentium computers controlled target motion, monitored the monkey's behavior, delivered rewards, and used A/D converters to sample voltages related to horizontal head velocity, horizontal and vertical eye position, eye velocity, and target position. Each channel was sampled at 1 kHz for experiments run as a

sequence of trials and at 500 Hz for experiments that delivered continuous sine waves of head and/or target motion. Eye-velocity voltages were obtained by analog differentiation of eye-position signals (DC to 25 Hz, -20 dB/decade). Target-position voltages were derived from the feedback from the mirror galvanometers used to move the targets. Head-velocity voltages were obtained from a tachometer attached to the drive shaft for the motor. The acceptance pulses from the hardware window discriminator used to recognize SS waveforms were timed to the nearest $10\ \mu\text{s}$.

The second Pentium computer sampled the amplified unit potentials at 50 kHz for later verification of the SS isolation and for off-line discrimination of the CS responses. Our data-analysis program provided a trigger level and multiple time and amplitude windows that were adjusted by the user to distinguish CSs from SSs. CSs were recognized by at least one and often several of the following attributes: a smaller negative deflection relative to SSs, a greater positive deflection, and oscillations in potential that lasted for up to 10 ms. Once all the CSs in a file had been identified, the computer program created a new channel of trigger pulses so that the CSs could be analyzed in exactly the same way as the SSs represented by the hardware discriminator pulse times recorded during the experiment. We isolated CSs throughout the recording for 21 of the 47 PCs in our population, and we were able to recognize but not reliably trigger CSs in an additional 10 PCs.

Data were analyzed after the experiment using a computer program that allowed inspection of the data from each trial, identification of saccades, and use of a mouse-controlled cursor to place marks for aligning subsequent averages. Usually, only 5–10% of trials showed saccades in the first 100 ms of pursuit because our step-ramp target motions were designed to produce saccadic latencies longer than 200 ms (Rashbass 1961). Trials were discarded if saccade latency was less than 200 ms. For the remaining trials, the rapid deflections of eye velocity associated with later saccades were removed and replaced by straight-line segments connecting eye velocity before and after the saccade. The beginning and end of these segments were identified either manually or by an automatic saccade-detection program that required user inspection and verification. Removing the rapid deflections of eye velocity associated with saccades served partly to make clean averages for presentation and partly to allow accurate analysis of sustained eye velocity during target motion at constant velocity. Only one PC (an HEVP) showed discernable saccade-related SS firing. Therefore we made no attempt to edit the firing rate of PCs near saccades, and saccade-related SS firing contaminated the analysis of only one PC. Further, most of our analysis was done during the first 200 ms of pursuit when all the trials retained for analysis were saccade-free.

For data from sinusoidal pursuit, 10–15 cycles of eye velocity and spike data were aligned on the zero-crossings of target position. Each cycle was divided into 64 bins, the analogue traces for the cycles were averaged, and the spikes were counted in histogram bins and converted to firing rates. The averages were then subjected to a Fourier analysis with a FFT, and the resulting DC and fundamental terms were used to compute the mean firing rate, amplitude of modulation of firing rate, and phase of firing rate relative to the stimulus. For data from pursuit of step-ramp target motion, the primary method of analysis involved determining the initiation of pursuit, measuring eye acceleration and SS firing rate in the interval from 0 to 100 ms after the initiation of pursuit, and averaging these measurements for the first and last 20 trials in the learning block. We also quantified performance during the sustained phase of pursuit by measuring eye velocity and firing rate during the interval from 300 to 400 ms after the onset of target motion. The time of initiation of pursuit was marked by the first author. This was possible because the onset of pursuit was abrupt in these experiments, and there was relatively little uncertainty about when eye velocity started to accelerate from baseline. Firing rate was calculated as the mean of the spike rate in the analysis interval minus the mean baseline firing in the interval from 100 to 300 ms before the initiation of pursuit in the same trial. The

conclusions were the same, and the actual numbers changed only slightly if we averaged the baseline firing over all 20 trials and subtracted that value from the measurements made in each interval of individual trials.

For each learning block, we measured the mean eye acceleration at the initiation of pursuit in individual trials as the change in eye velocity in the interval from 0 to 100 ms after the onset of pursuit divided by 0.1 s. We then assayed for behavioral learning by comparing the data from the first and last 20 trials in each learning block. Learning was considered “significant” and blocks were admitted for analysis of PC firing rate if *P* values were less than 0.01 in unpaired *t*-tests. Changes in firing were then analyzed in the same way and were considered significant if *P* values were less than 0.01 in unpaired *t*-tests. We also made averages of the responses to 15–20 identical target motions aligned on the initiation of pursuit to allow viewing of the time course of the responses and for demonstration purposes in our figures.

Results

We maintained isolation of 47 PCs long enough to evoke significant behavioral learning in pursuit. These included 28 HGVPs, 8 HEVPs, and 11 VEVs. During pursuit eye movements, the responses of the PCs were similar to those of previous studies. PCs had baseline SS rates ranging from 35 to 135 spikes/s. During pursuit of 0.5-Hz target motion at $\pm 31.4^\circ/\text{s}$, the amplitude of modulation of firing rate ranged from 10 to 57 spikes/s (Lisberger and Fuchs 1978; Miles et al. 1980). During step-ramp target motion, firing rate began to change with a latency similar to the onset of pursuit, approximately 100 ms after the start of target motion (Krauzlis and Lisberger 1991; Stone and Lisberger 1990a). Many PCs showed a pulse of firing at the onset of pursuit followed by a lower sustained response during sustained eye velocity near target velocity. Each PC had an *ON* direction for increased SS firing, usually ipsiversive or down, and an *OFF* direction for decreased firing, usually contraversive or up (Krauzlis and Lisberger 1996). Two HGVPs had contraversive *ON* directions.

Simple spike correlates of pursuit learning

Of 32 PCs followed during statistically significant changes in eye acceleration for learning trials in the *ON* direction, 10 showed statistically significant changes in SS firing during the initiation of pursuit. Eight of the changes were in the direction predicted for the learning paradigm used. Even though only a minority of PCs showed statistically significant SS correlates of learning, we begin by showing the time course of firing rate before and after two of the largest learning-induced changes in SS firing rate we recorded. In Fig. 2A, increases in initial eye acceleration were induced for a rightward learning direction by presenting many repetitions of learning trials consisting of rightward target velocity (dashed trace) at $10^\circ/\text{s}$ for 100 ms followed by $30^\circ/\text{s}$ for the remainder of the trial. Comparison of eye velocity before and after learning revealed substantial changes. In the first 20 learning trials (fine solid trace), eye velocity initially accelerated at a rate appropriate for target motion at $10^\circ/\text{s}$ and subsequently responded to the second step with a more rapid acceleration toward $30^\circ/\text{s}$. In the last 20 learning trials (bold trace), the initial eye velocity showed the effects of learning starting about 40 ms after the onset of pursuit (Fig. 2A, vertical arrow). Comparison of PC firing during the same trials revealed that increases in eye acceleration were accompanied by increases in the SS response (Fig. 2A). During the experiment illustrated in Fig. 2A, control trials to the left were randomly interspersed with the learning trials to the right. Comparison of the averages from the first and last 20 control trials (Fig. 2C) revealed no evidence of changes in either eye velocity or SS firing rate. Indeed, the eye velocity traces in Fig. 2C were so similar that the prelearning average is obscured by the postlearning average.

Figure 2, B and D, shows a comparable result for another PC when learned decreases in initial eye acceleration were produced for learning in the *ON* direction. Learning trials presented

rightward target velocity at 25°/s for 100 ms followed by 5°/s for the remainder of the trial. Comparison of the first 20 (fine trace) and last 20 (bold trace) learning trials shows that learning caused a substantial reduction in initial eye acceleration, again starting about 40 ms after the onset of pursuit (Fig. 2B, vertical arrow). The learned change in behavior was accompanied by a large decrease in the SS response of the PC under study. As before, learning induced by target motion in one direction had little or no effect on the eye velocity or SS response during control trials in the opposite direction (Fig. 2D). Thus when the learning direction was in the *ON* direction of the PC under study, the SS responses of these two PCs showed large correlates of pursuit learning.

