Role of the Cerebellar Flocculus Region in Cancellation of the VOR During Passive Whole Body Rotation

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Belton, Timothy and Robert A. McCrea. Role of the cerebellar flocculus region in cancellation of the VOR during passive whole body rotation. J Neurophysiol 84: 1599–1613, 2000. A series of studies were carried out to investigate the role of the cerebellar flocculus and ventral paraflocculus in the ability to voluntarily cancel the vestibuloocular reflex (VOR). Squirrel monkeys were trained to pursue moving visual targets and to fixate a head stationary or earth stationary target during passive whole body rotation (WBR). The firing behavior of 187 horizontal eye movement–related Purkinje (Pk) cells in the flocculus region was recorded during smooth pursuit eye movements and during WBR. Half of the Pk cells encountered were eye velocity Pk cells whose firing rates were related to eye movements during smooth pursuit and WBR. Their sensitivity to eye velocity during WBR was reduced when a visual target was not present, and their response to unpredictable steps in WBR was delayed by 80–100 ms, which suggests that eye movement sensitivity depended on visual feedback. They were insensitive to WBR when the VOR was canceled. The other half of the Purkinje cells encountered were sensitive to eye velocity during pursuit and to head velocity during VOR cancellation. They resembled the gaze velocity Pk cells previously described in rhesus monkeys. The head velocity signal tended to be less than half as large as the eye velocity–related signal and was observable at a short (~40 ms) latency when the head was unpredictably accelerated during ongoing VOR cancellation. Gaze and eye velocity type Pk cells were found to be intermixed throughout the ventral paraflocculus and flocculus. Most gaze velocity Pk cells (76%) were sensitive to ipsilateral eye and head velocity, but nearly half (48%) of the eye velocity Pk cells were sensitive to contralateral eye velocity. Thus the output of the flocculus region is modified in two ways during cancellation of the VOR. Signals related to both ipsilateral and contralateral eye velocity are removed, and in approximately half of the cells a relatively weak head velocity signal is added. Unilateral injections of muscimol into the flocculus region had little effect on the gain of the VOR evoked either in the presence or absence of visual targets. However, ocular pursuit velocity and the ability to suppress the VOR by fixating a head stationary target were reduced by approximately 50%. These observations suggest that the flocculus region is an essential part of the neural substrate for both visual feedback–dependent and nonvisual mechanisms for canceling the VOR during passive head movements.

INTRODUCTION

The vestibulo-ocular reflex (VOR) is a central pattern generator that rotates the eyes so that visual images of interest can be stabilized on the retina when the head moves. The vestibular signals that are the origin of the VOR eye movement commands carried by central VOR pathways must be modified or supplemented as the behavioral context of the reflex changes. One circumstance in which the VOR must be modified is when a visual image remains stationary with respect to the head while the head moves in space. The correspondence between image and head motion can occur in natural circumstances when the head is passively perturbed in space during either movements of the body or movements of the substrate on which it stands. An example of this is when a squirrel monkey looks at an object on the moving tree branch on which it stands. A second circumstance in which the VOR must be canceled occurs when voluntary head movements contribute to gaze shifts made to pursue moving visual targets. In that circumstance the VOR must be canceled in order for the gaze shift to progress.

One way for the VOR to be suppressed during passive head movements is through programming a predictive pursuit command that also functions to cancel the VOR (Barnes 1988; Barnes and Eason 1988; Lanman et al. 1978). A second method would be to produce a parametric reduction in the oculomotor signals of the VOR (Cullen et al. 1991; Robinson 1982; Tomlinson and Robinson 1981). Squirrel monkeys use both parametric and predictive mechanisms to cancel the VOR (Cullen et al. 1991), and different classes of secondary VOR neurons are preferentially related to each mechanism (Cullen and McCrea 1993; Cullen et al. 1993). During VOR cancellation the signals carried by position-vestibular-pause (PVP) neurons and secondary vestibular neurons whose firing behavior is related to smooth pursuit eye movements, the eye–head velocity (EHV) neurons, are modified. However, the changes in PVP head movement sensitivity do not depend on visual following or smooth pursuit signals, while the changes in EHV activity apparently do depend on pursuit inputs (Cullen et al. 1993). The two mechanisms for cancellation of the VOR possibly reflect two general mechanisms for changing VOR gain. One involves the input of visual feedback or pursuit signals to VOR pathways (Barnes 1993; Collins and Barnes 1999; Lisberger 1990; Lisberger et al. 1981). The second mechanism involves the removal or addition of vestibular reafferent signals as determined by behavioral context (Cullen and McCrea 1993; McCrea et al. 1996). The cerebellar flocculus and contiguous regions of the ventral paraflocculus are a critical part of the central neural subcortical and pharmacological systems that determine the gain of the VOR.
METHODS

The methods used for recording and analyzing eye movements and single-unit activity in squirrel monkeys were similar to those that have been described previously (Chen-Huang and McCrea 1999; Gdowski and McCrea 1999).

Surgical preparation

Three adult squirrel monkeys were prepared for recording both single-unit activity and eye movements. Surgeries were carried out under aseptic conditions on animals anesthetized with pentobarbital sodium (20 mg/kg ip, supplemented as necessary with 1–2 mg/kg iv). A woven coil of fine, Teflon-coated wire (Cooner) was sutured to the sclera of one eye for the recording of eye movements using the magnetic search coil technique. A small stainless steel stud was affixed to the occipital bone with dental acrylic for restraining the head in the plane of the horizontal semicircular canals. A Plexiglas well was fixed onto the parietal bone for the placement of microelectrodes, and a metal reference pin was permanently affixed to the skull adjacent to the probe insertion site.

Experimental recording conditions

During experiments animals were seated in a primate chair atop a vestibular turntable (Inland 832). A harness was placed over the animal’s shoulders and in front of the trunk to inhibit trunk and arm-raising movements. The implanted stud was attached to a rod that allowed the head to move in the plane of the horizontal semicircular canals. The rotational axis of the rod was coincident with the rotational axis of the turntable and was positioned within 5 mm of the C1–C2 axis of head rotation at the level of the external auditory meatus. The rod rotated within a low-friction ball bearing assembly fixed to the table and had a universal joint that permitted small postural adjustments. The head could be fixed to the turntable by disabling the universal joint and attaching a block that prevented angular rotation of the rod. The experiments described in this paper concern recordings obtained when the head was restrained from moving.

The monkeys were trained to fixate and pursue a small visual target (0.5 W HeNe laser, <0.2° diam) projected onto a cylindrical projection screen 90 cm distant from the monkey. The background presented by the screen was not an effective optokinetic stimulus during constant velocity turntable rotations (30–60°/s). The target movement was produced with a pair of galvanometer-controlled mirrors mounted on the turntable. The animals were rewarded for fixation of the target using a sweetened milk mixture according to a variable reinforcement schedule. After training, the monkeys were able to produce on-demand performance for sustained periods of 5 h or more, three to four times per week.

