Saccades and drifts differentially modulate neuronal activity in V1: Effects of retinal image motion, position, and extraretinal influences

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In natural vision, continuously changing input is generated by fast saccadic eye movements and slow drifts. We analyzed effects of fixational saccades, voluntary saccades, and drifts on the activity of macaque V1 neurons. Effects of fixational saccades and small voluntary saccades were equivalent. In the presence of a near-optimal stimulus, separate populations of neurons fired transient bursts after saccades, sustained discharges during drifts, or both. Strength, time course, and selectivity of activation by fast and slow eye movements were strongly correlated with responses to flashed or to externally moved stimuli. These neuronal properties support complementary functions for post-saccadic bursts and drift responses. Local post-saccadic bursts signal rapid motion or abrupt change of potentially salient stimuli within the receptive field; widespread synchronized bursts signal occurrence of a saccade. Sustained firing during drifts conveys more specific information about location and contrast of small spatial features that contribute to perception of fine detail. In addition to stimulus-driven responses, biphasic extraretinal modulation accompanying saccades was identified in one third of the cells. Brief perisaccadic suppression was followed by stronger and longer-lasting enhancement that could bias perception in favor of saccade targets. These diverse patterns of neuronal activation underlie the dynamic encoding of our visual world.

Introduction

Natural viewing in primates consists of abrupt voluntary saccades interspersed with periods of visual fixation containing small involuntary saccades and drifts. Both voluntary and fixational eye movements are integral components of the spatiotemporal inputs to the visual system. Not only do eye movements produce both fast motions (saccades) and slow motions (drifts) of the retinal image, they cause receptive fields to land on stimulus features, move over them, or leave them (Gur & Snodderly, 1987, 1997; Snodderly, Kagan, & Gur, 2001).

The utility of large saccades for searching through the environment is obvious, but the benefits of the smaller fixational eye movements are more subtle, and their functional role is not fully understood. In the parafovea, fixational eye movements are thought to prevent image fading (Clarke & Belcher, 1962; Ditchburn, 1973, 1980; Gerrits & Vendrik, 1974), and a faded stimulus often reappears after a fixational saccade (Martinez-Conde, Macknik, Troncoso, & Dyar, 2006; Timberlake & Snodderly, unpublished data). Nevertheless, the importance of fixational saccades (often called microsaccades[1]) has been questioned because they rarely occur during aspects of natural vision that do not require prolonged fixation

(Kowler & Steinman, 1979, 1980). Perhaps of greater significance than prevention of image fading, evidence is emerging that the involuntary movements of fixation improve the visibility of image detail in the fovea (Rucci & Desbordes, 2003; Rucci, Iovin, Poletti, & Santini, 2007). However, in the fovea, the improved stimulus visibility does not appear to require the occurrence of fixational saccades (Rucci et al., 2007; M. Rucci, personal communication). Instead, the perceptual enhancement must occur during the slow fixational drifts.

Although physiological effects of slow drifts have been relatively neglected, the effects of fixational saccades on V1 neurons have received considerable attention. Depending on the visual stimulus and the task, different authors have reported post-saccadic suppression (Leopold & Logothetis, 1998), enhancement ("bursts," Livingstone, Freeman, & Hubel, 1996; Martinez-Conde, Macknik, & Hubel, 2000, 2002), or both (Snodderly et al., 2001). To resolve these conflicting results, and to clarify the relative contributions of saccades and drifts, we have conducted a cell-by-cell analysis separating the effects of saccades and drifts. In natural vision, fixational drifts follow voluntary as well as fixational saccades; therefore we studied effects of both voluntary and fixational saccades and the drifts following them. Although some V1 neurons were transiently activated only by saccadic movements, others discharged vigorously during drift periods following both fixational and voluntary saccades, as long as the stimulus was optimally positioned on the receptive field. Many of the sustained neurons that were activated in drift periods had spatially selective receptive fields, providing a neural basis for perception of fine detail during drifts (Rucci et al., 2007).

We found a systematic relationship between eye movement activation patterns and neuronal responses to flashed or moving stimuli. This correspondence suggests a unified description of effects of spatiotemporal inputs to V1 receptive fields, either imparted by eye movements or caused by external stimuli. This outcome implies that much classical neurophysiological data on V1 spatio-temporal properties can provide plausible working hypotheses for the study of natural vision during eye movements.

Large saccadic movements are known to be accompanied by extraretinal modulations of neuronal excitability in the early visual pathway (Duffy & Burchfiel, 1975; Kayama, Riso, Bartlett, & Doty, 1979; Ramcharan, Gnadt, & Sherman, 2001; Reppas, Usrey, & Reid, 2002; Royal, Sáry, Schall, & Casagrande, 2006; Thiele, Henning, Kubischik, & Hoffmann, 2002). However, for the smaller fixational saccades, the occurrence of extraretinal effects has been debated (Martinez-Conde et al., 2002; Martinez-Conde, Macknik, & Hubel, 2004). Here we provide new data to show that similar extraretinal modulation occurs with both fixational saccades and small voluntary saccades in about a third of the V1 neurons that we studied. The extraretinal modulations may contribute to perceptual variations in visibility around saccades, including both suppression and enhancement (Ross, Morrone, Goldberg, & Burr, 2001; Rucci & Desbordes, 2003).

Methods

Data were collected from three adult female monkeys (*Macaca mulatta,* M42, M45, and M46). The monkeys were trained to fixate on a light-emitting diode (LED) and were prepared for electrophysiological recordings as previously described (Snodderly & Gur, 1995). All procedures complied with NIH guidelines and were approved by the Animal Care and Use Committee of the Schepens Eye Research Institute.

Data collection

Fiber electrodes (Reitboeck, 1983) with bare tip lengths ≤5 μm and impedance at 1 kHz of ~3–7 MΩ were most frequently used for extracellular recording. In some experiments, glass-insulated platinum-iridium electrodes (Snodderly, 1973) with a tip diameter of 1–1.5 μm, and bare tip length of 5–7 μm, were used. In a subset of experiments, we chronically implanted electrodes for several days (up to 2 weeks). During recording, the electrode could be moved along the vertical axis by the microdrive; at the end of the day, the electrode was firmly locked in place. In most cases, single-unit activity was recorded; when multi-unit activity was recorded, cells were sorted using principal components analysis and fuzzy k-means clustering (Abeles & Goldstein, 1977; Gur, Beylin, & Snodderly, 1999).

Eye position was monitored by a magnetic search coil (Judge, Wurtz, & Richmond, 1980; Robinson, 1963; 1–2 arcmin resolution, 200 Hz sampling rate) for 55% of the cells studied during fixation, as well as for all cells studied during voluntary saccades. For 45% of the cells studied during fixation, a double Purkinje image eye tracker (2–3 arcmin resolution; 100 Hz sampling rate) was used. Although not ideal, 100 Hz is sufficient to detect the overwhelming majority of even the smallest saccades, and to map saccadic start- and end-point position, because most fixational saccades result in a net change of position that would be detected. Therefore, even if we might sometimes miss the exact onset of a very small saccade, we would still detect the net displacement of the eye 10 ms later, introducing temporal jitter to saccade-triggered averages in a small fraction of trials that had a negligible impact on the results (see Supplemental Methods for a detailed treatment of the effects of sampling rate). Eye position was recorded together with spike arrival times (0.1-ms time resolution) and spike shapes collected at 20–25 kHz (Gur et al., 1999). To avoid breakage, search

coils were made of a thicker wire (AS 634, Cooner) and they were sutured to the sclera for maximum accuracy (Tang et al., 2007).

The trial started when the monkey correctly pressed the lever in response to illumination of the small (4 to 7 arcmin depending on the viewing distance) fixation LED. The stimulus was presented continuously for 5 s provided that the gaze remained within a fixation window (±0.5° to ±1°). To accommodate blinks, brief (100–200 ms) deflections from the fixation window were allowed. For voluntary saccade trials, the fixation window was briefly removed during the saccade when the fixation target changed position.

Visual stimulation

Stimuli were displayed on a Barco 7351 monitor at a 60-Hz frame rate, with a Truevision ATVista video graphics adapter (old system), or more recently on a Sony 500 PS monitor at a 160-Hz frame rate, driven by a Cambridge Research Systems VSG2/3F graphic card (new system). Bars were optimized for orientation, length, and color (white, green, or red), 0.9- or 1-log units brighter (*increment*) or darker (*decrement*) than the background of 1 or 5 cd/m^2. Chromatic stimuli were generated by activation of individual guns of the monitor. After the ocular dominance was established, stimuli were viewed binocularly, unless responses during monocular viewing were substantially stronger.