Changes in transient period firing in individual PCs

Previous studies of the firing of PCs in the floccular complex during pursuit have shown that there are at least two components in SS response during pursuit with the head stationary (Krauzlis and Lisberger 1991; Stone and Lisberger 1990a). One component is reflected as a transient in firing rate at the initiation of pursuit and is thought to result from visual inputs related to image motion. The second component is reflected as sustained firing during steady eye velocity and is thought to result from corollary discharge related to smooth eye velocity.

To assess learning-induced changes in the transient responses, we measured the average SS response in the first 100 ms after the initiation of pursuit. Figure 3 summarizes these measurements by plotting the changes in the SS response of PCs as a function of the concurrent changes in eye acceleration. Data from experiments that increased and decreased eye acceleration have been plotted in the same graphs, at positive and negative values on the *x* axis, respectively. The size of the change in pursuit eye acceleration varied across PCs and is indicated by where the point plots along the *x* axis in Fig. 3.

Figure 3A plots the results when learning trials were in the *ON* direction of the PC under study. Of the 14 PCs held long enough during *ON* direction learning to cause a significant increase in eye acceleration, 10 plotted in the top-right quadrant of Fig. 3A, indicating that most showed increased transient responses. Six of the 14 PCs (filled symbols) had statistically significant changes in transient SS responses (see *METHODS*, last paragraph), and 5 of those 6 had the expected increases in response size for increases in eye acceleration. Similarly, most PCs (13/18) held long enough to cause a significant decrease in eye acceleration plotted in the bottom-left quadrant of Fig. 3A, indicating decreased transient responses. Four of the 18 PCs had statistically significant changes in SS responses (filled symbols), and 3 of these 4 effects were the expected decrease in the size of the transient responses. Of the 10 PCs that showed significant changes in SS firing, 7 were HGVPs (circles), 1 was an HEVP (triangles), and 2 were VEVs (squares). Five were recorded in each monkey, although the two with wrong-way changes were both recorded in the same monkey. The two PCs with statistically significant changes in the wrong direction were an ipsiversive HGVP and a downward VEV.

Figure 3B shows the rather different results obtained when the learning direction was the *OFF* direction for the PC under study. Of the 17 PCs held long enough to elicit statistically significant changes in eye acceleration during *OFF*-direction learning, none showed statistically significant changes in transient SS response. One might contend that there is a tiny change in response that causes the points to plot in the second and fourth quadrants of Fig. 3B, which is the expected direction of change given the direction of changes in eye acceleration. However, these changes were small and failed to reach statistical significance in spite of learning-induced changes in initial eye acceleration as large as those associated with changes in transient simple-spike responses for *ON*-direction learning.

We looked for, but did not find, any changes in either sustained eye velocity or sustained SS firing, even in the PCs that showed statistically significant changes in transient SS firing (data

not shown). The tiny change in sustained response in Fig. 2A did not achieve statistical significance, and the clear effect on the second transient in Fig. 2B occurred before the analysis interval, which is marked by the bold horizontal line labeled “S.” We also analyzed eye acceleration and SS transient and sustained responses during the control trials that provided target motion in the direction opposite to the learning trials. Although three PCs did show a statistically significant change in one measure of response, we did not find consistent changes in the SS responses of PCs during target motion in the control direction (data not shown). During the minimum of 30 min required to complete a useful experiment, baseline firing often changed. However, there was no consistent trend in the change in baseline firing for any learning paradigm nor was there a correlation between changes in baseline firing and changes in SS response. Baseline firing rate was subtracted by our measurements of SS responses and therefore did not affect our analysis.

Figure 3A documents that a minority of PCs (8 of 32) shows statistically significant changes in transient SS responses in the expected direction and that the majority of PCs show changes in the correct direction that fail to reach statistical significance. Variation among PCs in the size of the changes in SS response cannot be attributed entirely to differences in the amount of learning. Some PCs in Fig. 3A showed little or no change in response after changes in eye acceleration that were big enough to cause large and statistically significant changes in other PCs. We do not think that the wide variation among PCs in Fig. 3A is an artifact of the analysis method, a reflection of cell variability over time, or a result of recording-related damage or changing PCs during a learning block. Any of these explanations should have produced similar variation in Fig. 3B, where we instead found little variation among PCs and no statistically significant changes. We can also exclude one obvious explanation based on the well-known sensitivity to eye velocity of these PCs (e.g., Lisberger and Fuchs 1978; Miles and Fuller 1975). In our PCs, the sensitivity to eye velocity averaged about 1 spike/s per %/s, and mean eye velocity in the first 100 ms of pursuit changed by an average of +1.5 and −3%/s in learning paradigms that caused increases or decreases in eye acceleration. Thus the eye velocity sensitivity would account for a change in transient responses of 1.5–3 spikes/s, much smaller than we saw in many PCs.

Relationship between PC response properties and learning-induced changes in simple spike responses

Most accounts of the mechanisms of plasticity in the brain imply that learning should occur selectively in neurons showing changes in activity during the conditions that induce learning. Thus one might assume that PCs with larger prelearning responses during pursuit eye movements would undergo larger changes in responses. We tested this possibility by correlating the size of the learning-related change in transient SS responses with a variety of measures of the prelearning response properties of the PCs.

Figure 4, A–D, shows that large SS responses during smooth pursuit before learning were a necessary, but not sufficient, condition for obtaining statistically significant changes in transient SS responses. Figure 4, A and B, plots sustained firing rate as a function of sustained eye velocity for all 32 PCs that were followed during a statistically significant change in eye velocity with a learning direction in the PCs_{ON} direction. Data were obtained in the interval from 350 to 450 ms after the onset of pursuit for trials the first full-test block, which provided target motion at 5, 10, 20, and 30%/s in the _{ON} and _{OFF} directions. In general, PCs showed increased firing during pursuit in the _{ON} direction and either decreased firing or little change in firing during pursuit in the _{OFF} direction. The PCs that showed statistically significant effects of learning (filled symbols connected by dashed lines) all conformed with this general trend, and plotted within the same range as did the PCs that did not show significant effects of learning (open symbols connected by dashed lines). The same general impression is given by Fig. 4, C

and D, which plots the transient firing in the prelearning full test as a function of the initial eye acceleration, both measured in the interval from 0 to 100 ms after the onset of pursuit. For PCs studied during learning that decreased eye acceleration (Fig. 4C), the two cells with the largest prelearning transients (curves indicated by arrows in Fig. 4C) both had statistically significant effects of learning. However, the other PCs with statistically significant effects of learning in Fig. 4, C and D (filled symbols connected by solid lines) again plotted within the range of the PCs that failed to show significant changes during learning (open symbols connected by dashed lines). It therefore appears that the size of a PC's transient SS response during the initiation of pursuit does not predict the effect of learning on its responses.

To quantify the relationship between the size of the learning-induced changes in transient SS responses and baseline response properties, we also conducted correlation analyses. The analysis included all 32 PCs that were followed through statistically significant changes in eye acceleration with the learning direction in the *ON* direction. For each PC, we computed a normalized change in transient SS response as the absolute change in firing divided by the change in eye acceleration in the first 100 ms of pursuit. The normalized change in transient SS response was significantly correlated with the size of the prelearning transient response to target motion at 25°/s for PCs studied during learned decreases in eye acceleration ($r = 0.78$, $P < 0.01$) but not with the size of the transient response to target motion at 10°/s in the first 20 learning trials for PCs studied during learned increases in eye acceleration ($r = 0.11$, $P = 0.39$). As illustrated in Table 1, the normalized change in the transient SS response was not significantly correlated with the size of the *OFF*-direction transient response or with the sustained SS response during pursuit at 30°/s in either the *ON* or *OFF* direction for learning paradigms that cause either increases or decreases in eye acceleration.

Correlates of learning in the population simple spike response

From the perspective of downstream structures that convert floccular output into commands for smooth eye movement, the relevant issue is not how the responses of individual PCs are modified during learning but rather how the overall response of the population is modified. To address this issue, we assumed that neurons contribute to the signals that drive smooth eye movement if they respond during pursuit. We computed the mean response of the population that responded during pursuit both before and after learning, giving equal weight to those PCs that showed or did not show changes in SS firing in relation to learning. If the postlearning population mean fell on the prelearning relationship, it would imply that the population response of the PCs was appropriate to drive the postlearning eye acceleration. This outcome is entirely possible even if learning causes changes in the transient SS responses of only a subset of the PCs that show modulation of SS firing during pursuit.