Two of the monkeys were also used in the head-free pursuit experiments described in the accompanying paper (Belton and McCrea 2000). The training paradigms and rewards were identical in the monkeys in the head-free and head-restrained conditions: the monkeys were rewarded when the angular position of the right eye was within 2° of the target. It is conceivable that monkeys attempted to follow targets with head movements when their heads were restrained; however, any such effort was unnecessary and unrewarded. One monkey was not used in head-free experiments until after recordings in the flocculus had been completed. The Purkinje cell responses recorded in that animal were similar to the responses observed in the other two animals when the head was restrained.

Eye and head movement recording

Eye movements were measured using a magnetic search-coil system (40 cm diam, Neurodata Instruments). The eye position signal was calibrated by assuming that the gain of the VOR recorded during fixation of an earth stationary target was unity. The position of the head was monitored with a second search coil. Behavioral monitoring, data acquisition, and stimulus generation were controlled with a personal computer. Turntable, target, gaze, and head signals were low-pass filtered (5–10 kHz) and sampled (2–500 Hz) at 16-bit resolution with a Cambridge Electronics 1401 data acquisition system.
Single-unit recording techniques

Single-unit recordings were made using varnish-coated tungsten microelectrodes (4–7 MΩ impedance) introduced into the FLR through a guide tube. The guide tube was mounted onto the slave cylinder of a hydraulic microdrive (Trent Wells). A manual micro-manipulator was used for moving the guide tube covering the electrode approximately 0.7 mm into the cerebellum. The electrode was then advanced into the FLR using the hydraulic microdrive.

Single-unit potentials were conventionally amplified (Dagan 2400) and filtered (band-pass 300 Hz to 8 kHz). Action potentials were discriminated with a window discriminator (BAK Electronics) that triggered an event channel of the Cambridge Electronics 1401 system at 0.1 ms resolution. These events were subsequently converted into values of instantaneous firing frequency corresponding to each A/D sample.

Histology and location of recording sites

Before recording from the FLR, an attempt was made to estimate the location of the cerebellar tentorium and the rostral and lateral extent of the ventral paraflocculus. Once these were ascertained, a region extending 4 mm caudal from the rostral end of the paraflocculus and 2–3 mm medial from the lateral edge of the ventral paraflocculus was systematically explored. On each track the location of the ventral aspect of the ventralmost folium, and, when encountered, of the vestibular nerve were noted. These landmarks were used as aids in reconstructing recording probes. In two monkeys the position of a microelectrode tract was confirmed histologically.

The most salient anatomical landmark during recording was the presence of VIIIth nerve axons 300–500 μm ventral to the cerebellum cortex. In squirrel monkeys the position of the vestibular nerve was determined histologically.

Single-unit recordings explored were bounded dorsally by an area approximately 700 μm ventral to the tentorium, and laterally by the petrosal bone. No Purkinje cells whose firing behavior was related to eye or head movement were encountered >2.25 mm medial to the estimated lateral edge of the ventral paraflocculus. Ventrally the FLR was bounded by bone and, most medially, the cochlear nuclei, where acoustically responsive, smaller neurons could be encountered. The results of our reconstructions suggest that only the caudal-medialmost folium of the flocculus was not explored (see Anatomical location of horizontal Pk cells). No effort was made to explore the dorsal paraflocculus.

Single-unit recordings

Isolated single units (signal-to-noise ratio was typically >3:1) were tested for responsiveness during horizontal smooth pursuit of a visual target moved sinusoidally at 0.5 Hz, 40°/s peak velocity, and during suppression of the VOR using the same stimulus. Units that were apparently unresponsive to horizontal pursuit and turntable rotation were often tested for their responsiveness during vertical pursuit. Units that were sensitive to vertical pursuit were usually not sensitive to horizontal pursuit and were not studied further.

Most Pk cells were easily isolated and discriminated from background activity. No recordings were obtained from poorly discriminated units or from recordings that included more than one unit.

EXPERIMENTAL PROTOCOLS. The responses of each Pk cell were studied using the following four paradigms.

1) Sinusoidal smooth pursuit (0.5 Hz, 40°/s peak target velocity)
2) VOR. Fixation of an earth stationary target during whole body rotation (WBR; 0.5 Hz, 40°/s peak velocity)
3) VOR cancellation. Fixation of an head stationary target during WBR (0.5 Hz, 40°/s)
4) Spontaneous eye movements in the absence of a visual target in dimly lit room
The following experiments were carried out in some Pk cells.

5) VOR without a target present. This was done either in the dark or in the light with the monkey facing the featureless projection screen

6) Procedures 1–3 above at stimulus frequencies of 1.0 or 2.3 Hz.

7) Steps in turntable acceleration, without a target present (400°/s², 100 ms)

8) Steps in turntable acceleration during fixation of visual target (400°/s², 100 ms)

9) Steps in turntable acceleration during ongoing VOR cancellation [step change to ∼400°/s² from a background WBR acceleration of 20–80°/s² (see Cullen et al. 1991)]

The firing rate of most units was also studied using paradigms when the head was free to move in the yaw plane. The results of those experiments are described in the accompanying paper.

Inactivation of the FLR using muscimol

After extensive recordings had been made from single units in the FLR, injections of muscimol were made in the estimated center of the region that contained the highest concentration of horizontal eye movement–related Pk cells. In each case this site was dorsal and slightly anterior to the VIIIth nerve. In one of the monkeys, the cannula used for muscimol injection was fixed in place immediately on completion of the behavioral experiment, and the animal was killed by transcardial perfusion of a fixative. Histological reconstruction showed that the tip of the cannula was located in the ventral-most folium of the ventral paraflocculus approximately 1 mm rostral to the flocculus.

After confirming the location by recording single-unit activity, the recording microelectrode was replaced with a Hamilton syringe (model 7002) that contained 1.2–1.4 μL of a 20-mg/ml solution of muscimol in normal saline. This solution was injected over 10 min. The concentration and volume injected were similar to that used in previously published studies in which muscimol was used to inactivate the squirrel monkey FLR (Partsal et al. 1995). VOR eye movements were periodically recorded postinjection to ensure the normal functioning of the vestibular nerve, and of vestibular nucleus neuronal pathways.

Data analysis

ANALYSIS OF PURKINJE CELL SENSITIVITY TO EYE POSITION.

Static eye position sensitivity was assessed by measuring firing rate during intersaccadic periods (0.5–2 s) of spontaneous stationary eye position in the absence of a target. Multiple regression estimates of the sensitivity to horizontal and vertical eye position were computed from at least 20 stable eye positions (>150 ms before or after a saccade). For those units with nonlinear firing rate–eye position relationships, separate fits of firing rate to eye position were made across sub-ranges of the averaged response that were at least 180° in length and that appeared to be linear by visual inspection.