For receptive field mapping, speed tuning, and flash stimulation trials, the eye position signal was added to the stimulus position signal at the beginning of each video frame to compensate for changes in eye position (e.g., Snodderly & Gur, 1995; Tang et al., 2007). Note that the maximum delay between shifts in the eye position and subsequent corrections could be as long as 28 ms in the old experimental system, and 10 ms in the new system; thus, this procedure could not eliminate effects of the fast saccadic movements, which was done offline by removing data epochs containing saccades. The aim of the online eye position compensation was to ensure that the stimulus had a known relationship to the receptive field during the slow inter-saccadic drift periods. The compensation corrected for the change in position that followed each fixational saccade and for the slow drift movements that occurred until the next saccade.

The width and location of receptive field activating regions (ARs) were estimated with narrow increment and decrement bars swept forward and back at 1.5–7°/s across the receptive field in a direction orthogonal to the axis of optimal orientation (Figure 1A; Kagan, Gur, & Snodderly, 2002). The total region of space occupied by ARs was considered the classical receptive field (CRF). The data for speed tuning, and direction selectivity recorded with sweeping bars, and for estimation of response transiency with flashing bars were also collected while compensating for eye position.

To study the visually driven effects of fixational and voluntary eye movements, compensation for eye position was turned off, and a stationary, optimally configured bar was placed in the CRF (Figure 1B). Fixational eye movements (saccades and drifts) moved the CRF over and around the bar. Voluntary saccades were elicited by switching between fixation targets (usually 3 times in a 5-s trial) positioned so that the CRF would land on the stimulus, cross it, or leave it (Figure 1C).

To study the effects of saccades on the ongoing discharge in the absence of deliberate visual stimulation, we used a uniformly illuminated screen of the monitor at 1 or 5 cd/m^2 ("light" condition) or covered it and recorded trials when the room was completely dark except for the small fixation LED ("dark" condition). Prior to trials run in the dark, monkeys were adapted to the 1 or 5 cd/m^2 background, so their sensitivity to any dim stray light would be minimized (Snodderly et al., 2001).

Precautions were taken to ensure that the extraretinal modulation was not a result of inadvertent visual stimulation of V1 receptive fields, especially in the light. We restricted the analysis of extraretinal effects to fixational saccades that kept the CRF away from monitor edges. Maximal saccadic displacement was ≤2.4° while the position of the CRF was at least 3° from the edges during central fixation. We also studied cells that were at least 2.5° eccentric from the fixation target, so that the fixation target would not stimulate the cells. In the case of larger voluntary saccades, we placed the fixation targets so that the receptive field stayed away from monitor edges along the entire saccade trajectory. These precautions, together with the fact that the latency of the enhancement and the biphasic time course of modulations were very different from visually driven responses, make it very unlikely that these saccade-related modulations were generated by unintended visual stimulation.

Data analysis

Data from three monkeys were analyzed for this study: 118 cells for fixational eye movement effects (53 cells from M45, data collected with the old system; 65 cells from M46, data collected with the new system), 44 cells with voluntary saccades (M46, new system), and 299 cells for extraretinal modulation of ongoing activity (M42—34 cells, M45—100 cells, M46—165 cells). Many cells contributed to more than one analysis. The eccentricity of receptive fields ranged from 0.6° to 10° (4.5 ± 1.7°). Based on physiological criteria and cortical depth (Gur, Kagan, & Snodderly, 2005; Snodderly & Gur, 1995), recording sites included a broad sample of all laminar locations.

Saccades were detected by a velocity threshold of 10°/s and a preceding 50-ms period of stable fixation (Bair & O'Keefe, 1998; Snodderly et al., 2001). Blink-related eye movements were automatically detected and excluded from further analyses. The velocity threshold was chosen
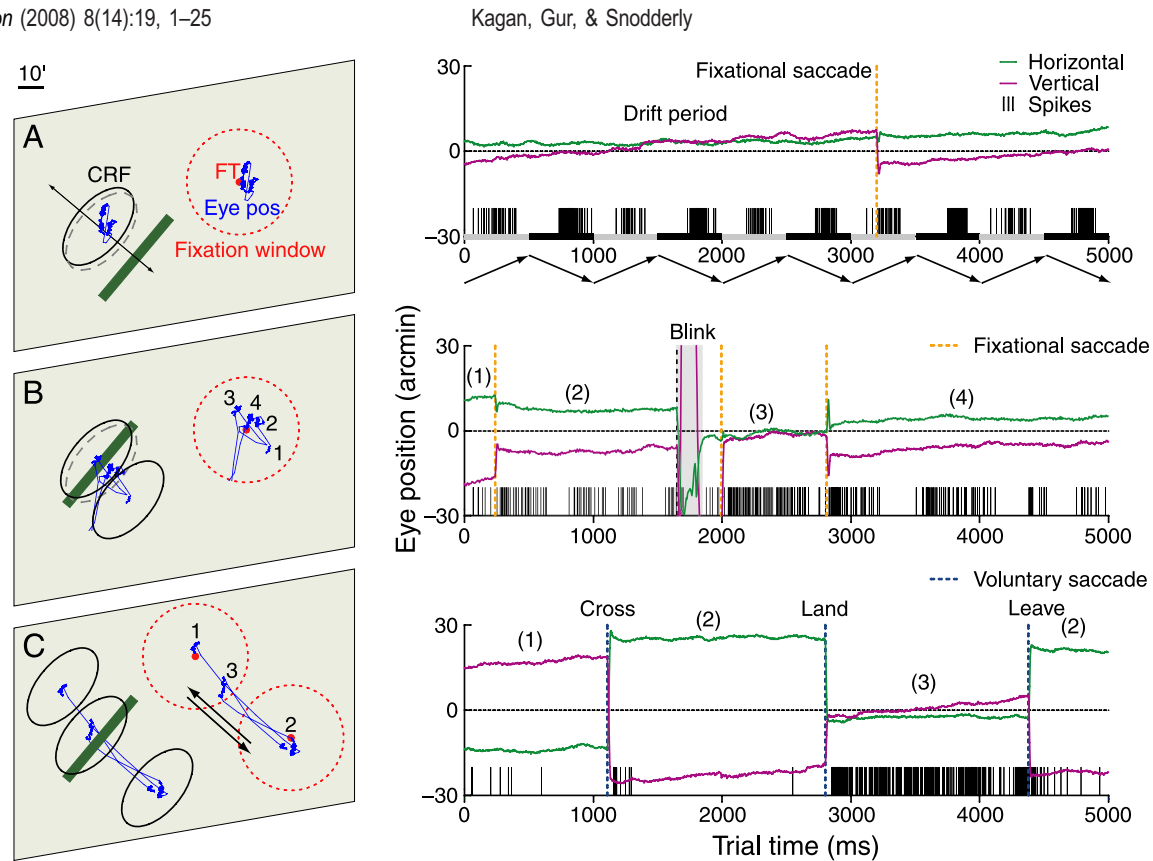
Figure 1. Experimental procedures. Left panels present a schematic view of the fixation target (FT, red dot), the fixation window (red dashed circle), and the eye position trajectory (wiggly blue line) during behavioral trials for three types of experiments. The classical receptive field (CRF, black ellipse) moved in space with the eye and it was stimulated with a rectangular bar stimulus (shown in green). Right panels depict records of 5-s experimental trials for the corresponding conditions. Horizontal (green line) and vertical (magenta line) eye position are plotted as a function of time along with arrival times of nerve spikes (short vertical lines). Dashed vertical lines denote onset times of fixational and voluntary saccades. (A) Receptive field mapping with sweeping bars while compensating for changes in eye position. An optimally oriented bar was moved forward and back across the neuron's response field. Light gray and black horizontal bars above the time axis represent the two sweep directions. Several cycles of stimulus movement were presented in each trial. The position and extent of the CRF was estimated online from several such trials (the dashed gray ellipse shows the estimated CRF at its position relative to the fixation target). The black ellipse indicates the position of the CRF at the beginning of the trial. Vertical eye position shows a characteristic upward drift during fixation. Since the position of the bar was continuously adjusted to compensate for changes in eye position, drift and position offsets following saccades did not affect the estimation of the CRF—note the consistent timing of the spike train relative to timing of the sweep across repetitions. (B) Effects of fixational eye movements. A stationary bar was placed at the spatial location of the CRF (estimated as shown in A), and fixational eye movements moved the CRF over and around the bar. Inter-saccadic drift periods during fixation are indicated by numbers 1–4. The CRF (black ellipse) is shown at positions 1 and 3. Blinks were automatically detected and excluded from analyses. This cell was activated continuously while the CRF was on the stimulus, but differences in firing rate between different fixations, corresponding to different locations of the bar on the sensitivity profile of the CRF and the stimulus, are apparent (cf. 4 vs. 2, 3). Dense spike bursts after every fixational saccade are also evident, reflecting "mixed" activation by both slow drifts and fast saccades. (C) Voluntary saccades were elicited by switching between two fixation targets 1 and 2, positioned so that the CRF would cross the stationary bar or land on the bar and then leave it (see also Figure 2). Note bursts of spikes after crossing and landing saccades, and sustained firing in the drift period (3) following the landing saccade, but not after crossing saccade (2). This cell (24822) had an ongoing firing rate of ~3 spikes/s, hence a few scattered spikes occurred in the drift periods even when the CRF was not on the stimulus (1, 2).