To assess the prelearning relationship between the transient SS response and eye acceleration, we made measurements from the transient responses to target motions presented during the first block of full-test trials for each of the 47 PCs that showed statistically significant changes in eye acceleration during learning. A subset of these data has been presented already in Fig. 4, C and D, for PCs that were studied during learning paradigms with target motion in the *ON* direction. We then divided the PCs into four groups: learning in the *ON* direction, decreases in eye acceleration; learning in the *OFF* direction, decreases in eye acceleration; learning in the *ON* direction, increases in eye acceleration; and learning in the *OFF* direction, increases in eye acceleration. We divided the data into four groups so that the postlearning data for each learning direction and for increases versus decreases in eye acceleration would be compared with the prelearning data from the same group of PCs. For each group, we sorted the points relating firing rate to eye acceleration in the prelearning full-test trials into bins that were 50°/s² wide along the *x* axis. We then computed the mean of the transient SS response and the mean eye acceleration in each bin for each group. This yielded the four sets of connected lines (×) in Fig.

4, E and F. Figure 4E contains two sets of connected lines for PCs followed during decreases in eye acceleration for learning trials in the _{ON} and _{OFF} direction (positive and negative values of eye acceleration). Figure 4F contains two different sets of connected lines for PCs followed during increases in eye acceleration in the two directions.

Once the prelearning relationship between transient SS firing rate and eye acceleration had been established from the prelearning full-test block, we measured transient SS firing rate and eye acceleration for the first 20 and last 20 learning trials for each PC included in the analysis group (large, filled symbols in Fig. 4, E and F). For each of the four combinations of learned increases and decreases in eye acceleration and learning in the _{ON} or _{OFF} direction, the postlearning values of transient SS response as a function of eye acceleration (Fig. 4, E and F, filled triangles) plot very close to the prelearning relationship (×). The same is true for the prelearning values (Fig. 4, E and F, filled circles), as would be expected given that the connected lines and filled circles are prelearning responses to the same target velocity presented in different blocks of trials. Thus the population response of our sample of PCs changed in a way that is quantitatively consistent with the associated change in eye acceleration. We conclude that the population SS output from the floccular complex remains as appropriate to drive the eye acceleration after learning as it was before learning.

Complex spike correlates of learning

Previous recordings from PCs in monkeys have implied that CS responses are caused by image motion with a direction selectivity opposite that for SS responses (e.g., Stone and Lisberger 1990b). The present section reports the CS sensitivity to image motion in our experiments, asks whether learning trials cause any unexpected CS responses that could signal the need for learning, and looks for changes in CS activity during learning. We were able to isolate and follow CS responses of 21 PCs through statistically significant changes in eye acceleration. Of these, eight PCs were held through learned increases in eye acceleration in the _{ON} direction, eight PCs were held through learned decreases in eye acceleration in the _{ON} direction, and five PCs were held through increases or decreases in eye acceleration in the _{OFF} direction (2 increase, 3 decrease). Baseline CS firing rates ranged from 0.15 to 1.67 spikes/s, which is similar to those reported in previous studies (e.g., Stone and Lisberger 1990b).

Figure 5 shows examples of averaged CS firing rate along with averaged SS firing rate, target velocity, eye velocity, and image velocity for both learning and control trials in four representative PCs. Collectively, the four PCs show examples of responses during all four combinations of learned increases or decreases in eye acceleration and learning in the _{ON} or _{OFF} direction for the SS responses of the PC under study. For example, Fig. 5A shows responses during learning (A1) and control (A2) trials for an experiment that caused increases in eye acceleration in the SS _{ON} direction of the PC under study. Figure 5B shows averaged responses for a different PC held through learned decreases in eye acceleration in its SS _{ON} direction. The most notable CS response during the learning trials is a burst of CSs about 300–400 ms after the onset of target motion in decrease-acceleration trials (Fig. 5B1), beginning about 100 ms after a reversal of the direction of image motion from ipsiversive to contraversive (Fig. 5B1, diagonal arrow). In addition, CS firing may have been suppressed slightly starting approximately 100 ms after the onset of ipsiversive image motion in Fig. 5, A1 and B1. For _{OFF}-direction control trials (Fig. 5, A2 and B2), CS activity showed a transient increase that started about 100 ms after the initiation of contraversive image/target motion.

For comparison with the _{ON}-direction learning conditions used to obtain Fig. 5, A and B, Fig. 5 C and D summarize the CS firing of two PCs held through pursuit learning in the _{OFF} direction for the SS response. In _{ON}-direction control trials, CS firing was unmodulated or weakly suppressed (Fig. 5, C1 and D1). In _{OFF}-direction learning trials, CS firing showed a transient increase after contraversive image motion (Fig. 5, C2 and D2). Thus CS firing consistently

showed a transient increase in all OFF-direction learning and control trials (Fig. 5, A2, B2, C2, and D2) and was either weakly suppressed or unmodulated in all ON-direction learning and control trials (Fig. 5, A1, B1, C1, and D1). The only deviation from the general pattern was in Fig. 5B1, where CS firing sharply increased late in the learning trials that caused decreases in eye acceleration during ON-direction motion.

Relationship between complex spike firing and image velocity in control trials

Based on the data of Stone and Lisberger (1990b), we had expected to find that CS firing increased or decreased during the initiation of pursuit for contraversive or ipsiversive target motion, respectively. We analyzed this relationship by dividing the interval from 100 to 400 ms after the onset of target motion into three 100-ms intervals. We then paired the image motion from 0 to 100 ms after the onset of target motion with the complex spike firing 100 to 200 ms after the onset of target motion and so on. For each interval, we measured the mean CS firing rate by counting the number of CSs that occurred in the interval and converting to firing rate. We then defined the magnitude of the CS “response” for a given PC as the mean CS firing rate in each interval minus the baseline firing rate 100–300 ms before the target started to move.

The icon at the upper right of Fig. 6A shows that target motion at 25°/s produced a characteristic profile of image velocity that started at target speed (1st 100-ms interval, shown with white fill) and declined in the last 200 ms of the analysis interval (2nd and 3rd 100-ms intervals, shown with gray and black fill, respectively). For the eight PCs studied during control trials that delivered image motion in the SS OFF-direction (negative values on the x axis), CS responses were positive. CS responses were largest for the first interval, which provided image motion at 25°/s (open symbols). In the second and third intervals (gray and solid symbols), both the magnitude of OFF-direction image velocity and the size of the CS responses were smaller. For the three PCs that were studied for control trials at 25°/s in the SS ON direction (positive values on the x axis), CS responses were negative. Because of the low spontaneous rates of CS firing in most PCs, the apparently smaller responses to image motion in the SS ON direction may reflect saturation at close to zero firing rate. Different PCs were used to study responses to image motion in the SS ON and OFF direction because the only way to obtain sufficient data for analysis of the relatively infrequent CSs was to use the control direction trials interleaved with learning trials in the opposite direction.

Figure 6B shows the results of the same analysis for control target motion at 10°/s. As mentioned in the preceding text, responses to image motion in the SS OFF and ON direction were obtained from the control trials for PCs followed during learning paradigms that increased eye acceleration for target motion in the SS ON or OFF direction, respectively ($n = 8$ and $n = 2$). For image motion in the SS OFF direction, CS responses were largest for image motion at 10°/s in the first analysis interval (open squares) and were smaller in the next two analysis intervals, during which image motion was about 4°/s (gray filled squares) and 1°/s (black filled squares). For image motion in the SS ON direction, CS responses were negative in the first analysis interval, indicating decreases in firing. In the second and third analysis intervals, the responses were small and could be either positive or negative for a given image motion.

Figure 6B allows comparison of the data for control target motion at 10°/s (symbols) with the mean CS responses for target motion at 25°/s. The latter are shown by the bold dashed lines (labeled “From A”), which were obtained by averaging along both the ordinate and abscissa for the data in each interval in Fig. 6A. During the first interval of control trials with target motion at 10°/s, CS responses displayed a sensitivity to image motion with response amplitudes that surrounded the mean from the second interval of control trials with target motion at 25°/s. The comparison does not hold up as well for the second and third 100-ms intervals for target motion at 10°/s. Instead the responses were generally smaller for target motion at 10°/s than predicted based on the response for target motion at 25°/s and were not direction selective.