ANALYSIS OF DATA OBTAINED WITH SINUSOIDAL STIMULI. Most of the experimental paradigms described in this paper utilized sinusoidal target or turntable movements. Unit activity was recorded during periods of 30 s duration, with multiple tests usually obtained for each of the behavioral paradigms described above. Cycles were selected for further analysis only if the monkey’s gaze position was within 2–3° of the visual target. In experiments in which the visual target was not present, cycles in which sleepy eye movements occurred (epochs of 10 s or more without saccades or where the position of the eye drifted) were eliminated. Records from 20–100 selected cycles (mean, 26 at 0.5 Hz) were concatenated, desaccaded, averaged, and fit with sinusoidal functions. An iterative fitting technique was used to eliminate the significant deviations from linearity associated with periods of low firing rate (Chen-Huang et al. 1997). A significant minority of the Purkinje cells exhibited nonlinear responses during sinusoidal pursuit and VOR cancellation that were not associated with low firing rate. In these cases an estimate of unit response gain was obtained by fitting a sinusoidal function to the units firing rate to a portion of the cycle (always >180°) in which the response was linearly related to the stimulus (see Fig. 5 for an example).

ANALYSIS OF DATA OBTAINED WITH VELOCITY TRAPEZOID STIMULI. The methods used for analysis of unit responses during steps in head acceleration generated during VOR cancellation or fixation have been described in detail elsewhere (Cullen and McCrea 1993). Briefly, S–22 records of behavioral and neural responses when the monkey was either fixating the target or not moving the eyes prior to and after brief unpredictable steps in head acceleration (∼400°/s²) were averaged. Records were included in the analysis only if saccades were absent 100 ms prior to and 100 ms after the onset of the head acceleration step and if the monkey reacquired the target within 200 ms after onset.

RESULTS

The results are based on recordings from 187 Pk cells in the ventral paraflocculus and flocculus of three squirrel monkeys whose firing behavior was sensitive to horizontal smooth pursuit eye movements and/or VOR cancellation in the plane of the horizontal semicircular canals. Horizontal Pk cells constituted a large minority (∼45%) of the Pk cells encountered in this region of the cerebellum. The remaining Pk cells were neither sensitive to horizontal smooth pursuit eye movements nor to cancellation of the VOR. They were usually sensitive to vertical eye movements and were strongly modulated during vertical smooth pursuit when tested. Approximately 10% of the units encountered appeared to be unresponsive during both horizontal and vertical pursuit eye movements.

Anatomical location of horizontal Pk cells

An attempt was made to systematically explore the entire flocculus and ventral paraflocculus. After reconstruction of electrode tracks we estimate that only the most caudal and medial folium of the flocculus was unexplored. Pk cells with firing behavior related to horizontal smooth pursuit eye movements could readily be grouped into two categories, eye velocity Pk cells (Ev Pk cells) and gaze velocity Pk cells (Gv Pk cells), based primarily on their responses during whole body rotation (see below). The two cell types were intermixed and were found in every region of the flocculus and ventral paraflocculus that was explored.

Figure 1 shows the estimated location of the Ev and Gv Pk cells in the cerebellar FLR in all three monkeys. Figure 1A is a photograph of a lateral view of the FLR of one of the monkeys. In Fig. 1B the estimated location of Ev and Gv Pk units in all three monkeys in the sagittal plane is superimposed on an outline drawing of the flocculus and ventral paraflocculus. The location of each cell in the plot was calculated on the basis of the relative position of the tip of the electrode to the estimated rostral edge of the paraflocculus and to the estimated location of the vestibular nerve. Dorsal and coronal views of the distribution of Pk cells recorded are illustrated in Fig. 1, C and D.

Neither Ev nor Gv Pk cells were confined to one region of the FLR, although Purkinje cells within a small local region of cortex tended to be of the same variety. In 22 of 39 tracts in which 3 or more Pk cells were encountered, both Ev and Gv Pk...
cell types were recorded. However, when three or more Pk cells were separated by <1 mm on the same probe, they were more likely (4 of 6 instances) to be of the same type. No significant physiological differences were found in the eye or head movement sensitivity of Pk cells recorded in the most rostral part of the FLR as compared with Pk cells located in the most caudal regions of the FLR. In sum, Pk cells within a small local region of cortex tended to be of the same variety, but neither of the two types of units were confined to any one rostro-caudal region of the FLR.

Eye velocity and gaze velocity Purkinje cells

The firing rate of Pk cells in the FLR was modulated during smooth eye movements and during passive whole body rotation. In some units (n = 17; 9%) this modulation was small (gain re stimulus velocity was <0.2 spikes/s/deg/s) and difficult to analyze quantitatively. The remaining 170 units were classified on the basis of firing behavior during smooth pursuit eye movements and passive WBR. Nearly half of the Pk cells tested (80/170) were sensitive to head velocity during turntable rotation and were designated as Gv Pk cells, since they were similar to the Gv Pk cells that have been described in rhesus monkeys (Lisberger and Fuchs 1978a; Miles et al. 1980). The firing rate of the remaining Pk cells (90/170) was not significantly related to head velocity during WBR but was related to horizontal eye movements, particularly during smooth tracking of visual targets. Although the firing rate of some of these eye movement only Pk cells was related to both eye velocity and eye position (see below) in some circumstances, we refer to them as eye velocity (Ev) Pk cells because the eye position signals were not present in every paradigm (see below).

Sample records of the firing behavior of a typical Ev Pk cell and Gv Pk cell during sinusoidal smooth pursuit, VOR cancellation, and WBR are shown in Figs. 2 and 3. The firing rate of Ev Pk cells was modulated when the monkey pursued a moving target (Fig. 2A) and when it fixated an earth stationary target (Fig. 2B). When the monkey attempted to cancel its VOR (Fig. 2B), any modulation apparent in the activity of these cells was in phase with the uncanceled eye velocity. No activity related to head velocity could be discerned. The averaged response of the unit in each condition is illustrated on the right in Fig. 2.

The distinguishing characteristic of Gv Pk cells in the squirrel monkey FLR was their sensitivity to head velocity during passive whole body rotation. The Gv Pk cell shown in Fig. 3 was typical. Like the Ev unit illustrated in Fig. 2, it was sensitive to ipsilateral smooth pursuit eye movements (Fig. 3A). However, during VOR cancellation (Fig. 3B) the cell’s firing rate was related to ipsilateral head velocity. This head velocity signal was smaller than the eye velocity–related modulation recorded during ocular pursuit. When the monkey fixated an earth stationary target during WBR (Fig. 3C), the Gv Pk cell’s response was in phase with eye velocity, but reduced. The reduction could be accounted for by the presence of the concomitant head velocity signal, which in this condition was out of phase with the eye velocity signal.

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**FIG. 2.** Firing behavior of an Ev Pk cell during ocular pursuit (A), vestibulo-ocular reflex (VOR) cancellation (B), and VOR during fixation of an earth stationary target (C). The records on the left of the figure are sample records of stimulus velocity, eye velocity, and unit firing rate. The single cycles illustrated in each condition on the right of the figure are averaged desaccaded responses. The dashed traces are target velocity (Tgv) or turntable velocity (Tbv). The solid traces are eye velocity (Ev). The histograms are unit instantaneous firing frequency (25-ms bins). The calibrations in B also apply to C.