to detect even very small saccades and to ensure that inter-saccadic periods did not contain any abrupt eye movements. With 200-Hz sampling, saccades >3′ could be detected. With 100-Hz sampling, saccades >6′ could be detected. The inter-saccadic periods of relatively stable fixation, containing slow eye movements of varying

velocity and often exhibiting a consistent directional component (e.g., upward shift, Snodderly, 1987) are termed *drift* periods (Skavenski, Robinson, Steinman, & Timberlake, 1975). The resolution of our eye tracking systems did not allow resolving the potential presence of tiny high-frequency physiological *tremor*; therefore, the

drift periods may also contain tremor of very low amplitude. Detailed metrics of ocular drifts of the monkeys used in this study are given in the Supplemental Methods, along with a discussion of the limits of detection of small saccades.

To get reliable estimates of receptive field maps, speed tuning, and direction selectivity with sweeping bars, and the transiency index in response to flashing bars, the epochs of 200 ms following saccades or blinks were excluded from the analysis. The speed tuning peak was estimated from a cubic spline interpolation function fitted to the values of peak firing rates in 10-ms bins for 1.5–36°/s sweeps in the preferred direction. A direction index DI was computed as DI = 1 − (# spikes fired in the non-preferred direction) / (# spikes fired in the preferred direction). Simple and complex cells were distinguished by the degree of spatial overlap of increment and decrement activating regions in the CRF (Kagan et al., 2002). Monocontrast cells were defined as those cells responding to only to one sign of contrast, either light or dark bars, but not both (Kagan et al., 2002).

To examine effects of eye movements during presentation of stationary bars without position compensation, various perisaccadic and inter-saccadic intervals were analyzed around saccades or between successive saccades, as described below. All saccade-triggered averages were aligned to the saccade onset. To quantify the relative strength of post-saccadic and inter-saccadic firing, a normalized saccade-drift difference (*SDD*) was calculated using saccades separated from the nearest saccade by at least 250 ms. For fixational eye movements, the index was calculated using data from saccades that led to an increase in firing in the 250-ms period after the saccade compared to the 250-ms before the saccade (*increasing* saccades, 62% of all fixational saccades); for voluntary eye movements, the index was calculated using landing saccades. Increasing fixational saccades comprised most saccades in saccade-activated and mixed classes; in the position/drift class, increasing saccades were primarily landing saccades or "within" saccades (CRF stays on the stimulus). The saccade-drift difference compared the mean firing rate in the periods 0–150 ms immediately after the saccade (FR_{sac}) to the firing rate (FR_{drift}) during drift periods from 250 ms after the saccade to the next saccade—therefore drift periods had variable duration (829 ± 570 ms fixational, 691 ± 255 ms voluntary). Firing rates were corrected for the ongoing firing rate measured in the "light" condition (FR_{base}):

$$SDD = \frac{(FR_{sac} - FR_{base}) - (FR_{drift} - FR_{base})}{(FR_{sac} - FR_{base}) + (FR_{drift} - FR_{base})}$$

$$= \frac{FR_{sac} - FR_{drift}}{FR_{sac} + FR_{drift} - 2FR_{base}}. \qquad (1)$$

To evaluate the validity of using "increasing" fixational saccades for SDD computation as the closest equivalent of voluntary landing saccades, we re-calculated SDD for fixational landing saccades in 28 cells where ≥5 precisely mapped landing trajectories could be identified. The SDD for these landing saccades ranged from −0.25 to 1.01 and it corresponded closely to the SDD based on *all increasing* saccades ($r = 0.94$, $p < 1e^{-5}$, mean $SDD_{increasing}$ − $SDD_{land} = 0.07 ± 0.14$).

To characterize the immediate time course of post-saccadic modulation, we used a transiency index (TI), which is similar to the SDD index, but computed for firing rates in shorter periods of 0–120 ms after the saccade (response *peak*) and 120–250 ms after the saccade (*tail* of the post-saccadic response):

$$TI = \frac{FR_{peak} - FR_{tail}}{FR_{peak} + FR_{tail} - 2FR_{base}}. \qquad (2)$$

The peak was calculated as the mean of the 3 highest 10-ms bins in the 0- to 120-ms period to accommodate variations in response latency. These peak estimates were also used for analysis of peak firing rates (Figure 6A). There was a close correspondence between the SDD and TI ($r = 0.74$ and 0.8 for fixational and voluntary landing saccades, respectively; $p < 0.0001$; Supplemental Figure S1).

The cross-covariance function between eye velocity (sampled each 10 ms or 5 ms, in the latter case down-sampled to 10 ms) and instantaneous spike rate (binned at 10 ms) was estimated separately for each 5-s trial (or portions of the trial after removal of blinks) and averaged across all trials for each cell, yielding several measures: minimal latency of first significant lag, amplitude of maximal covariance, latency of maximal covariance, and duration of significant portion of the function. The MATLAB function *xcov* was used for the computation. Before averaging, each individual trial function was normalized so that the auto-covariances at 0-lag were 1 ("*coeff*" scaling option). The significance of the cross-covariance function at each lag to be larger than zero was assessed by the one-tailed *t* test ($p < 0.025$).

For the analysis of saccadic modulation of the ongoing activity ("extraretinal" modulation in the absence of a deliberate visual stimulus), only saccades of ≤100 arcmin amplitude and within ±100 arcmin from the mean fixation locus were analyzed to ensure that the fixation target or monitor edges did not appear within the CRF at any moment (Snodderly et al., 2001). To avoid overlap of effects from adjacent saccades, we only analyzed saccades that were not preceded by another saccade in a period at least 300 ms before and were not followed by another saccade for at least 400 ms after the saccade onset.

To assess statistical significance of the modulation, we estimated a mean and standard deviation (*SD*) of the baseline ongoing firing rate using 600-ms epochs sampled

from drift periods starting 300 ms after any saccade and ending 50 ms before the next saccade. Using at least 5 perisaccadic intervals, and a similar number of drift periods, we adopted ±2.5 *SD* confidence limits around the baseline as the criterion for statistical significance. If at least two consecutive bins of 25 ms crossed the ±2.5 *SD* threshold, the modulation was deemed "significant" (Figure 7B; Reppas et al., 2002). This criterion led to only 3% of false positives in sets constructed from drift intervals (8/250 cells in the light and 2/83 cells in the dark). These cells had only a few (5 to 10) valid drift intervals, so some random fluctuation of the ongoing firing did not average out. In contrast, 34% (86/250 cells) in the light and 31% (26/83 cells) in the dark were found to be significantly modulated by the saccades. Visual inspection of individual saccade-triggered averages confirmed that the significance test was relatively conservative. In a few cases, when the ongoing activity was high, a clear modulation was present but it was not strong enough to cross the ±2.5 *SD* limits since the *SD*, which scales with the mean firing rate, was too large. For cells with zero ongoing rate (and thus zero *SD*) at least 2 *non-empty* perisaccadic intervals were required for significance (only 5 of 59 cells with zero ongoing rate were significant).