Relationship between complex spike firing and image velocity in learning trials

We next analyzed the relationship between CS responses and image velocity in learning trials to reveal whether there were any features of the responses that could not be explained by the same relationship already documented for control trials. The latter relationship is summarized by the means from Fig. 6, A and B, which are shown as bold dashed lines in Fig. 6, C and D. Figure 6C shows the results for learning trials that decreased eye acceleration in the SS_{ON} direction ($n = 8$ PCs). In the first analysis interval (white fill in *inset*), image velocity was 25°/s in the SS_{ON} direction and CS responses were negative in all eight PCs, as expected given the responses to control trials (Fig. 6A). In the second analysis interval (gray fill in *inset*), image velocity was small and crossed from positive to negative because of the concurrent step decrease in target velocity and the increase in eye velocity. CS responses were variable among PCs as was the sign of the mean image velocity.

In the third 100-ms analysis interval for decrease acceleration learning trials (Fig. 6C, black fill in *inset*), mean image velocity was about 5°/s in the SS_{OFF} direction. The CS response was an increase in firing as expected for this direction of image motion, but the size of the CS response was much larger than expected based on the control responses to small image velocities in the SS_{OFF} direction. The large CS response in the third analysis interval in Fig. 6C corresponds to the large, late burst of CS firing seen in the histogram of Fig. 5B1.

Figure 6D shows a similar analysis for learning trials that increased eye acceleration in the SS_{ON} direction for SSs ($n = 8$ PCs). In the first analysis interval, image motion was equal to initial target motion of 10°/s (interval shown with white fill in *inset*) and CS firing was slightly below baseline ($\square$ in Fig. 6D). In the second analysis interval (interval shown with gray fill in *inset*), image velocity was close to 25°/s because target velocity had increased and eye velocity was still small. CS responses for this interval ($\square$ in Fig. 6D), which plotted at the largest image velocity of the trio of points for each PC, were slightly negative, except for one that was strongly positive. In the third analysis interval (interval shown with black fill in *inset*), image velocity started to decline toward zero as eye velocity increased, and more CS responses were negative than positive ($\blacksquare$ in Fig. 6D). The data in Fig. 6D would not support the conclusion that the CS responses during learning trials reflect anything more than the relationship to image velocity documented during control trials.

Analysis of suitability of complex spike responses to guide pursuit learning

We now ask whether CS responses are suitable to guide pursuit learning. The answer to this question comes from a comparison of the responses to double and single steps of target velocity in the same direction. To guide learning, the CS responses should be different, indicating whether or not there has been a second step of target velocity. Figure 7A compares the CS firing in learning trials that delivered double steps of target velocity from 10 to 30°/s (open symbols) with test trials that delivered single steps to 10°/s (filled symbols). Because of the way our experiments were designed, the learning and test data come from different PCs. In each of the three analysis intervals, plotted along the x axis, the responses to test and learning trials were similar. For conditions that increase eye acceleration, CS responses do not appear to report whether or not there was a second step of target velocity.

Figure 7B suggests that CS responses could report whether or not there was a second step of target velocity for conditions that caused decreases in eye acceleration. Comparison of learning trials that delivered double steps of target velocity from 25 to 5°/s (open symbols) with test trials that delivered single steps to 25°/s (filled symbols) reveals that CS firing diverged in the third analysis interval and possibly in the second analysis interval. These intervals occur 100 and 200 ms after the interval in which eye acceleration and SS firing rate are subject to change by learning. Thus CS responses during pursuit learning for decreases in pursuit gain is in

qualitative agreement with our finding for the vestibuloocular reflex that CSs would provide consistent guidance for learning in the floccular complex if they were compared with parallel fiber inputs from 100 ms earlier (Raymond and Lisberger 1998).

Correlation of learning-related changes in simple spike firing with complex spike responses

Figure 8 compares the magnitude of CS response modulation in learning trials with the learning-related changes in SS responses of the same PCs. It contains data from the third analysis interval, in which CS firing could distinguish test trials from double-step learning trials that decrease eye acceleration. As an index of the change in transient SS responses, we again used the normalized value defined as the mean absolute change in SS response divided by the size of the learned change in eye acceleration. In experiments that caused learned increases in eye acceleration (squares), the population of PCs available for CS analysis showed a range of learning-related changes in the transient SS response. However, the size of the CS response during learning trials did not vary much among PCs and was not related to the size of the learning-related changes in the transient SS response. In experiments that caused learned decreases in eye acceleration (circles), only one of the PCs available for CS analysis showed a large change in the transient SS response. Yet the CS response of this PC (pointed to by upward arrow) was within the range of CS response from the other PCs, which showed little or no learning-related modification of transient SS responses. Thus Fig. 8 implies a double dissociation of the presence of CS responses that could guide learning and the effect (or noneffect) of learning on the SS responses of the same PCs.

Effect of learning on complex spike responses

Previous analyses of changes in PC responses during motor learning have suggested that CS responses might be present early in learning but disappear as learning proceeds (Gilbert and Thach 1977; Kim et al. 1998). To evaluate this prediction for learning in pursuit eye movements, we computed mean CS firing rate in each of the three 100-ms analysis intervals over the first and last 50 learning and control trials obtained from each PC. To control for changes in the image motion stimulus during learning, we also measured the mean image velocity in the 100 ms preceding each interval used for analysis of CS responses. Since good isolation of CS waveforms and a minimum of 100 learning trials were necessary for this analysis, we completed it for only five PCs held through decreases and seven PCs held through increases in eye acceleration in the $_{ON}$ direction for the SSs from the PC under study.

Learning caused consistent changes in the mean CS response. CS direction-selectivity and sensitivity to image motion increased in experiments that caused learned increases in eye acceleration and decreased in experiments that caused learned decreases in eye acceleration. Each point in Fig. 9 plots the mean CS responses of our sample of PCs as a function of the mean image velocity measured in the previous 100 ms in the same trials. For PCs held through increases in eye acceleration (Fig. 9A), the mean CS response to image motion in the $_{ON}$ direction was more negative in the last 50 trials (large symbols) than in the first 50 trials (small symbols). For the same PCs, the mean CS responses to image motion in the $_{OFF}$ direction underwent smaller changes but was more positive in the last than in the first 50 trials for the second and third analysis intervals. Thus for learned increases in eye acceleration, CS responses in each of the three analysis intervals showed stronger modulation by the direction of image motion in the last 50 trials than in the first 50 trials. In Fig. 9A, the responses to image motion in the $_{ON}$ direction came from learning trials in which target motion stepped from 10 to 30°/s, while the responses to image motion in the $_{OFF}$ direction came from the 10°/s control trials. As a result, the range of image velocities is quite different for image motion in the $_{ON}$ versus $_{OFF}$ direction.

For PCs held through learned decreases in eye acceleration (Fig. 9B), the mean CS response to image motion in the last 50 trials was smaller than the mean response in the first 50 trials. For image motion in the SS_{ON} and SS_{OFF} directions, the CS response in all three analysis intervals plotted closer to zero (horizontal dashed line) in the last 50 learning and control trials (large symbols) than in the first 50 trials (small symbols). The change in CS response is particularly clear in the responses to image motion in the SS_{OFF} direction; the responses are plotted at negative values of image velocity and came from control trials. Note that the CS responses in the third analysis interval of learning trials in the SS_{ON} direction result from image motion in the SS_{OFF} direction and plot at small negative values of image velocity (points marked by diagonal arrows in Fig. 9B). The large excitatory CS response, which corresponds to the late burst of CS firing in Figs. 5B1 and 6D, was much smaller in the last 50 than in the first 50 trials.

Figure 9, C and D, plots the data from the learning trials again to show how learning affected the CS responses in each of the three analysis intervals. For learning that caused increases in eye acceleration (Fig. 9C), CS firing was inhibited in each analysis interval at the end of learning (large symbols) even though it had been nearly unmodulated before learning (small symbols). For learning that caused decreases in eye acceleration (Fig. 9D), CS firing was modulated to a lesser extent after learning than before learning, showing less inhibition in the first and second intervals and less excitation in the third interval.