**Sensitivity of FLR Pk cells to eye movements**

**FIRING BEHAVIOR DURING SACCADES AND STEADY FIXATION.** The majority (112/187, 59%) of the Ev and Gv Pk cells in the FLR exhibited some sensitivity to ocular saccades, but the signals they generated were typically weak and were not consistent in the absence of a visual target. Most of the units that were sensitive to saccades generated inconsistent bursts of spikes in the pursuit eye velocity on-direction and/or were inconsistently inhibited during saccades to the unit’s off-direction. Some of the saccade-sensitive units (22%) exhibited the opposite behavior; i.e., they were excited during saccades to their pursuit off-direction and inhibited during saccades to the on-direction. A few cells (11%) generated bursts or pauses during saccades in all directions.

Most FLR Pk cells whose firing rate was modulated during horizontal smooth pursuit eye movements were not sensitive to eye position during steady fixation. Static eye position sensitivity was estimated in 119 Pk cells by multiple regression of tonic firing rate versus horizontal and vertical eye position recorded during periods of steady fixation in the absence of a target. The tonic firing rate of a minority of those units (29/119, 24%) was significantly related to horizontal eye position (mean, 4.5 spikes/s/deg). The horizontal eye position signal, when present, was related to eye movements in the same direction as the pursuit signal and was usually similar to an estimate of dynamic eye position sensitivity measured during ocular pursuit. A comparison of unit sensitivity to static eye
position ($K_s$) and “dynamic” eye position sensitivity recorded during sinusoidal smooth pursuit ($K_d$) was comparable. Dynamic eye position sensitivity was calculated from 0.5-Hz responses of 23 units using the following formula

$$K_d = K_1 \sin(K_2)/E_h$$

where $K_1$ and $K_2$ are unit response amplitude and phase and $E_h$ is the amplitude of horizontal eye position during pursuit. In this subset of neurons, the correlation between $K_s$ and $K_d$ was 0.88, and the slope of the regression was 0.86, which suggests that the phase lag in the responses observed in these Pk cells during 0.5-Hz pursuit could be related to an eye position signal.

**FIRING BEHAVIOR DURING SMOOTH PURSUIT EYE MOVEMENTS.** The firing rate of nearly half of the Purkinje cells in the flocculus and ventral paraflocculus was related to horizontal smooth pursuit eye movements, but their sensitivity varied over a large range (0.13–4.49 spikes/s/deg/s re eye velocity). Only a few horizontal eye movement–related Purkinje cells were sensitive to vertical eye position or to vertical smooth pursuit eye movements. Five units were encountered that were sensitive to both horizontal and vertical pursuit eye movements; four of these exhibited nonlinear responses during horizontal smooth pursuit (see below). Three were related to downward eye velocity and two to upward eye velocity during vertical pursuit. Vertical and horizontal directional preferences were not related in any consistent way.

The gain and phase of Pk cell responses during 0.5-Hz horizontal ocular pursuit are summarized in Fig. 4 and Table 1.

Both Ev and Gv Pk cells could be segregated into two subgroups based on the phase of their response during pursuit.

Most units had responses that were roughly in phase with ipsilateral eye movements (type I units); but 39% of the cells were sensitive to head and/or eye movements in the contralateral direction (type II units). For Ev Pk cells, type I and type II responses were found in nearly equal numbers (48% type II), but only 29% of the Gv Pk cells had type II responses.

Ev and Gv Pk cells differed in their response phase during sinusoidal smooth pursuit eye movements. On average, Gv units tended to fire in phase with eye velocity during pursuit (mean phase lag re eye velocity, 13.5°). The phase lag in Ev Pk cell pursuit responses tended to be larger (mean phase lag re eye velocity, 55°). The relatively large phase lag was observed both in Ev Pk units whose firing rate was correlated with static eye position and those that were not.

The mean sensitivity of all Pk cells to eye velocity during ocular pursuit at 0.5 Hz was $1.37 \pm 0.98$ (SE) spikes/s/deg/s, and the median sensitivity was 1.06 spikes/s/deg/s. In Fig. 4A gains that were in phase with ipsilateral eye movements are plotted to the right of zero on the abscissa, and units whose responses were in phase with contralateral movements are plotted to the left of zero. Note that the entire population of FLR Pk cells tended to be distributed zero eye velocity sensitivity. Thus if type I and type II Pk cells projected to the same population of neurons in the vestibular nuclei, the influence of the two types of Pk cells would tend to cancel one another during pursuit.

The modulation in the firing rate of FLR Pk cells during smooth pursuit decreased in conjunction with decreases in pursuit eye velocity if stimulus frequency was increased. During pursuit at 1 Hz, the gain of the responses of some units increased; but on average unit sensitivity to eye velocity was...
One bilaterally responsive Gv Pk cell is illustrated in Fig. 5 sensitive to eye movements during pursuit in both directions.

Eye velocity until it had achieved a threshold value. A significant fraction of both Ev Pk cells (22/90, 24%) and Gv Pk nonlinear responses were observed in the squirrel monkey. A when eye velocity achieves a threshold value (Lisberger et al.

NONLINEAR PURSUIT SIGNALS. In macaque monkeys some Pk cells code the velocity of smooth pursuit eye movements only with respect to target velocity (Tv), turntable velocity (Tb), or eye velocity (Ev). The gain and phase of the average eye movement responses evoked in each condition are shown in the first row. The table only includes data for units with response gains larger than 0.2 spikes/s/deg/s re target velocity during pursuit. Negative values are responses that lead ipsilateral velocity. VOR, vestibuloocular reflex; Hv, head velocity; Pk, Purkinje; Gv, gaze velocity.

unchanged from that observed during 0.5-Hz pursuit (1.94 – 1.97 spikes/s/deg/s; n = 10), despite a doubling of eye acceleration at the higher frequency and a halving of eye position excursion. The response phase re eye velocity advanced somewhat, from a lag of 21° at 0.5 to a lead of 0.2° at 1.0-Hz pursuit. At 2.3 Hz the gain of ocular pursuit declined to <0.2, and the modulation in Pk unit responses was proportionally reduced, yet the response gain re eye velocity was little changed (mean gain at 0.5 Hz = 1.45 and at 2.3 Hz = 1.57 spikes/s/deg/s; n = 8).

NONLINEAR PURSUIT SIGNALS. In macaque monkeys some Pk cells code the velocity of smooth pursuit eye movements only when eye velocity achieves a threshold value (Lisberger et al. 1994a; Miles et al. 1980; Noda and Warabi 1982). Similar nonlinear responses were observed in the squirrel monkey. A significant fraction of both Ev Pk cells (22/90, 24%) and Gv Pk cells (16/80, 20%) did not generate signals related to pursuit eye velocity until it had achieved a threshold value.

Fourteen other units (9 Ev Pk cells and 5 Gv Pk cells) were sensitive to eye movements during pursuit in both directions. One bilaterally responsive Gv Pk cell is illustrated in Fig. 5A. This unit’s firing rate was best fit by two sinusoidal functions that were out of phase with one another during sinusoidal smooth pursuit. In Fig. 5B the averaged firing rate of the cell is plotted versus eye velocity. Since the unit’s response was approximately in phase with eye velocity in both directions, it was possible to plot a linear fit to the firing rate/eye velocity relationship on either side of the “knee” at which the directional preference of the unit switched from ipsilateral to contralateral. The point at which this reversal occurred varied from unit to unit (Fig. 5C), although it was on average near zero eye velocity (mean, 0.5 ± 10.4°/s).