Perisaccadic modulation of the ongoing activity was summarized with two indices: normalized enhancement (EI) and suppression (SI), using 25-ms bin saccade-triggered averages plotted around the baseline firing rate (Figure 7B; cf. Reppas et al., 2002). EI and SI were calculated as integrals of areas falling above or below the baseline, divided by the integral of the baseline, in the interval of [-100 to +300] ms around the saccade (Reppas et al., 2002). To illustrate, a doubling of the firing rate for the entire interval would lead to EI = 1. In the special case when the baseline rate was 0 spikes/s, the SI was always zero and EI was not normalized. The latency of the significant post-saccadic enhancement was estimated as the beginning of the first two consecutive bins exceeding the 2.5 *SD* confidence thresholds.

Correlations between variables were calculated using the Spearman *r* or the Pearson r (for normally distributed variables). Unless stated otherwise, statistical significance of differences between distributions was assessed with *t* test. Values reported for individual parameters are means ±*SD* Analyses were done with custom software written in MATLAB (MathWorks).

Results

Sections 1–3 of the Results present analyses of the interactions between visual stimuli and V1 receptive fields introduced by eye movements. Figure 1 illustrates the three types of trials used in these experiments. First, we mapped each neuron's classical receptive field (CRF) with

increment and decrement bars sweeping across the receptive field while monkeys maintained fixation within a small eye position window. Figure 1A, left panel, schematically shows the spatial relationships between the fixation target, FT, eye position, the CRF, and the moving bar. The right panel shows the time courses of eye position, cell firing, and stimulus timing during the trial. The position of the stimulus bar was continuously adjusted during receptive field mapping to compensate for slow fixational drifts and for position offsets associated with fixational saccades ("position compensation"; see Methods). This technique was not intended to compensate for fast transients during saccades, but it yielded reliable online estimates of CRF width and location that were used in subsequent eye movement experiments select locations of stimuli and fixation targets.

For eye movement experiments, illustrated in Figures 1B and 1C, position compensation was turned off. Effects of fixational eye movements were studied by placing a stationary, optimally oriented bar in the CRF (Figure 1B left panel). Fixational saccades and drifts moved the CRF over and around the bar, activating the cell in characteristic patterns (Figure 1B right panel). To study effects of voluntary eye movements, the fixation target abruptly changed position, eliciting a visually guided saccade. We adjusted the positions of the fixation targets to produce three types of voluntary saccades, all illustrated in Figure 1C: *landing* saccades (CRF initially not on the stimulus; saccade causes CRF to land on the stimulus), *leaving* saccades (the reverse of landing saccades), and *crossing* saccades (CRF crosses the stimulus during the saccade but is not on the stimulus *before* or *after* the saccade). The cell used for this example responded to each saccade with a burst of spikes, but also fired continuously while the CRF was on the stimulus, after a landing saccade (Figure 1C, right panel).

Analysis of fixational and voluntary eye movements reveals three activation patterns

During presentation of a stationary stimulus, each neuron in V1 was activated in one of three characteristic patterns when eye movements caused the CRF to land on the stimulus or to leave it. Figure 2A illustrates these three firing patterns during behavioral trials that elicited landing and leaving voluntary saccades. The cell in the top row (termed a "saccade" cell) discharged only short post-saccadic bursts after landing saccades but was silent in inter-saccadic drift periods. At the other extreme, the bottom row shows data from a "position/drift" cell that fired during drift periods as long as the CRF was positioned on the stimulus. Finally, the cell illustrated in the middle row exhibited a mixture of these two patterns, firing bursts of spikes after saccades and continuing to discharge above its ongoing rate during drift periods after
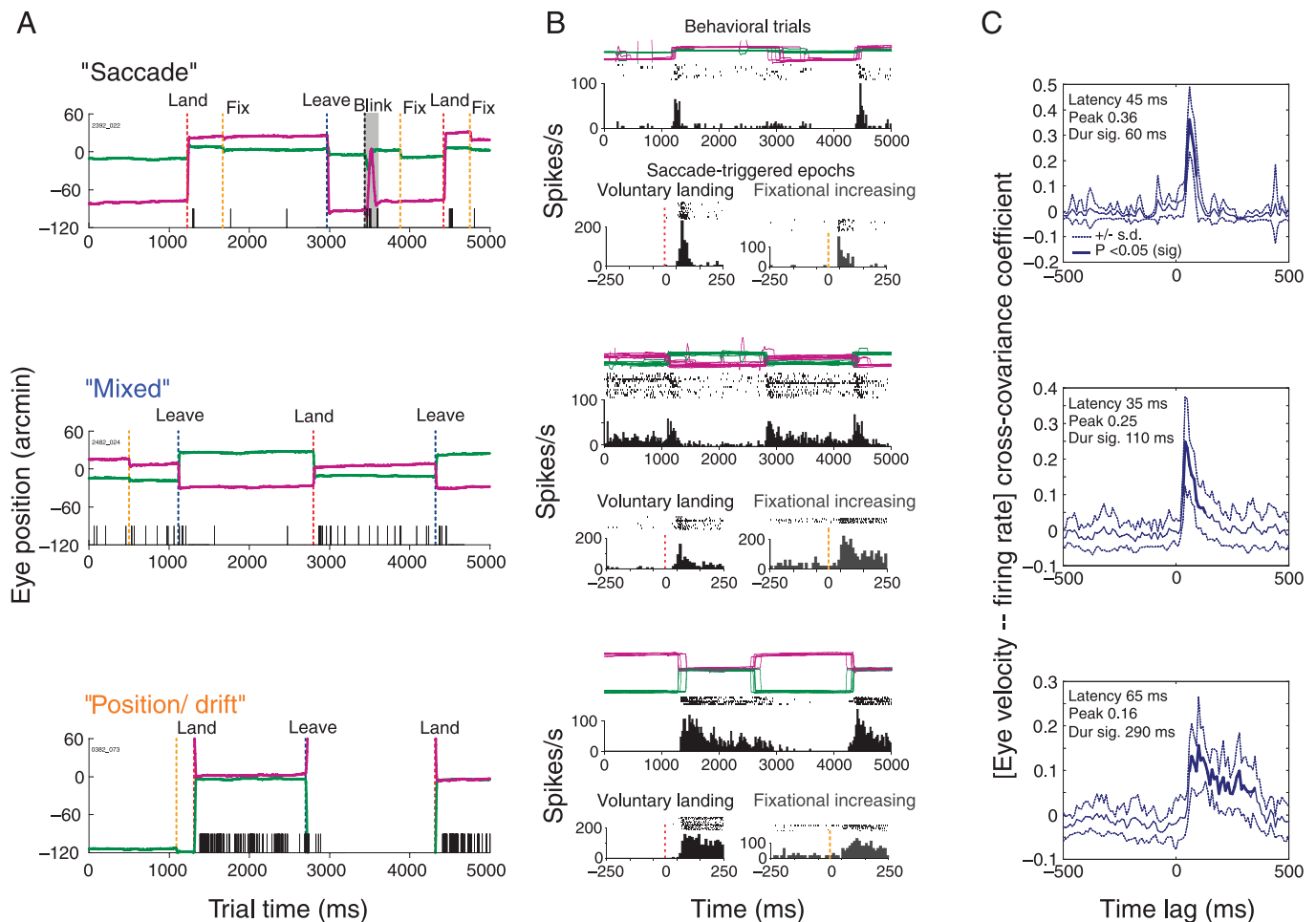
Figure 2. Three neuronal classes of eye movement activation. (A) Records from three example neurons: "saccade-activated" (*sac,* top row), "mixed" (*mix,* middle row), "position/drift-activated" (*pos,* bottom row). Five-second trials were recorded while the monkey made voluntary saccades that caused the CRF to land on an optimally oriented stationary bar (landing saccades denoted by vertical red dashed lines) or to leave it (blue dashed lines). Fixational saccades are marked by orange dashed lines. See S10 for similar examples of records for *sac* and *pos* cells during fixation. (B) Compiled results from repeated trials in for each of the cells in column A. Each panel consists of superimposed eye position traces, rasters of spike arrival times, and saccade-triggered histograms for voluntary landing saccades (black) and for fixational increasing saccades (gray). (C) Cross-covariance analysis of eye velocity and neuronal firing rate performed on complete sets of trial records like those shown in panel A (with blink periods excluded) for the same three neurons. Plots show the cross-covariance coefficient as function of time lag between eye velocity and instantaneous firing rate. Thick traces denote epochs significantly different from zero. The text inset in each panel shows the latency of first significant time lag, the peak of cross-covariance function, and the duration of the significant epoch.

landing saccades ("mixed" cell). Supplemental movies show analogous examples for fixational eye movements.