Discussion

It is widely accepted that the SS and CS activity of PCs arises from different anatomical sources and plays different functions in cerebellar function. In the floccular complex of the monkey, SS activity is thought to be driven by vestibular sensory inputs, eye velocity motor feedback from the brain stem, and visual inputs that signal image motion (e.g., Krauzlis and Lisberger 1991; Lisberger and Fuchs 1978; Miles and Fuller 1975; Stone and Lisberger 1990a,b). CS activity is driven by visual inputs that arise in the accessory optic system and are transmitted via the inferior olive (e.g., Maekawa and Simpson 1973; Stone and Lisberger 1990b). A popular theory of cerebellar learning implies that SS activity is involved in the processing of neural signals as they pass through the cerebellum, while CS activity acts as an error signal that guides plasticity in the cerebellar cortex (Albus 1971; Ito 1982; Marr 1969). Previous studies in other movement systems have shown that SS activity is modified in relation to learning, while CS activity occurs at the correct time to guide learning (e.g., Gilbert and Thach 1977; McCormick and Thompson 1984; Ojakangas and Ebner 1992). One goal of our study was to determine whether the activity of floccular PCs during pursuit learning fit with the earlier findings in other motor systems.

Possible causes of diversity in the SS correlate of learning

One of our principle findings is that a minority of PCs in the floccular complex show changes in transient SS responses as a result of learned modifications in the initiation of pursuit. In a well-supported model of floccular Purkinje cell firing (e.g., Miles and Fuller 1975; Stone and Lisberger 1990a), both feedforward visual inputs and feedback corollary discharge have roles in pursuit. Because the sustained responses of PCs did not change in association with learning, we suggest that learning does not induce modifications in the gain of the eye velocity feedback pathway to PCs. Instead changes in transient SS responses seem to represent a selective change in a feedforward image motion input to some PCs. Changes in transient responses, when they occur, are much too large to be attributed to the meager changes in the magnitude of eye velocity feedback during the first 100 ms of pursuit.

The neuronal correlate of pursuit learning in the floccular complex did not appear uniformly across the full population of PCs. Some PCs displayed large and statistically significant changes

in the transient SS response while others showed no changes. We think that the diversity of changes in SS responses represents genuine variation in the amount of modification of SS responses across PCs rather than temporal nonstationarities or damage during recording. The latter explanations would predict similar variation we did not see in responses during control trials or learning trials in the *OFF* direction for SS responses.

Our data raise the possibility that PCs with similar response properties might receive rather different inputs and encode different kinds of information. Some of the PCs in our population had strong transient SS responses that were related to eye acceleration before learning but did not show changes in association with learning. The transient SS responses of these PCs seemed to represent information about image motion since the same image motion evoked the same firing. Other PCs showed convincing changes in transient SS responses as eye acceleration changed and therefore seemed to encode a property of the evoked movement, such as eye acceleration. Although all the PCs in our sample had the large sustained responses reported before (e.g., Lisberger and Fuchs 1978) during sustained eye velocity, it seems plausible that the transient SS responses arise from two different, possibly overlapping, pathways through the floccular complex. These pathways could represent different sensory or motor variables using neural codes that appear very similar. Our data suggest that learning may be expressed only in the pathway that represents motor variables. There is an anatomical basis for two different, possibly overlapping, pathways that would create visually driven transient SS responses. Visual mossy inputs to the floccular complex arise from both the dorsolateral pontine nucleus and the nucleus reticularis tegmenti pontis (Gerrits et al. 1984; Glickstein et al. 1994). Pursuit learning may be associated with changes in the strength of transmission through only one of these visual input pathways.

An alternate explanation for the diversity of the expression of learning in transient SS responses would allow all PCs to receive the same inputs. Instead it would place the source of the diversity of SS representations of learning in the properties of the learning mechanism(s). In principle, mechanisms of learning that depend on postsynaptic neural activity should cause PCs with bigger transient SS responses to undergo greater changes. Our data were broadly consistent with this expectation for experiments that caused decreases in eye acceleration for target motion in the SS *ON* direction but not for experiments that caused increases in eye acceleration for target motion in the SS *ON* direction. Both of these learning conditions are associated with increases in SS firing, but they differ in that increases in eye acceleration require potentiation of neural responses while decreases require depression. Thus one possible explanation of our data would be that the mechanism of depression is activity dependent while that of potentiation is not.

Consistent correlate of learning in the population simple spike response of PCs

Some floccular PCs showed changes in association with learning while others did not. However, the mean output of the population of PCs we recorded maintained a consistent encoding of motor parameters related to pursuit eye movements. It follows that the changes in SS responses in the floccular complex, although present in only a minority of PCs, are potentially suitable to drive eye acceleration after learning. Our data do not reveal whether there are parallel pathways that transmit signals to drive eye acceleration after learning. However, the data do suggest that the floccular complex plays a role in pursuit learning in proportion to its role in normal pursuit.

We had expected to find large changes in SS responses because of the precedent provided by studies of the VOR, which compared population responses of HGVPs after large, long-term changes in the gain of the VOR that had been acquired over days or weeks (e.g., Lisberger et al. 1994b; Miles et al. 1980). When floccular PCs were followed during smaller, short-term learning in the VOR, the results were strikingly similar to those reported here (Hirata et al. 1999): not all PCs showed changes in the same direction as the general population over the

course of 1 h, and the magnitude of the changes varied widely across PCs. We do not yet know whether the SS firing of all individual PCs changes in the same direction during long-term changes in the VOR or if only a fraction of PCs show appropriate changes, as is the case in our data for pursuit learning. It would be helpful to know whether all PCs would show changes in the appropriate direction during long-term learning in pursuit.

Our results demonstrate that some amount of pursuit learning occurs in or before the cerebellar cortex of the floccular complex. Our finding that learning is associated with changes in the transient SS responses of some floccular PCs argues that the floccular complex plays a role in the initiation of pursuit, and against the prior suggestion that the vermis alone controls the initiation of pursuit (Fuchs et al. 1994). Our finding that the average output from floccular PCs provides a consistent representation of the motor command before and after learning also places some constraints on other possible sites of learning. If the vermis is making a significant contribution to pursuit, then there must be either parallel modifications in visual transmission through the vermal and floccular pathways or an additional site of learning downstream from the cerebellum if pursuit learning does not cause changes in the output from the vermis.

Do complex spikes function as error signals guiding changes in Purkinje cell output?

The cerebellar cortex has been implicated as one site of motor learning in the vestibuloocular reflex (e.g., Ito 1982; Lisberger and Sejnowski 1992; Raymond et al. 1996). By analogy, one possible locus for neural plasticity underlying pursuit learning is at the synapses from visually responsive parallel fibers onto PCs. Specific theories of neural plasticity at cerebellar sites suggest that complex spikes encode an error signal during learning: conjunction of a complex spike with parallel fiber activity should induce changes in the strength of parallel fiber inputs onto Purkinje cells (Albus 1971; Ito 1982; Marr 1969). Our data test several predictions made by this hypothesis.

PREDICTION 1: BASED SOLELY ON ITS LOCAL PARALLEL FIBER AND CS INPUTS, THE PLASTICITY MECHANISMS IN A GIVEN PC SHOULD BE ABLE TO DISTINGUISH BETWEEN LEARNING AND TEST TRIALS. Our analysis of CS responses in the floccular complex showed that a plasticity mechanism based on CS activity would be able to distinguish between test and learning trials for conditions that cause decreases but not increases in eye acceleration. The inadequacy of CS activity for guiding learned increases in eye acceleration suggests different plasticity mechanisms are used for conditions that cause learned increases in eye acceleration. Note that this conclusion is not altered by considering CS activity at different times after the interval when SS activity is modified. Indeed, CS activity is appropriate for decreasing initial eye acceleration in the intervals 100 and 200 ms after the interval when learning is measured in pursuit, in agreement with our data on the VOR (Raymond and Lisberger 1998).

PREDICTION 2: CONJUNCTION OF CS RESPONSES WITH A VISUALLY MEDIATED TRANSIENT SS RESPONSE IN PCS SHOULD BE A SUFFICIENT CONDITION TO CAUSE LEARNING-INDUCED CHANGES IN THE TRANSIENT SS RESPONSES OF PCS. All floccular PCs showed similar CS responses under conditions that decreased initial eye acceleration whether or not they expressed learning-induced changes in transient SS responses. These data argue that temporal correlation of CS responses with visually mediated parallel fiber inputs is not sufficient to induce learning, even if the CS responses are compared with parallel fiber inputs present 100 or 200 ms earlier (Raymond and Lisberger 1998). However, CS responses may still be *necessary* for learning. Perhaps a subset of responsive PCs receives as-yet-unknown signals that enable plasticity driven by CS inputs. If so, then only PCs that receive the modulatory input would express learning in their SS responses, and the diversity of changes in SS responses in our sample would be expected.