Firing behavior of Purkinje cells during VOR cancellation

During single-unit recordings, VOR cancellation was rarely perfect, and the average gain of the VOR at 0.5 Hz recorded concomitantly with unit recordings was 0.21 ± 0.12. When VOR cancellation was near perfect, the firing rate of Ev Pk cells was not modulated (Fig. 6A1). Ev Pk unit responses recorded during cycles in which the VOR cancellation was relatively poor were always larger than responses recorded when VOR cancellation was effective, and they increased in tandem with uncanceled eye velocity at higher stimulus frequencies. The sensitivity of Ev Pk cell to eye velocity during VOR cancellation was comparable to that measured during ocular pursuit (see Table 1), although responses were more phase-advanced.

In contrast to Ev Pk cells, the response amplitude of Gv Pk cells during VOR cancellation was inversely related to the gain of the VOR. The sensitivity of Gv Pk cells to head velocity during VOR cancellation varied over a wide range (Fig. 6B1), and the gain of their responses (Fig. 6B2) tended to be slightly more phase-advanced with respect to head velocity than with respect to eye velocity during ocular pursuit. In Fig. 6C1 unit sensitivity to eye velocity during smooth pursuit is plotted as a function of unit response gain re head velocity during VOR cancellation. The filled circles represent Gv Pk cells, and the open symbols represent Ev Pk cells. The dashed line indicates equal sensitivity to eye and head velocity. The mean eye velocity sensitivity of all Gv Pk cells during pursuit was 1.31 spikes/s/deg/s, while the mean gain of the head velocity signals generated by these cells during VOR cancellation was 0.56 spikes/s/deg/s.

The disparity between pursuit and VOR cancellation responses of Gv Pk cells could not be attributed to incomplete cancellation of the VOR. Figure 6C2 plots unit eye velocity sensitivity during pursuit versus an estimate of head velocity sensitivity calculated after subtraction of any residual eye velocity signal from the responses recorded during VOR cancellation. The change in unit responses as a function of canceled eye velocity was also estimated by comparing responses recorded during cycles when the VOR was less well canceled (but with eye position still kept within 2° of the target) to responses recorded when cancellation was more effective. The sensitivity of these cells to canceled eye velocity was similar to the corrected estimate of head velocity sensitivity obtained after subtraction of any residual eye velocity-related component of the response.

The VOR cancellation response of 13 Gv Pk cells was studied at higher stimulus frequencies (1.0 and 2.3 Hz). Gv Pk cells were equally sensitive to head velocity at stimulus frequencies of 0.5 and 1.0 Hz (1.27 and 1.25 spikes/s/deg/s; n = 7). At 2.3 Hz monkeys were not able to cancel their VOR, which increased to a mean gain of 0.80. The modulation in firing rate of most (4/6) of the Gv units tested at 2.3 Hz

### Table 1. Purkinje cell responses during smooth pursuit and whole body rotation

<table>
<thead>
<tr>
<th>Smooth Pursuit</th>
<th>WBR (VOR Cancellation)</th>
<th>WBR (Earth Target)</th>
<th>WBR (No Target)</th>
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<td></td>
<td>Gain re Tgv</td>
<td>Phase re Tgv</td>
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<td>Type III</td>
<td>1.02</td>
<td>229</td>
<td>1.47</td>
</tr>
<tr>
<td>Gv Pk cells</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Type I</td>
<td>0.82</td>
<td>57</td>
<td>1.41</td>
</tr>
<tr>
<td>Type II</td>
<td>0.68</td>
<td>211</td>
<td>0.95</td>
</tr>
</tbody>
</table>

n is number of cells. Summary of Purkinje cell responses during smooth pursuit eye movements and passive whole body rotation (WBR). Values in the table were obtained during sinusoidal ocular pursuit and turntable rotation at 0.5 Hz, 40°/s peak velocity. Each column summarizes the gain and phase of unit responses with respect to target velocity (Tv), turntable velocity (Tb), or eye velocity (Ev). The gain and phase of the average eye movement responses evoked in each condition are shown in the first row. The table only includes data for units with response gains larger than 0.2 spikes/s/deg/s re target velocity during pursuit.
reversed in direction and was in phase with unsuppressed eye velocity rather than with head velocity.

The poor VOR cancellation performance during high-frequency head rotation was particularly evident when the head was unpredictably accelerated during fixation of a head stationary target. Steps in head acceleration evoke compensatory eye movements for brief periods, and the VOR does not begin

FIG. 5. Nonlinear responses of FLR Pk cells during pursuit. A: averaged response of a Gv Pk cell that exhibited nonlinear responses during ocular pursuit. The dashed lines are sinusoidal fits to each half cycle. During ipsilateral pursuit the unit was sensitive to ipsilateral eye velocity (2.16 spikes/s/deg/s), while during contralateral pursuit the unit was sensitive to contralateral eye velocity (1.28 spikes/s/deg/s). B: plot of firing rate vs. eye velocity of the same unit. Each data point is the average firing rate of the unit illustrated in A at a different point of the pursuit cycle. The superimposed linear fits intersected at approximately zero eye velocity in this unit. C: linear regression analysis of nonlinear responses in 10 other Pk cells. Note that the point of intersection between ipsilateral and contralateral eye velocity sensitivity was idiosyncratic for each cell.

to be suppressed until 80–100 ms after the onset of such a perturbation (Cullen et al. 1991). Responses to head acceleration steps (300–400/s²) were studied in 14 Gv Pk cells and 4 Ev Pk cells. Figure 7A shows the averaged firing behavior of a type I Gv and a type II Ev Pk cell during contralateral steps in head acceleration (cH₀) that were generated during fixation of a target that remained stationary with respect to the head. The steps produced a transient VOR eye velocity (—) that was in the ipsilateral, ON-direction of the Gv Pk cell and in the Ev Pk cell's OFF-direction. The compensatory eye velocity evoked by the step [Eᵥ (VORc)] did not begin to be suppressed until approximately 90 ms after acceleration onset. The responses of Pk cells were also delayed. Gv Pk cells typically began to respond approximately 40 ms (mean latency = 37 ± 20 ms) after the onset of the acceleration step. The response of Ev Pk cells was even more delayed with a mean latency of 77 ± 27 ms.