The interpretation of activations by eye movements is most straightforward when the saccadic displacements cause the CRF unambiguously to land on the stimulus, leave it, or cross it (Snodderly et al., 2001). However, when studying fixational eye movements, their unpredictability, the idiosyncratic differences among animals, and the range of CRF sizes, make it difficult and tedious to obtain a complete set of cleanly separated landing, leaving, and crossing interactions for each cell. For example, with larger CRF sizes, few of the small fixational saccades cause the CRF to cross cleanly over

the stimulus without either landing on it or leaving it. To overcome these difficulties, we have employed two approaches. First, we analyzed data from all fixational saccades that caused an *increase* in firing in the 250-ms following a saccade. Based on control analyses for 28 cells, we found that including all such "increasing" saccades yielded results that differed only slightly from results based on precisely mapped landing saccades (see Methods). Second, we utilized visually guided voluntary saccades to generate distinct landing and leaving (Figure 2A), as well as crossing (Figure 1C) trajectories (44 cells). In 33 of the 44 cells data were collected for both fixational and voluntary saccades.

Figure 2B shows plots of eye position (superimposed), spike rasters, and spike histograms compiled from repeated behavioral trials for the three cell types. In addition, saccade-triggered rasters and histograms for both voluntary landing and "increasing" fixational saccades are presented (data for fixational saccades were derived from several trials like one shown in Figure 1B). These plots illustrate two main points: the consistency of activation patterns across trials and the similarity of the patterns of activation produced by voluntary and fixational eye movements (cf. black and gray PSTHs, see Supplemental Results for population data). Transient, saccade-activated cells like the one in the top row of Figure 2 responded well to any abrupt change in the CRF, firing briefly following saccades that caused the CRF to land, to leave, or to cross the stimulus. The strength of the response when the CRF left the stimulus was relatively weak for this cell, but it can be seen at about 3000–3500 ms in the histogram in panel B. Saccade-activated cells also responded to small saccades that moved the stimulus within the CRF (notice responses to small fixational saccades while the CRF is on the stimulus; Figure 2A, top row). Consequently, in these cells 69% of all fixational saccades led to an increase of firing. Notably, saccade-activated cells did not continue to fire during drift periods that followed landing saccades, even though the CRF remained on the stimulus. Hence, their transient post-saccadic response was very different from the sustained activation that was observed in the position/drift cells, which continued to discharge as long as the CRF was on the stimulus (Figure 2, bottom row). We refer to this sustained activation during inter-saccadic drift periods as a "position/drift" response since we cannot distinguish how much of the response is caused by the velocities imparted by slow drifts rather than the mere presence of the stimulus in the receptive field. In contrast to the less selective firing of saccade-activated cells, position/drift cells only increased their firing after saccades that landed the CRF on the stimulus or moved it to a more sensitive part of the CRF (51% of all fixational saccades). Mixed cells exhibited both types of responses—post-saccadic bursts and inter-saccadic sustained firing when the CRF overlapped the stimulus (Figure 2B, middle row). Because post-saccadic bursts accompanied landing, crossing, leaving, and "within CRF" saccades, the proportion of fixational increasing saccades, 65%, was larger in mixed cells than in position/drift cells. To summarize, data for fixational and voluntary saccades demonstrate a consistent link connecting neuronal response patterns and the spatiotemporal interactions between the stimulus and the CRF resulting from eye movements.

Quantification of strength of post-saccadic and drift responses

To quantify the relative strengths of the post-saccadic bursts and the maintained firing during drifts on a cell-by-cell basis, we calculated the normalized difference between the firing rates in 150-ms epochs immediately after saccades and firing rates during the rest of the drift periods (saccade-drift difference, SDD, see Methods; Supplemental Figure S1 shows the distribution of SDD). Values of SDD for the entire sample of cells, based on fixational saccades causing increased firing, ranged from -0.4 to 1.2, with 91% of the values falling between 0 and 1. In our previous work, we found that cells could be separated into the three types of eye movement activation based on comparing their responses to landing and to crossing saccades, and the boundaries between these groups corresponded to particular values of SDD (Snodderly et al., 2001). Saccade-activated cells, with strong post-saccadic bursts and little or no discharge in the drift periods, had high values of SDD (>0.7, 25% of present sample); position/drift-activated cells, with comparable burst firing rates and drift firing rates had low values of SDD (<0.3, 38%); and mixed cells (37%) had intermediate values. The choice of these boundaries to distinguish activation types is not critical for present purposes, and we do not imply a clear-cut separation of all three types based on a single index. Rather, we find that V1 neurons have a range of response patterns in post-saccadic and drift periods, with distinctly non-overlapping saccade- and position/drift-activated classes representing the two ends of the range, and mixed cells in between.

Additionally, to provide an assessment of eye movement activation patterns unbiased by choice of specific time epochs, we estimated a cross-covariance function between eye velocity and instantaneous firing rate, using complete records for each cell after excluding blinks (see Methods). Figure 2C illustrates results of this analysis for the same three example cells used in Figures 2A and 2B, for voluntary eye movement trials, and Supplemental Figure S2 presents population data. As expected, for saccade-activated cells, which have transient discharges, the peak of the cross-covariance function was high, sharp, and well localized within time lags matching the expected latency of visual responses. The position/drift-activated cells had either low broad humps spanning a wide range of latencies or no statistically significant peak, and mixed cells had medium-height peaks followed by lower values. The cross-covariance peak amplitude and other parameters derived from this analysis correlated well with the SDD derived from mean rates in specific time epochs ($r = 0.7$ for both fixational and voluntary saccades; $p < 0.0001$; Supplemental Figure S2A). These results confirm the existence of a broad range of response patterns, including many with strong sustained activation during drift periods.

Finally, Figure 3A summarizes the average firing patterns in the perisaccadic period for the three eye movement activation classes, separately for fixational increasing saccades and for voluntary landing saccades. Importantly, cells tested with *both* fixational and voluntary eye movements behaved similarly for the two types of eye

movements (additional comparisons are illustrated in the Supplemental Results).

Although they have usually been ignored, most V1 neurons had considerable drift responses. To illustrate this point, we calculated the ratio of the mean drift-firing rate to the mean post-saccadic firing rate (using the same time periods as in the SDD computation) and expressed it as a percentage. Figure 3B shows the cumulative distribution of cells with drift firing rates achieving specific percentages of the post-saccade firing rate, both for fixational saccades that caused an increase in firing and for voluntary landing saccades. About ¾ of the cells had at least 25% as high a mean firing rate in the drift period as in the post-saccade period, and nearly half the cells had mean firing rates at least 50% as high in the drift period as in the post-saccade period. Comparing different activation types defined by the SDD values, saccade cells fired less than 17% as fast in the drift period as in the post-saccade period (mean $\pm$ *SD* = 4 $\pm$ 11%), mixed cells fired up to 50% as fast in the drift period (38 $\pm$ 18%), and position/drift cells fired more than 50% as many spikes in the drift period as in the post-saccade period (80 $\pm$ 24%). Importantly, the *mean* post-saccadic firing rate of position/drift cells (44 $\pm$ 30 spikes/s) was as high as that of saccade cells (43 $\pm$ 28 spikes/s), so the similarity between post-saccadic and inter-saccadic firing of the position/drift-activated population was not due to low response strength.