PREDICTION 3: AS LEARNING PROCEEDS, THE CS RESPONSES THAT ARE PRESENT AT THE OUTSET OF LEARNING SHOULD DIMINISH, SO THAT THE SIGNALS GUIDING LEARNING ARE REMOVED ONCE OPTIMAL PERFORMANCE HAS BEEN REESTABLISHED. In our data, CS responses did not diminish as learning proceeded for both increases and decreases in eye acceleration. Instead they were modified in parallel with the learning, becoming stronger or weaker depending on whether the learning was causing increases or decreases in the initial eye acceleration of pursuit. If one considers, however, only learned decreases in eye acceleration, when the CSs could guide learning, then the strength of the CS error signal indeed decreases during learning, as predicted for the hypothesis of cerebellar learning.

The logic given in the preceding text reveals some disagreements with the predictions of the cerebellar learning hypothesis. However, our data do not disprove the hypothesis. Instead they highlight the possibility of additional contingencies and mechanisms that would supplement the hypothesis of cerebellar learning as currently understood.

We note that our data on learning in pursuit are similar to those of Ojakangas and Ebner (1992) and Gilbert and Thach (1977) for motor learning in arm movements. These groups both found CS responses that seemed appropriate to guide learning but also found no consistent relationship between the presence or absence of the relevant CS responses and learning-related changes in SS responses. These data and ours could still be consistent with modified forms of the cerebellar learning hypothesis as outlined in the preceding text. Still some forms of learning, such as VOR learning or eyelid conditioning (Raymond et al. 1996), might depend on cerebellar plasticity while others, such as pursuit, might be precerebellar. We think it is important to remain open to the possibility that learning in cortical motor behaviors could occur in precerebellar sites and could therefore be understood best by analyzing large parts of the relevant neural system rather than by focusing on a single structure such as the cerebellar cortex.

Acknowledgements

We thank Dr. Jennifer Raymond for many helpful discussions during the course of this research and Drs. Fred Miles and Albert Fuchs for critical comments on an earlier version of the paper. S. G. Lisberger is an Investigator of the Howard Hughes Medical Institute.

This research was supported by National Institute of Neurological Disorders and Stroke Grant P01-NS-34835 (S. G. Lisberger) and by stipend support from the Boyer Fund and the University of California Regents (M. Kahlon).

References

- Albus JS. A theory of cerebellar function. *Math Biosci* 1971;10:25–61.
- Boussaoud D, Desimone R, Ungerleider LG. Subcortical connections of visual areas MST and FST in macaques. *Vis Neurosci* 1992;9:291–302. [PubMed: 1390388]
- Brodal P. The pontocerebellar projection in the rhesus monkey: an experimental study with retrograde axonal transport of horseradish peroxidase. *Neuroscience* 1979;4:193–208. [PubMed: 106327]
- Brodal P. Further observations on the cerebellar projections from the pontine nuclei and the nucleus reticularis tegmenti pontis in the rhesus. *J Comp Neurol* 1982;204:44–55. [PubMed: 6276445]
- Dursteler MR, Wurtz RH. Pursuit and optokinetic deficits following chemical lesions of areas MT and MST. *J Neurophysiol* 1988;60:940–965. [PubMed: 3171667]
- Dursteler MR, Wurtz RH, Newsome WT. Directional pursuit deficits following lesions of the foveal representation within the superior temporal sulcus of macaque monkey. *J Neurophysiol* 1987;5:1262–1287. [PubMed: 3585468]
- Ferrera VP, Lisberger SG. Neuronal responses in visual areas MT and MST during smooth pursuit target selection. *J Neurophysiol* 1997;78:1433–1446. [PubMed: 9310434]

- Fuchs AF, Robinson FR, Straube A. Participation of the caudal fastigial nucleus in smooth pursuit eye movements. I. Neuronal activity. *J Neurophysiol* 1994;72:2714–2728. [PubMed: 7897484]
- Fukushima K, Tanaka M, Suzuki Y, Fukushima J, Yoshida T. Adaptive changes in human smooth pursuit eye movement. *Neurosci Res* 1996;25:391–398. [PubMed: 8866520]
- Fushiki H, Sato Y, Miura A, Kawasaki T. Climbing fiber responses of Purkinje cells to retinal image movement in cat cerebellar flocculus. *J Neurophysiol* 1994;71:1336–1350. [PubMed: 8035218]
- Gerrits NM, Epema AH, Voogd J. The mossy fiber projection of the nucleus reticularis tegmenti pontis to the flocculus and adjacent ventral paraflocculus in the cat. *Neuroscience* 1984;11:627–644. [PubMed: 6326000]
- Gilbert PFC, Thach WT. Purkinje cell activity during motor learning. *Brain Res* 1977;128:309–328. [PubMed: 194656]
- Glickstein M, Cohen JL, Dixon B, Gibson A, Hollins M, Labossiere E, Robinson F. Corticopontine visual projections in macaque monkeys. *J Comp Neurol* 1980;190:209–229. [PubMed: 7381057]
- Glickstein M, Gerrits N, Kralj-Hans I, Mercier B, Stein J, Voogd J. Visual pontocerebellar projections in the macaque. *J Comp Neurol* 1994;349:51–72. [PubMed: 7852626]
- Hirata Y, Arikan R, Highstein SM. Analysis of the discharge pattern of cerebellar Purkinje cells before and during adaptation of the vestibulo-ocular reflex in the squirrel monkey. *Soc Neurosci Abstr* 1999;25:371.
- Ito M. Cerebellar control of the vestibulo-ocular reflex—around the flocculus hypothesis. *Annu Rev Neurosci* 1982;5:275–296. [PubMed: 6803651]
- Judge SJ, Richmond BJ, Chu FC. Implantation of magnetic search coils for measurement of eye position: an improved method. *Vision Res* 1980;20:535–538. [PubMed: 6776685]
- Kahlon M, Lisberger SG. Coordinate system for learning in the smooth pursuit eye movements of monkeys. *J Neurosci* 1996;16:7270–7283. [PubMed: 8929434]
- Kahlon M, Lisberger SG. Vector averaging occurs downstream from learning in smooth pursuit eye movements of monkeys. *J Neurosci* 1999;19:9039–9053. [PubMed: 10516321]
- Keller EL, Heinen SJ. Generation of smooth-pursuit eye movements: neuronal mechanisms and pathways. *Neurosci Res* 1991;11:79–107. [PubMed: 1656345]
- Kim JJ, Krupa DJ, Thompson RF. Inhibitory cerebello-olivary projections and blocking effect in classical conditioning. *Science* 1998;279:570–573. [PubMed: 9438852]
- Kitazawa S, Kimura T, Yin PB. Cerebellar complex spikes encode both destinations and errors in arm movements. *Nature* 1998;392:494–497. [PubMed: 9548253]
- Kobayashi Y, Kawano K, Takemura A, Inoue Y, Kitama T, Gomi H, Kawato M. Temporal firing patterns of Purkinje cells in the cerebellar ventral paraflocculus during ocular following responses in monkeys. II. Complex spikes. *J Neurophysiol* 1998;80:832–848. [PubMed: 9705472]
- Krauzlis RJ, Lisberger SG. Visual motion commands for pursuit eye movements in the cerebellum. *Science* 1991;253:568–571. [PubMed: 1907026]
- Krauzlis RJ, Lisberger SG. Directional organization of eye movement and visual signals in the floccular lobe of the monkey cerebellum. *Exp Brain Res* 1996;109:289–302. [PubMed: 8738377]
- Langer T, Fuchs AF, Scudder CA, Chubb MC. Afferents to the flocculus of the cerebellum in the rhesus macaque as revealed by retrograde transport of horseradish peroxidase. *J Comp Neurol* 1985;235:1–25. [PubMed: 3989000]
- Leichnetz GR. Inferior frontal eye field projections to pursuit-related dorsolateral pontine nucleus and middle temporal area in the monkey. *Vis Neurosci* 1989;3:171–180. [PubMed: 2487099]
- Lisberger SG, Fuchs AF. Role of primate flocculus during rapid behavioral modification of vestibulo-ocular reflex. I. Purkinje cell activity during visually guided horizontal smooth-pursuit eye movements and passive head rotation. *J Neurophysiol* 1978;41:733–763. [PubMed: 96225]
- Lisberger SG, Morris EJ, Tychsen L. Visual motion processing and sensory-motor integration for smooth pursuit eye movements. *Annu Rev Neurosci* 1987;10:97–129. [PubMed: 3551767]
- Lisberger SG, Pavelko TA, Brontë-Stewart HM, Stone LS. Neural basis for motor learning in the vestibuloocular reflex of primates. II. Changes in the responses of horizontal gaze velocity Purkinje cells in the cerebellar flocculus and ventral paraflocculus. *J Neurophysiol* 1994b;72:954–973. [PubMed: 7983548]