FIG. 6. Firing behavior of FLR Pk cells during VOR cancellation. Averaged responses of an Ev (A1) and a Gv (A2) Pk cell are shown at the top. During VOR cancellation, Ev Pk cell responses were always related to any residual, uncanceled eye velocity. The firing rate modulation of Gv Pk cells was typically in phase with head velocity and was always associated with the same direction as the response elicited during smooth pursuit. B: gain (B1) and phase (B2) of the Gv Pk cell responses re ipsilateral turntable velocity during VOR cancellation at 0.5 Hz. C: FLR Pk cell sensitivity to eye velocity during ocular pursuit vs. head velocity sensitivity measured during VOR cancellation. C1: VOR cancellation responses uncorrected for residual eye movements. C2: unit responses during VOR cancellation corrected for eye movement–related activity related to imperfect cancellation. Dashed lines indicate equal sensitivity to eye and head velocity.
When a step in head acceleration occurs in the context of ongoing VOR cancellation, VOR eye movements evoked by the step are suppressed at a shorter latency (Cullen et al. 1991). The responses of a Gv Pk cell during steps in head acceleration that were generated while the monkey was suppressing its VOR are illustrated in Fig. 7B. Unit and eye movement responses to steps in both the contralateral (cH", red traces) and ipsilateral (iH", blue traces) direction are superimposed on a response evoked in the dark in the absence of a target (black traces). When the step in head acceleration was produced in the dark, the Gv Pk cell did not respond. However, a head velocity–related modulation was present in the Gv Pk cell both prior to and during the step when the monkey was already canceling VOR eye movements by fixating a head stationary target. The response of the cell (red histogram) closely matched the response predicted by a model of firing rate based on its sensitivity to head velocity measured during 0.5-Hz VOR cancellation and to eye velocity during ocular pursuit at 0.5 Hz (dashed line superimposed on firing rate histograms in Fig. 7B).

Signals generated by Purkinje cells during whole body rotation in the presence and absence of targets

The firing rate of most Pk cells was modulated during whole body rotation when the monkey fixated an earth stationary target. However, these rotational responses were reduced or absent when the rotation was done either in darkness or in the lit room without the visual target present. Figure 8 shows the responses of both an Ev Pk and a Gv Pk cell during 0.5-Hz sinusoidal WBR in the presence of an earth stationary target (Fig. 8A) and during rotation in the light when no target was present (Fig. 8B). The gain and phase of the VOR in the presence and absence of a target were comparable (Table 1), but the gain and phase of the signals generated by Pk cells were substantially affected by target presence.

The firing rate of Ev Pk cells was strongly modulated during WBR when an earth stationary target was present. The amplitude and phase of the rotational responses of Ev Pk cells were similar to those recorded during ocular pursuit. Eye velocity Pk units were slightly less sensitive to eye velocity during VOR with a target present (1.37 spikes/s/deg/s) than during smooth pursuit (1.53 spikes/s/deg/s), yet the phase lag re eye velocity observed during WBR was very similar to that observed during WBR when an earth stationary target was present. The amplitude and phase of the rotational responses of Ev Pk cells were similar to those recorded during ocular pursuit. Eye velocity Pk units were slightly less sensitive to eye velocity during VOR with a target present (1.37 spikes/s/deg/s) than during smooth pursuit (1.53 spikes/s/deg/s), yet the phase lag re eye velocity observed during WBR was very similar to that observed during WBR when an earth stationary target was present. The amplitude and phase of the rotational responses of Ev Pk cells were similar to those recorded during ocular pursuit. Eye velocity Pk units were slightly less sensitive to eye velocity during VOR with a target present (1.37 spikes/s/deg/s) than during smooth pursuit (1.53 spikes/s/deg/s), yet the phase lag re eye velocity observed during WBR was very similar to that observed during
ocular pursuit. Units with nonlinear responses during pursuit also exhibited similar response nonlinearity during WBR.

In the absence of a target, the signals generated by Ev Pk cells during WBR were reduced by more than half and phase led rather than lagged eye velocity (Fig. 8B). The reduction in response amplitude was observed both during WBR in the dark and during WBR in ambient light when the monkey faced the projection screen. In Fig. 8C Ev Pk unit response gains re eye velocity during WBR are plotted as a function of the gain of their response during smooth pursuit. The open symbols were responses recorded during fixation of an earth stationary target; the closed symbols were responses recorded when the target was not present. The dashed line indicates identical eye velocity sensitivity during VOR and pursuit. The dotted line is the best linear fit to the responses recorded in the absence of a target. The slope of the regression of unit WBR responses in the absence of a target re responses during pursuit (dotted line in Fig. 8C) was 0.29. Thus the sensitivity of Ev Pk cells to eye movements during the VOR in the absence of a foveal target was less than one-third of that observed during pursuit.

Most Gv Pk cells were modulated during WBR when a target was present. That modulation was approximately equal to the vector sum of the eye velocity–related response measured during pursuit and the head velocity–related response recorded during VOR cancellation (e.g., dashed line superimposed on the averaged firing rate histogram in Fig. 8A). For most Gv Pk cells this response was in phase with eye velocity, rather than head velocity, since the eye velocity signal was usually the larger of the two. In Fig. 8D the Gv Pk unit responses evoked by WBR during fixation of an earth stationary target are plotted as a function of the response predicted from their sensitivity to eye velocity during smooth pursuit and to head velocity during VOR cancellation (open circles). The correlation between recorded and predicted responses was 0.93.

Gv Pk cell responses during VOR were reduced in gain, and in some cases were abolished in the absence of a target (Fig. 8B). In a few cells the response recorded without the target was reversed in direction from the response recorded when it was present. In Fig. 8D the filled symbols are the responses recorded in the absence of a target. The slope of a regression line fit to the responses recorded in the absence of a target (dotted line in Fig. 8D) was 0.5. This decreased slope, and the 180° phase reversal of some units, suggests that Gv Pk cells were nearly twice as sensitive to VOR eye velocity when a foveal target was present than when it was absent.

In sum, Ev and Gv Pk cells responded in opposite ways during VOR cancellation. Ev Pk cells generated signals that increased with VOR gain, while Gv Pk cells generated signals that increased as the gain of the VOR decreased. The eye velocity signals generated by both types of Pk cell were proportionally reduced if eye movements were suppressed and were significantly reduced in the absence of a visual target. These signals were apparently not used to suppress the VOR, since they were related primarily to unsuppressed eye velocity during VOR cancellation. The head velocity–related signals of Gv Pk cells were inversely proportional to eye velocity during VOR cancellation and led eye velocity suppression when the head was passively perturbed during fixation of a head stationary target and thus could be used to cancel the VOR.

Effects of unilateral muscimol injection into the FLR on VOR cancellation and smooth pursuit

In two monkeys, small injections of muscimol were made into a caudal region of the ventral paraflocculus where horizontal Gv Pk cells were concentrated. The muscimol injections were carried out only after single-unit recordings had been completed, and a map of the FLR had been constructed based...
on encountered eighth nerve axons and other prominent adjacent landmarks (see METHODS).

After recording behavioral responses during smooth pursuit, VOR and VOR cancellation in both head-restrained and head-free conditions, 1.25 μl of a 2% muscimol solution was injected. Behavioral responses during sinusoidal experiments were then recorded over a period of several hours. During this time the amplitude of the slow phase of spontaneous nystagmus in the dark and the gain of the VOR in the presence and absence of a target were carefully monitored to determine whether the muscimol had diffused to the vestibular nuclei. In both monkeys a small (3–8°/s) spontaneous nystagmus began to appear 10–15 min after completion of the injection. In one monkey this spontaneous nystagmus increased to 8°/s in the ipsilateral direction and 1°/s in the downward direction. In the other monkey the spontaneous nystagmus was 5°/s in the contralateral direction. The gain of the VOR in the dark was unaffected by muscimol injection in both monkeys.