Comparison of activation by eye movements with activation by external stimuli

Flashed stimuli vs. saccades: Response transiency

Our working hypothesis is that the different eye movement activation patterns result primarily from interactions between the dynamic input imparted by the eye movements, and the spatial and temporal properties of the V1 neurons. To test this idea, we compared eye movement activation with activation by externally modulated stimuli. When a saccade moves the CRF with respect to a stimulus, there is an abrupt change in flux in the CRF. For comparison with this situation, we have presented cells with a flashed stimulus positioned on the CRF and compared the response with that evoked by saccades that move the CRF abruptly with respect to a steady stimulus. To describe the immediate time course of the response, we used a transiency index (TI), which is similar to the saccade-drift difference, but is based on firing rates in shorter periods of 0–120 ms after the saccade (*peak* of the response) and 120–250 ms after the saccade (*tail* of the response, see Methods). To derive the TI for saccadic activation, the peak was calculated as the mean of the 3 highest 10-ms bins in the 0- to 120-ms post-saccadic
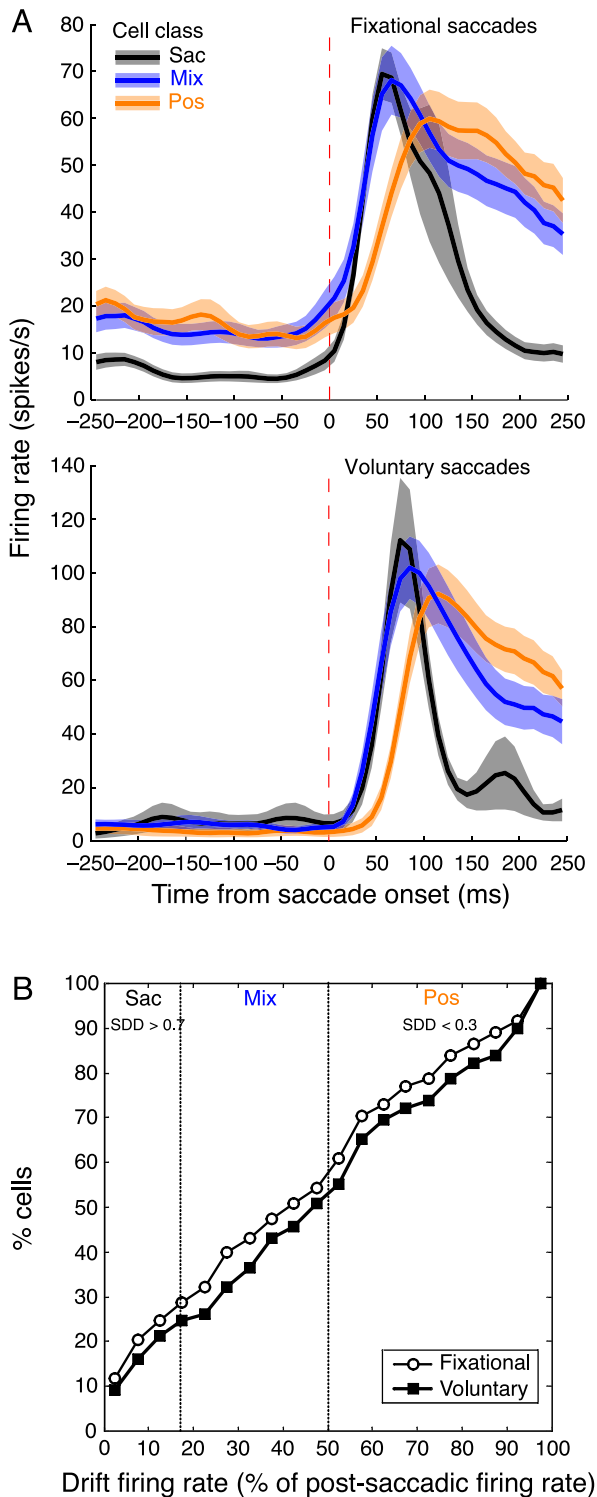

Figure 3. (A) Population perisaccadic averages triggered by fixational increasing saccades for 118 cells (30 *sac*, 43 *mix*, 45 *pos*) and voluntary landing saccades for 44 cells (10 *sac*, 17 *mix*, 17 *pos*). Cells were assigned to eye movement activation classes according to the saccade-drift difference SDD as described in the text. Dashed vertical lines denote saccade onset. (B) Cumulative distribution of V1 cells as a function of the firing rate in the drift period relative to the firing rate in the post-saccadic burst period for fixational increasing saccades (open circles) and voluntary landing saccades (filled squares). Vertical dotted lines mark the borders between cell classes based on the SDD index.

period to accommodate variations in response latency. TI values close to 1 indicate peak $\gg$ tail, i.e., very transient response, and values close to zero indicate that peak and tail are similar because of sustained firing after saccades. For comparison, we analyzed the transiency of responses during drift periods for the same stimulus bar used for saccadic activation, but now the bar was flashed on and off in the middle of the CRF. The otherwise stationary stimulus was moved slightly on each video frame to compensate for fixational eye movements and to maintain the position of the stimulus on the CRF. The same TI formula was used to compute the flash response transiency: values for the peak were derived from the period 0–120 ms after flash onset but values for the tail were calculated for the period 120–150 or 120–200 ms after flash onset (shorter tail periods were used for flashes because many of the flash "on" periods were <250 ms: either 150 or 200 ms).

Figure 4B demonstrates a remarkably good correspondence between the transiency of the responses to flashes and the transiency of activation by voluntary landing saccades, the two most similar conditions ($r = 0.85$, $p < 0.00001$). Similarly, Figure 4A shows that the correspondence between activation by flashes and by fixational saccades that caused an increase in firing is also strong, but because there are more diverse interactions, with some crossing, leaving and "within CRF" fixational saccades in addition to landing saccades, the correlation is slightly lower ($r = 0.79$, $p < 0.00001$). These results indicate that the abrupt retinal image motions imparted by saccades affect neuronal activity in a manner very similar to the abrupt temporal transient of a stationary flashed stimulus. A smaller and slower extraretinal modulation accompanying saccades is considered later in the Results. There was a close correspondence between the saccade-drift difference measure, calculated using entire drift periods, and the post-saccadic transiency index based on shorter perisaccadic periods ($r = 0.74$ and 0.8 for fixational increasing and voluntary landing saccades respectively; $p < 0.0001$; Supplemental Figure S1). This shows a high degree of consistency between the time course of the responses immediately after the saccade and firing rates later in the drift periods.

Flashed stimuli and saccades: Response transiency vs. response latency

Figure 4C illustrates the additional relationship between eye movement activation patterns (measured as the SDD) and the *latency* of the responses to saccades (r ranging from -0.5 to -0.7; see legend for details). This relationship shows an association between the two temporal characteristics, the onset latency and the transiency of the response: low-pass, sustained position/drift-activated cells have longer latency than band-pass, transient, saccade-activated cells. One possible confound to this result could

be that we calculated the SDD using a fixed post-saccadic time interval of [0 to 150 ms], so longer response latencies could trivially cause a smaller post-saccadic firing rate and hence a lower SDD. To exclude this possibility, we re-calculated a variant of the saccade-drift difference that took into account post-saccadic firing rate in the 100-ms interval beginning *at the first significant post-saccadic bin*, i.e., in $[L_{land}$ to $L_{land} + 100$ ms], which should accommodate any variations in latency. The correlation coefficient for re-calculated SDD changed only by 5% ($r = -0.55$ as compared to original $r = -0.58$, $p < 0.001$; re-calculated SDD shown in the Figure 4C, filled squares), demonstrating the validity of this comparison.

Smoothly moving stimuli vs. saccades: Speed selectivity

Another factor determining activation by eye movements was the speed preference of the cells. Figure 5 shows that the speed preference measured with stimuli swept smoothly across the CRF was positively correlated with the saccade-drift difference for the same cells, indicating that cells responding to saccades, but not drifts, did so in part because of their speed tuning. In particular, cells that were activated by the high speeds of voluntary crossing saccades preferred higher sweep speeds ($11.0 \pm 8.1°/s$, $n = 22$) than cells that were not activated ($3.2 \pm 4.3°/s$, $n = 16$, t test, $p < 0.01$). We also found statistically significant correlations between the saccade transiency index, TI, and the direction selectivity index, DI ($r = 0.45$, fixational increasing saccades; $r = 0.57$, voluntary landing saccades; $p < 0.0001$). In summary, cells that preferred faster movement fired more transient discharges were more likely to be activated by crossing saccades, were not activated by drifts, and were more selective for direction of movement.