- Lisberger SG, Pavelko TA, Broussard DM. Responses during eye movements of brain stem neurons that receive monosynaptic inhibition from the flocculus and ventral paraflocculus in monkeys. *J Neurophysiol* 1994a;72:909–927. [PubMed: 7983546]
- Lisberger SG, Sejnowski TJ. Motor learning in a recurrent network model based on the vestibulo-ocular reflex. *Nature* 1992;360:159–161. [PubMed: 1436091]
- Lisberger SG, Westbrook LE. Properties of visual inputs that initiate smooth pursuit eye movements in monkeys. *J Neurosci* 1985;5:1662–1673. [PubMed: 4009252]
- MacAvoy MG, Gottlieb JP, Bruce CJ. Smooth pursuit eye movement representation in the primate frontal eye field. *Cereb Cortex* 1991;1:95–102. [PubMed: 1822728]
- Maekawa K, Simpson JJ. Climbing fiber responses evoked in vestibulo-cerebellum of rabbit from the visual system. *J Neurophysiol* 1973;36:649–666. [PubMed: 4541327]
- Marr D. A theory of cerebellar cortex. *J Physiol (Lond)* 1969;202:437–470. [PubMed: 5784296]
- McCormick DA, Thompson RF. Neuronal responses of the rabbit cerebellum during acquisition and performance of a classically conditioned nictitating membrane-eyelid response. *J Neurosci* 1984;4:2811–2822. [PubMed: 6502205]
- Miles FA, Fuller JH. Visual tracking and the primate flocculus. *Science* 1975;189:1000–1002. [PubMed: 1083068]
- Miles FA, Fuller JH, Braitman DJ, Dow BM. Long-term adaptive changes in primate vestibuloocular reflex. III. Electrophysiological observations in flocculus of normal monkeys. *J Neurophysiol* 1980;43:1437–1476. [PubMed: 6768853]
- Newsome WT, Wurtz RH, Dursteler MR, Mikami A. Deficits in visual motion processing following ibotenic acid lesions of the middle temporal visual area of the macaque monkey. *J Neurosci* 1985;5:825–840. [PubMed: 3973698]
- Newsome WT, Wurtz RH, Komatsu H. Relation of cortical areas MT and MST to pursuit eye movements. II. Differentiation of retinal from extraretinal inputs. *J Neurophysiol* 1988;60:604–620. [PubMed: 3171644]
- Noda H, Suzuki DA. The role of the flocculus of the monkey in fixation and smooth pursuit eye movements. *J Physiol (Lond)* 1979;294:335–348. [PubMed: 117101]
- Ogawa T, Fujita M. Adaptive modifications of human postsaccadic pursuit eye movements induced by a step-ramp-ramp paradigm. *Exp Brain Res* 1997;116:83–96. [PubMed: 9305817]
- Ojakangas CL, Ebner TJ. Scaling of the metrics of visually-guided arm movements during motor learning in primates. *Exp Brain Res* 1991;85:314–323. [PubMed: 1893982]
- Ojakangas CL, Ebner TJ. Purkinje cell complex and simple spike changes during a voluntary arm movement learning task in the monkey. *J Neurophysiol* 1992;68:2222–2236. [PubMed: 1491268]
- Optican LM, Zee DS, Chu FC. Adaptive responses to ocular muscle weakness in human pursuit and saccadic eye movements. *J Neurophysiol* 1985;54:110–122. [PubMed: 4031979]
- Rashbass C. The relationship between saccadic and smooth tracking eye movements. *J Physiol (Lond)* 1961;159:326–338. [PubMed: 14490422]
- Raymond JL, Lisberger SG. Neural learning rules for the vestibulo-ocular reflex. *J Neurosci* 1998;18:9112–9129. [PubMed: 9787014]
- Raymond JL, Lisberger SG, Mauk MD. The cerebellum: a neuronal learning machine? *Science* 1996;272:1126–1131. [PubMed: 8638157]
- Segraves MA, Goldberg ME, Deng SY, Bruce CJ, Ungerleider LG, Mishkin M. The role of striate cortex in the guidance of eye movements in the monkey. *J Neurosci* 1987;7:3040–3058. [PubMed: 3668615]
- Simpson JJ, Alley KE. Visual climbing fiber input to rabbit vestibulo-cerebellum: a source of direction-specific information. *Brain Res* 1974;82:302–308. [PubMed: 4441896]
- Stone LS, Lisberger SG. Visual responses of Purkinje cells in the cerebellar flocculus during smooth-pursuit eye movements in monkeys. I. Simple spikes. *J Neurophysiol* 1990a;63:1241–1261. [PubMed: 2358872]
- Stone LS, Lisberger SG. Visual responses of Purkinje cells in the cerebellar flocculus during smooth-pursuit eye movements in monkeys. II. Complex spikes. *J Neurophysiol* 1990b;63:1262–1275. [PubMed: 2358873]

- Tusa RJ, Ungerleider LG. Fiber pathways of cortical areas mediating smooth pursuit eye movements in monkeys. *Ann Neurol* 1988;23:174–183. [PubMed: 3288083]
- Ungerleider LG, Desimone R, Galkin TW, Mishkin M. Subcortical projections of area MT in the macaque. *J Comp Neurol* 1984;223:368–386. [PubMed: 6323553]
- van Donkelaar P, Fisher C, Lee RG. Adaptive modification of oculomotor pursuit influences manual tracking responses. *Neuroreport* 1994;5:2233–2236. [PubMed: 7881034]
- Yamada T, Noda H. Afferent and efferent connections of the oculomotor cerebellar vermis in the macaque monkey. *J Comp Neurol* 1987;265:224–241. [PubMed: 3320110]

FIG. 1.

Basic structure of test and learning trials. *A*: a test trial. The pursuit target moved at 20°/s for the entire duration of the trial. *B*: a learning trial that caused decreases in eye acceleration. The pursuit target began to move at 25°/s, but after 100 ms decelerated suddenly to 5°/s. *A* and *B*, *top*: superimposed eye (E) and target (T) position; *bottom*: superimposed eye (E) and target (T) velocity as a function of time. — and - - -, show eye and target motion, respectively. Data are shown starting 300 ms before the pursuit target (T) began to move. *C*: schematic showing how the 2 different blocks of trials were created. Icons show the target velocity trajectories we used. Full-test blocks provided target motion at 5, 10, 20, and 30°/s in both the learning and control directions. Learning blocks for increasing eye acceleration consisted of double steps

to 10 and then 30°/s in the learning direction combined with target motion at 10°/s in the control direction. Learning blocks for decreasing eye acceleration consisted of double steps to 25 and then 5°/s in the learning direction combined with target motion at 25°/s in the control direction.

FIG. 2.

Simple spike responses of 2 Purkinje cells (PCs) from the minority that showed large correlates of learning when held through significant changes in eye acceleration. *Top*: target (dashed) and eye (solid) velocity. *Bottom*: the outlines of poststimulus time histograms (PSTHs) of simple spike (SS) firing. Bin width was 16 ms. Light and bold traces depict averages taken from the first and last 20 learning trials. *A* and *C*: SS responses of a PC held through increases in eye acceleration in the *ON* direction. *B* and *D*: SS responses of a PC held through decreases in eye acceleration in the *ON* direction. *A* and *B*: learning trials in the *ON* direction. *C* and *D*: control trials in the *OFF* direction. In *C*, the prelearning average of eye velocity is obscured by the bold, postlearning trace. In *A* and *B*, the bold horizontal bars labeled T and S indicate the intervals used for analysis of transient and sustained responses.

FIG. 3.

Quantitative analysis of learning-related changes in the transient SS response. Each point plots the response of 1 PC and shows its change in SS transient firing as a function of the concomitant change in eye acceleration at the initiation of pursuit. *A*: learning trials in the *ON* direction of the PCs under study ($n = 32$). *B*: learning trials in the *OFF* direction ($n = 17$). Different symbols show data from PCs with different response properties: ● and ○, HGVPs; ▲ and △, HEVPs; ■ and □, VEVs. ●, ▲, and ■ represent PCs that showed significant changes ($P < 0.01$) in SS responses. Data are from 47 PCs held through statistically significant changes in eye acceleration ($P < 0.01$) in 2 monkeys. Vertical arrows show the points from the 2 PCs used to create the example traces in Fig. 2.