The earliest detectable effect of muscimol injection on visual or vestibular evoked eye movements was a decrease in the gain of the ipsilateral optokinetic following response evoked by rotation of a striped pattern-projection drum mounted above the monkeys head. No attempt was made to study this behavior in detail. The effects of muscimol injection on head-restrained smooth ocular pursuit, VOR cancellation, and VOR will be presented here, and the effects of the injections on responses recorded in the head-free condition will be presented in the accompanying paper (Belton and McCrea 2000).

The changes in smooth pursuit eye movements, VOR cancellation, and WBR resulting from muscimol injection are shown in Fig. 9. The first column (A1–A3) illustrates records of eye velocity evoked in each condition just prior to muscimol injection. The second column (B1–B3) contains sample records recorded 40–60 min after the injection. The records in the last column (C1–C3) are the averaged, desaccaded records of eye velocity evoked prior to injection (dark traces) and after injection (shaded, filled traces) are superimposed. In this animal muscimol injection produced a small ipsiversive constant velocity nystagmus (not shown). When corrected for this spontaneous nystagmus, the muscimol injection reduced ipsilateral and contralateral pursuit eye velocity during ocular pursuit nearly equally. Note that the gain of the eye movements evoked in the VOR cancellation paradigm increased to approximately 0.5 after the injection, but that the VOR was relatively unaffected.

VOR cancellation was strongly affected by muscimol injection in both animals. Within 30 min after the injection, the gain of the eye movements evoked during VOR cancellation increased from 0.1 to more than 0.5 (Fig. 9, A2–C2). The monkey was able to keep its eye position within 2° of the head stationary target only by generating numerous saccades. Slow phase eye velocity increased during both ipsilateral and contralateral turntable rotations, but it was slightly larger in the direction of the spontaneous nystagmus. The effect of muscimol on VOR cancellation developed earlier than the deficit in smooth pursuit eye movements and recovered earlier.

In sum, a unilateral injection of muscimol in the FLR produced a bilateral deficit in the ability to produce smooth pursuit
Eye movements and to suppress the VOR. It also produced a small decrease in the gain of compensatory eye movements evoked by WBR in the presence of a visual target.

Discussion

Purkinje cells in the flocculus and ventral paraflocculus are an important part of the neural substrate involved in producing visual following responses and in modifying the gain of the VOR (Fukushima et al. 1996, 1999; Lisberger and Fuchs 1978a; Miles et al. 1980; Noda and Suzuki 1979; Waespe and Henn 1981; Waespe et al. 1983). However, a moving visual target can be followed with different combinations of eye, head, and trunk movements for matching the speed of the gaze in space to that of the image. We had two questions when we began our experiments. One was whether Pk cells in the FLR play a more general role in gaze control when head and trunk movements contribute to gaze velocity. The other asked what part the FLR has in canceling VOR eye movements during passive or active, i.e., imposed or voluntary head movements. In this study we compared the contribution of the FLR of the squirrel monkey to control of gaze during pursuit eye movements and during passive whole body rotation. We found that most Purkinje cells whose firing behavior was related to smooth pursuit eye movements were also sensitive to eye velocity when passive rotation of the head contributed to smooth tracking. Approximately half of these cells were also sensitive to head velocity, but this signal was usually half as large as the eye velocity signal.

Although the firing rate of most Pk cells was modulated during passive whole body rotation, the output of the squirrel monkey FLR is apparently balanced in such a way that it has little net effect on the VOR in the absence of visual targets. This idea is supported by three observations. Pk cells whose firing rate was related to contralateral eye velocity were almost as frequently encountered as Pk cells that were related to ipsilateral eye velocity. Second, the modulation in firing rate of most Purkinje cells during WBR was absent or significantly reduced when a target was absent, while the gain of the VOR was essentially unaffected by removal of a target. Finally, as noted by Hightstein and colleagues (Partsalis et al. 1995; Zhang et al. 1995b), inactivation of the FLR had little effect on the VOR.

If the FLR has little effect on the VOR in the absence of a visual target or during fixation of earth stationary targets that are at a distance, it is clearly necessary for changing the gain of the VOR when the eye velocity required to fixate a visual target is different from head velocity (Waespe et al. 1983; Zee et al. 1981). The firing rate of all FLR Pk cells is profoundly modified during VOR cancellation. When squirrel monkeys suppressed their VOR the eye velocity signals produced by Ev and Gv Pk cells were also suppressed. In addition, the modulation in Gv Pk cell output reversed in direction; presumably this was because vestibular inputs to those cells were not opposed by a concomitant eye velocity signal. These modifications in the output of the FLR were apparently necessary for VOR cancellation, since the ability to suppress the VOR was significantly compromised when muscimol was injected. Similar observations have been made previously in squirrel monkeys and other primates (Partsalis et al. 1995; Takemori and Cohen 1974; Waespe et al. 1983; Zee et al. 1981; Zhang et al. 1995b).

Mixture of different Pk cell types in the FLR

The regions of the vestibular nuclei that give rise to the horizontal canal-related VOR receive inputs from Purkinje cells distributed in a rostrocaudal strip of cortex that extends throughout the flocculus and ventral paraflocculus (Balaban et al. 1981; Belknap and McCrea 1985; Langer et al. 1985; Voogd et al. 1996). Both Ev and Gv Pk cells were in every part of the FLR explored, which included all but the most caudal-medial folium of the flocculus. On some probes Ev and Gv Pk cells were found on the same tract within less than 1 mm of one another. The results do not support the idea that the flocculus and ventral paraflocculus contain fundamentally different types of Purkinje cells (Nagao 1992).

A mixture of Ev and Gv Pk cell types has been observed in other species. In goldfish, Ev and Gv Pk cells as well as a third class of cell, related only to head velocity, have been found (Pastor et al. 1994). In the rabbit, most of the Pk cells in the flocculus are of the Ev Pk type (Ghelarducci et al. 1975; Leonard and Simpson 1986), although some may have head velocity signals (Nagao 1990). Two types, comparable to the Ev and Gv types described in this study, have been found in the cat (Cheron et al. 1997; Fukushima et al. 1996). Although most studies in rhesus monkeys have focused on Gv Pk cells, the presence of Ev Pk cells in the FLR has often been noted (Fukushima et al. 1999; Kahlon and Lisberger 1997; Lisberger and Fuchs 1978a; Miles et al. 1980; Raymond and Lisberger 1997). In early studies as many as a quarter of the Purkinje cells in the FLR of the rhesus monkey were Ev Pk cells (Lisberger and Fuchs 1978a; Miles et al. 1980).