Spatial selectivity

In the spatial domain, receptive fields of position/drift cells in the parafovea (eccentricity <7°) had smaller mean activating regions than the other two classes ($24 \pm 17'$ vs. $31 \pm 23'$; $p < 0.05$), and a higher frequency of very small (<15′) receptive fields (17/49, as compared to 14/80 for saccade and mixed cells; $p < 0.05$, Fisher's exact test; see also Supplemental Results). The relative numbers of cells responding to only one sign of contrast at each spatial location, i.e., simple and monocontrast neurons, was significantly higher for position/drift cells than for mixed and saccade cells (pos: 40% (17/43) were simple/monocontrast; mix/sac: 18% (13/71) were simple/monocontrast; Fisher's exact test, $p < 0.02$). These data show that position/drift cells are more selective for small spatial features and sign of contrast, which may facilitate encoding stimulus position and fine spatial detail.
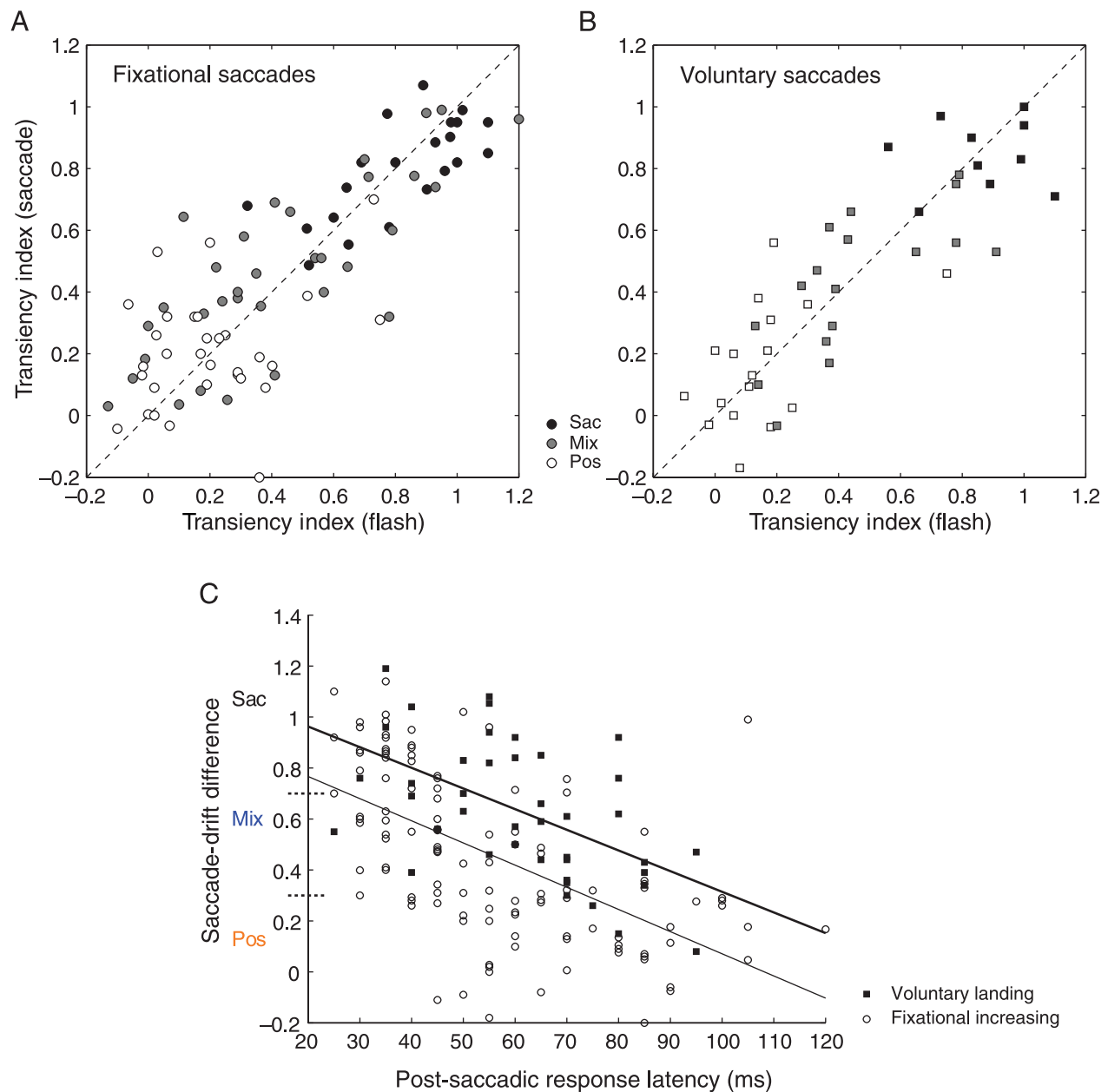
Figure 4. Correlation between temporal patterns of neuronal firing evoked by saccades in the presence of a stationary stimulus and responses to flashed stimuli during drift periods with compensation for eye movements. *Top row:* Transiency Index, TI, for (A) fixational saccades that produced increased firing and (B) voluntary landing saccades vs. stimulation with a flashed bar. Symbols' color denotes cell classification (black: "saccade," gray: "mixed," white: "position-drift"). The line of equality is plotted for comparison. (C) Correlation between latency of response to saccades and saccade-drift difference, SDD. Short dashed horizontal lines on the vertical axis denote SDD borders between cell classes. Response latency was estimated as the first of two consecutive 5-ms bins significantly above the pre-saccadic baseline (exceeding 2.5 *SD* confidence limits). There was a significant correlation ($r = -0.58$, $p < 0.001$) between the SDD and the latency of response to voluntary landing saccades (filled squares), with saccade cells having the shortest latencies (54 ± 15 ms) and position/drift cells the longest latencies (74 ± 15 ms). A similar relationship was found for fixational increasing saccades (open circles; $r = -0.68$; $p < 1e^{-7}$). Least-square fits are shown for voluntary landing (thick line) and fixational increasing saccades (thin line). Flash response latency (not shown) was also highly correlated with voluntary landing saccade response latency ($r = 0.78$, $p < 1e^{-7}$) and SDD ($r = -0.5$, $p < 0.001$), as well as fixational saccade response latency ($r = 0.58$ $p < 1e^{-7}$). See S1 for response latency in each eye movement activation class for fixational increasing and voluntary landing saccades, and Supplemental Methods for discussion of the issue of temporal jitter in estimated saccade onset times.
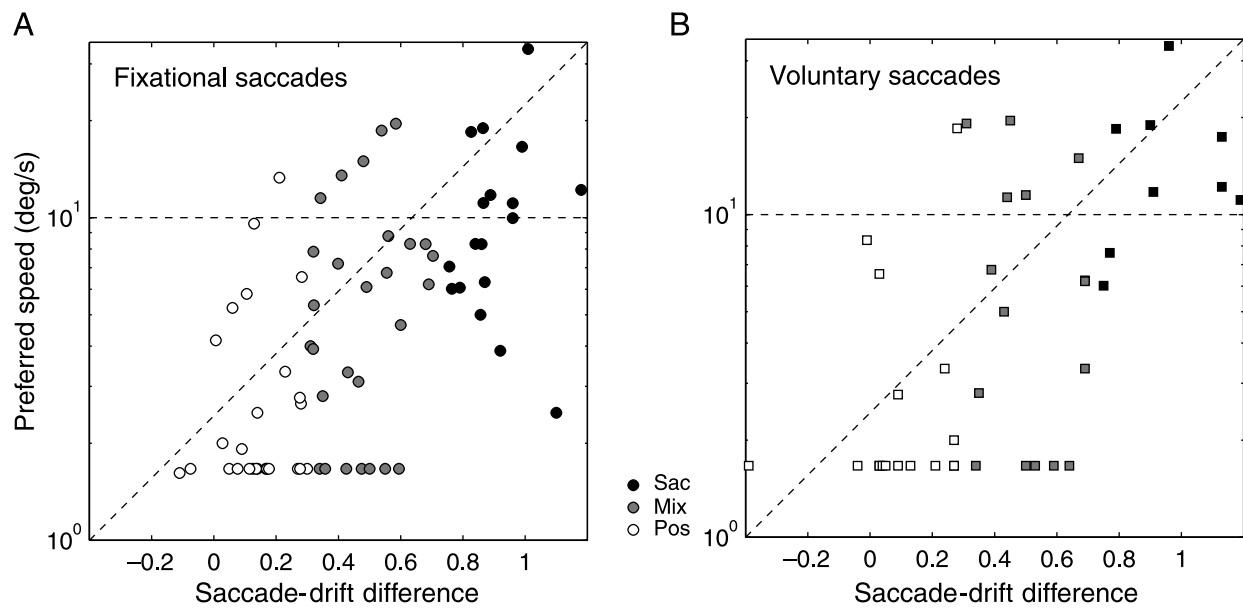
Figure 5. Speed tuning peak for an optimally oriented sweeping bar vs. saccade-drift difference in the firing rate evoked by (A) fixational increasing saccades for the same bar when it was stationary (118 cells) and (B) voluntary landing saccades (44 cells). Note that stimulus motion was imposed in addition to compensation for fixational eye movements, so the speed of the image on the retina was precisely controlled. For comparison, the horizontal dashed lines indicate the 10°/s threshold eye velocity used to detect fixational and voluntary saccades. The correlation between SDD and the preferred speed was highly significant for both voluntary saccades ($r = 0.59$, $p < 0.0001$) and fixational saccades ($r = 0.62$, $p < 0.00001$).