FIG. 4.

Analysis of mean change in SS response across the population of PCs. *A* and *D*: relationship of sustained firing rate to eye velocity for PCs held through learned decreases (*A*) or increases (*B*) in eye acceleration. Measurements were made in the interval from 350 to 450 ms after the onset of pursuit for targets moving at 5, 10, 20, and 30°/s. *B* and *E*: prelearning relationship of transient SS response to eye acceleration for PCs held through learned decreases (*C*) or increases (*D*) in eye acceleration. Measurements were made in the 1st 100 ms after the onset of pursuit for targets moving at 5, 10, 20, and 30°/s. In *A–D*, positive values of eye acceleration indicate tracking in the ON direction of the PC under study. Each set of connected points shows data for an individual PC; each PC contributed separate sets of points for ON- and OFF-direction

tracking. Different symbols indicate PCs with different response properties: ● and ○, HGVPs; ▲ and △, HEVPs; ■ and □, VEVs. Filled symbols and solid lines show data from PCs that had statistically significant changes in transient SS responses. Vertical arrows point to data from the 2 PCs with the largest prelearning transient responses during the initiation of pursuit. *E* and *F*: comparison of mean relationship between eye acceleration and transient SS response before and after learning that caused decreases (*E*) or increases (*F*) in eye acceleration in the _{ON} direction of the PC under study. Points plotted as ×'es show the mean transient SS responses before learning, averaged by dividing all the points in *A* and *B* into 50°/s² bins. In each graph, the ×'es plotted at positive values of eye acceleration were taken from PCs that were studied during learning in the _{ON} direction. The ×'es plotted at negative values were obtained from PCs studied during learning in the _{OFF} direction. ● and ▲ plot the mean transient SS response as a function of eye acceleration from the 1st 20 (●) and last 20 (▲) learning trials. Error bars represent SEs of the mean.

FIG. 5.

Typical complex-spike (CS) and SS firing profiles in learning and control trials. Each panel shows data for 1 kind of target motion, and each vertical pair of panels shows paired learning and control responses for 1 PC. *A1*: learning trials that increase eye acceleration in the SS_{ON} direction for PC 1. *B1*: learning trials that decrease eye acceleration in the SS_{ON} direction for PC 2. *C1*: control trials in the SS_{ON} direction for PC 3. *D1*: control trials in the SS_{ON} direction for PC 4. *A2*: control trials in the SS_{OFF} direction for PC 1. *B2*: control trials in the SS_{OFF} direction for PC 2. *C2*: learning trials that increase eye acceleration in the SS_{OFF} direction for PC 3. *D2*: learning trials that decrease eye acceleration in the SS_{OFF} direction for PC 4. From top to bottom, the traces show: superimposed target velocity (---) and eye velocity (—) as a function of time; image velocity, computed as target velocity minus eye velocity; PSTH of CS firing in 20-ms bins; and PSTH of SS firing in 20-ms bins. Upward deflections of the traces show SS_{ON} direction eye, target, or image motion. For PCs 1–4, data were averaged over 200, 150, 50, and 150 trials, respectively. The arrow in *B1* shows the time of transition from ipsiversive to contraversive image motion.

FIG. 6.

Quantitative analysis of complex spike responses as a function to image motion in learning and control trials. Data are taken from the 21 PCs for which we obtained well-isolated complex spikes. Each point plots CS responses for a single PC in a 100-ms interval as a function of the mean image velocity 100 ms earlier. *A*: responses during steps of target velocity to 25°/s. *B*: responses during steps of target velocity to 10°/s. *C*: responses during trials that cause learned decreases in eye acceleration for target motion in the SS_{ON} direction of the PC under study. *D*: responses during trials that cause learned increases in eye acceleration for target motion in the SS_{ON} direction of the PC under study. Different symbols show different 100-ms analysis intervals: □ and ○, 1st 100-ms analysis interval; ▣ and ▤, 2nd analysis interval; ■ and ●, 3rd analysis interval. Circles and squares show data taken from experiments that caused learned decreases or increases in eye acceleration. The icons in the top right corner of each graph show examples of the time course of image motion profiles that precede the plotted CS responses by 100 ms, with the 3 100-ms analysis intervals indicated by the shading under the traces. Bold dashed line in *B* shows the mean CS responses for target motion at 25°/s. Bold dashed lines in *C* and *D* show the mean CS responses for target motion at 25 and 10°/s, respectively. *A* and *B* show data from SS_{OFF}-direction control trials. *C* and *D* show data from SS_{ON}-direction learning trials. As indicated by the arrows on the x-axis label, image velocity in the ON and OFF direction for SS responses is plotted at positive and negative values on the x axis.

FIG. 7.

Comparison of CS responses in test vs. learning trials in intervals that followed by 0, 100, and 200 ms the time of learning in SS firing and eye acceleration. *A*: learning-induced increases in eye acceleration. *B*: learning-induced decreases in eye acceleration. ○, data from learning trials that presented double-steps of target velocity. ●, data from test trials that presented single steps of target velocity in the same direction as the learning trials. Intervals 1–3 along the *x* axis were defined as in Fig. 6. Data for double-step (learning) and single-step (test) trials were obtained from different PCs.

FIG. 8.

Relationship between complex spike modulation in 3rd analysis interval learning trials and degree of learning-induced change in SS responses in the same PCs. Each point shows data for 1 PC and plots the CS response as a function of the normalized change in SS response. The value on the x axis was computed as the change in SS response divided by the change in eye acceleration. ○, data obtained during learning-induced decreases in eye acceleration ($n = 8$). ■, PCs recorded during learning-induced increases in eye acceleration ($n = 8$). Vertical arrow shows the 1 PC in this sample that showed statistically significant changes in the transient SS responses for learned decreases in eye acceleration.

FIG. 9.

Changes in CS sensitivity to image motion during learning. *A* and *B*: CS response is plotted as a function of image velocity in the previous 100 ms for learned increases ($n = 7$) or decreases ($n = 5$) in eye acceleration ($n = 7$). Small and large symbols plot averages for the 1st and last 50 learning or control trials. Different fillings show data from different analysis intervals: open symbols, 1st 100-ms analysis interval; gray symbols, 2nd analysis interval; filled symbols, 3rd analysis interval. As indicated by the arrows on the x axis, positive and negative values of image velocity indicate motion in the ON and OFF directions for SS responses. The arrows in *B* mark matching data points showing CS responses for the 3rd 100-ms analysis interval in the first and last 50 trials of experiments that caused learning induced increases in eye acceleration in the SS ON direction. The icons in the top right corner of each graph show examples of the time course of image motion profiles that precede the plotted CS responses by 100 ms, with the 3 100-ms analysis intervals indicated by the shading under the traces. *C* and *D*: data for learning trials are plotted as a function of the interval of CS firing measurement. These graphs show the effects of learning on CS responses that followed by 0, 100, and 200 ms the interval where learning was measured in SS responses and eye acceleration. Small and large symbols show data from first and last 50 learning trials. In all 4 graphs, error bars depict one standard error of the mean.

TABLE 1

Correlation coefficients and statistical significance from statistical analysis of relationship between amount of change in transient SS response and other parameters of prelearning SS responses

	Increase Acceleration		Decrease Acceleration	
	<i>r</i>	<i>P</i>	<i>r</i>	<i>P</i>
Transient response				
ON direction	0.11	0.39	0.78	0.0001
OFF direction	-0.11	0.35	-0.37	0.11
Sustained response				
ON direction	0.35	0.17	0.25	0.23
OFF direction	-0.11	0.36	0.05	0.38

The four rows of data show the correlations for four different measures of prelearning simple spike (SS) responses during pursuit. The two columns of *r* and *P* values come from separate experiments using learning paradigms that increased or decreased initial eye acceleration. The transient responses were measured in the interval from 0 to 100 ms after the onset of pursuit in the first 20 learning trials in the learning block. The sustained response were measured in the interval from 350 to 450 ms after the onset of pursuit for target motion at 30°/s in the full-test block of trials that preceded each learning block.