While the primary difference between Ev Pk cells and Gv Pk cells was their sensitivity to head velocity, the two classes of cells differed in other respects. Ev Pk cells were equally likely to be related to ipsilateral or contralateral eye movements while only a minority of the Gv Pk cells was sensitive to contralateral eye and head movements (29%). Ev Pk cells generated signals during pursuit eye movements that lagged eye velocity substantially, while the signals of Gv Pk cells were in phase with eye velocity. Ev Pk cells tended to be more sensitive to ocular pursuit than Gv Pk cells. Finally, the eye movement signals generated by Ev Pk cells were disturbed when the head was passively perturbed during fixation of a head-stationary target, while the responses of Gv Pk cells led eye velocity suppression.

The different responses of different classes of Pk cell might be presumed to be due to different afferent inputs. However, we were not able to detect a clear segregation of Ev and Gv cells, which suggests that the anatomical organization of those inputs may not be grossly detectable. A similar lack of segregation was described by Fukushima et al. (1999) in the Japanese macaque. Consequently, it seems unlikely that different Purkinje cell types reside in different folia of the ventral paraflocculus or the flocculus, or are located exclusively in one rostro-caudal region. On the other hand, if different classes of Pk cells were confined to parasagittal microzones extending throughout the FLR, it is possible that a single microelectrode tract might pass from one microzone to another. Such a level of organization might not have been detected with the methods we used.

Both Ev and Gv Pk cells appear to be involved in producing visual following reflexes, smooth pursuit eye movements, and
plastic changes in the VOR (Raymond and Lisberger 1997, 1998). However, the differences in the responses of Ev and Gv Pk cells during cancellation of the VOR suggest that each cell type has a different role in controlling eye movements. Gv Pk cells provide a gated vestibular reafferent input to VOR pathways that can be used to cancel the VOR, whereas during VOR suppression the eye movement signals normally produced by Ev Pk cells are removed. The relatively small size of the vestibular signal in squirrel monkey Gv Pk cells and the relatively large fraction of Ev Pk cells encountered compared with rhesus monkeys probably reflect important differences in the strategy each species uses to modify signal processing in VOR pathways. The pursuit eye velocity signals generated by Ev Pk cells are clearly not used to suppress VOR oculomotor signals since their firing behavior is related only to unsuppressed eye velocity during VOR cancellation.

Target-dependent responses of FLR Pk cells

The eye velocity–related signals produced by FLR Pk cells were strongly influenced by the presence of a foveal target. Target-related activity could account for 50% or more of observed Purkinje cell modulation. An analogous target sensitivity has been observed from neurons receiving input from the flocculus in the superior vestibular nucleus of the squirrel monkey (Zhang et al. 1995a). When those monkeys were rotated within a lighted optokinetic drum, as against simple rotation in darkness, unit response amplitudes increased. Similar phenomena have been previously described in cats and rabbits (Fukushima et al. 1996; Gelardi et al. 1975), although in the rabbit this may be attributed in some cases to a retinal slip signal (Leonard 1986).

Neurons in many regions of the brain, including several precerebellar nuclei, exhibit target-dependent oculomotor responses. Target-dependent eye movement signals have been observed in many regions of the cerebral cortex that are involved in controlling smooth pursuit eye movements (Bizzi and Schiller 1970; Bruce and Goldberg 1985; Komatsu and Wurtz 1988; Newsome et al. 1988; Sakata et al. 1980). The target dependence of eye movement signals in the FLR presumably reflects the predominant role of this structure in modifying signal processing in VOR pathways as a function of visual context (Goldreich et al. 1992; Grasse and Lisberger 1992; Robinson 1965; Robinson et al. 1986; Stahl and Simpson 1995).

It is doubtful that this effect is due to a retinal slip signal. There was little evidence of an augmentation of Pk simple spike activity when target slip on the retinal changed during periods of varying pursuit efficacy (see also Suh et al. 1999). The visually driven changes in Pk firing rate observed by Stone and Lisberger were in the opposite direction from that reported here. During on-direction pursuit for ipsilaterally responsive Gv Pk, the peak activity was reduced, not increased by the visual component of firing rate (Stone and Lisberger 1990). Neither is the target sensitivity likely to be the influence of olivary inputs. When Leonard inactivated the inferior olive using lidocaine in the rabbit, only DC (background) shifts of simple spike activity were observed. No changes in simple spike eye movement signal gain were evident (Leonard 1986). As for full field slip signals, the results from the VOR no-target experiments (Fig. 8), one where the room lights were on, the other where they were off, demonstrated essentially unchanging firing rate modulation across the two conditions.

Role of the FLR in VOR cancellation

Squirrel monkeys have at least two means of canceling or suppressing the VOR during passive whole body rotation (Cullen et al. 1991, 1993). One mechanism involves the use of visual feedback to suppress the VOR. The visual feedback mechanism depends on retinal image slip signals to produce VOR suppression and is closely related to mechanisms that produce visual following reflexes and smooth pursuit eye movements (McCrea and Cullen 1992). A second, nonvisual mechanism is also utilized that produces a parametric reduction in the gain of passive head movement signals in central VOR pathways (Cullen et al. 1991; Lisberger 1990; Robinson 1982).

It has a shorter latency than the visual mechanism, and contributes 30–50% of the reduction in sensitivity of central VOR pathways to head movements during VOR cancellation (Cullen et al. 1993). The FLR is clearly an essential part of the central neural substrate that mediates the ability to cancel or suppress the VOR during passive whole body rotation (Zhang et al. 1995b). It seems likely that it is part of the neural substrate that mediates both visual and nonvisual VOR suppression (Tomlinson and Robinson 1981).

The contribution of FLR Pk cells to VOR cancellation appears to be more complex than the simple addition of a pursuit signal to central VOR pathways. The output signals produced by Gv Pk cells during VOR cancellation are only half as large as those observed during pursuit, and the pursuit-related signals produced by Ev Pk cells are abolished when the VOR is canceled. Thus the output of the FLR during VOR cancellation is best characterized as a reduction in target-contingent eye velocity signals plus the addition of a head velocity signal. The former may be approximately three times as important as the latter (Fig. 10). The head velocity signal produced by Gv Pk cells is demonstrable at a short latency, which suggests that these cells are involved...
in nonvisual and well as visual mechanisms for canceling the VOR.

The nonvisual mechanism for VOR cancellation may be part of a general mechanism for context-dependent modification of the gain of the VOR (McCrea et al. 1996). The head movement signals produced by secondary vestibular neurons involved in producing the VOR are not only modified when the reflex is suppressed, but also when gain or phase of the reflex is modified as a function of viewing distance (Chen-Huang and McCrea 1998, 1999). We have previously suggested the possibility that these modifications involve gated feedback of inhibitory or excitatory vestibular signals to VOR pathways as a function of behavioral context. These transformed vestibular signals correspond to central eye velocity commands when imposed on secondary VOR pathways and are thus strongly correlated with slow phase eye velocity. It seems likely that the FLR is not only an important component of the central pathways for mediating visual feedback-induced modification of signal processing in VOR pathways, but also part of a central mechanism that adds or subtracts vestibular and eye velocity signals to VOR pathways as a function of behavioral context.

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