### Flashed stimuli vs. saccades: Response strength

Finally, to evaluate the strength of responses evoked by saccadic eye movements, we compared peak firing rates of responses to flashed stimuli with the firing rates caused by a steadily illuminated stimulus moved about the receptive field by saccades. Figure 6A illustrates peak firing rates for fixational saccades, voluntary landing saccades, and voluntary crossing saccades of different amplitudes, normalized by flash firing rate for each cell before averaging. Values close to 1 demonstrate that the peak firing rates for flashes and for landing saccades, the most similar conditions, were remarkably comparable in all three eye movement classes (gray bars). As in previous sections, only fixational saccades that led to increases in neuronal firing were selected because this subset comprised mostly landing, "within CRF", and crossing trajectories that activated the cell. Activation by fixational saccades was slightly lower but still at least 60% of the flash response (purple bars). However, there was a major difference in the effects of crossing saccades on different cell types (blue bars). Saccade-activated cells responded fairly well to crossing saccades of amplitudes up to 5° (the largest size tested), mixed cells' response was considerably weaker, and most position/drift cells did not respond at all to crossing saccades of any size. In fact, only 2/17 cells classified as position/drift according to their SDD values had a weak response to crossing saccades. For all activation types, larger crossing saccades caused smaller responses. Although limits imposed by the video frame rate (160 Hz, 6.25 ms) could contribute to

this effect, it is unlikely to be the entire explanation, given the CRT phosphor persistence. Furthermore, weak responses of V1 neurons to fast-moving stimuli have previously been well established using classical optics (Judge et al., 1980). These results indicate that small saccades causing the CRF to cross stimuli are more effective than large saccades in activating V1 neurons of all eye movement classes.

The similarity between the responses to flashes and the activation by fixational saccades contrasts sharply with a prior report indicating that flashes are ~7 times as effective as fixational saccades for V1 neurons (Martinez-Conde et al., 2002). That report was based on saccade-triggered or flash onset-triggered average spike probability for all saccades and all cell types (but based on 6 cells). For comparison, we performed the same computation, combining data from *all* fixational saccades (causing either increase, decrease, or no change in firing) and all eye movement activation types. Figure 6B shows, for both monkeys, that activation by fixational saccades reached at least 50% of the response to flashes, even when data from all saccades and all cell types were averaged (thin black curve). In terms of the time course, the average post-saccadic responses in Figure 6B have less sharp peaks than the flash responses because the latency was more variable. Also, when data were included only for those saccades that caused increases in firing (thick black curve), there was a small increase in the post-saccadic burst magnitude, and the drift response of position/drift and mixed cells became evident in the later part of the drift period. Spike

probability stayed above the pre-saccadic baseline for increasing saccades, but returned to baseline for "all" saccades. This happened because inclusion of saccades that cause both increases and decreases diluted the sustained drift response, underlining the need to separate different saccadic effects for a comprehensive analysis.
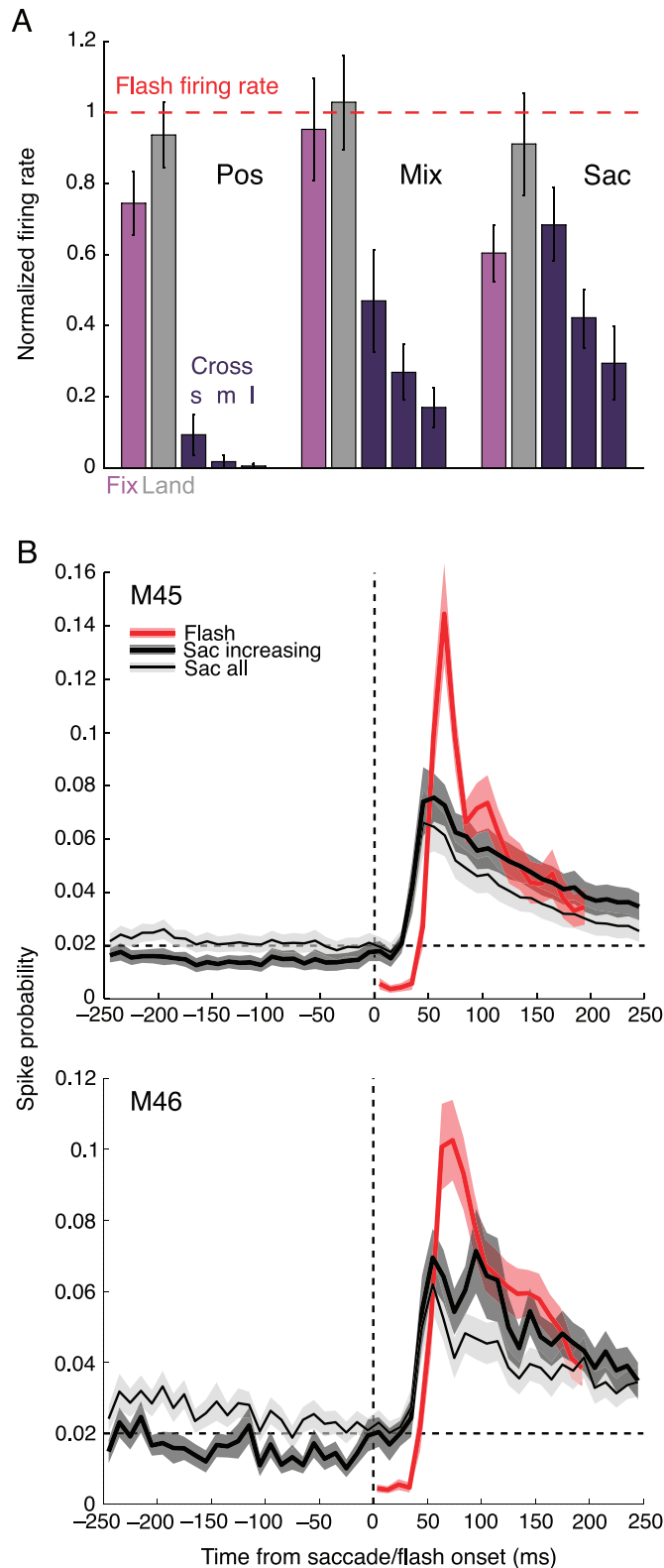

Extraretinal modulation of ongoing activity by fixational and voluntary saccades

Experiments described in this section were done in the absence of a deliberate visual stimulus, either with a lighted uniform monitor screen, or in the dark (see Methods). Figures 7A and 7B illustrate the basic result with data from 5 individual cells showing that both fixational and voluntary saccades in the light and in the dark had a considerable effect on the firing rate. The perisaccadic modulation of the ongoing firing typically had a biphasic time course—initial weak suppression followed by stronger enhancement peaking 100 to 200 ms after saccade onset.

To quantify suppression and enhancement on a cell-by-cell basis, we calculated suppression (SI) and enhancement (EI) indices and assessed the significance of the modulation for each neuron (Figure 7B; Methods). SI and EI are integrals of areas in the saccade-triggered histogram that are below the ongoing firing rate (SI) or above it (EI), normalized by the integral of the baseline ongoing rate. If individual bins of the histogram exceeded ±2.5 SD of the baseline, the modulation was considered statistically significant. Cells exhibiting significant extraretinal modulation were common in M45 and M46, the two monkeys with less frequent fixational saccades (see Figure 8). For fixational saccades in the dark, 31% (26/83) of the cells were significantly modulated, as were 34% (86/250 cells) of the cells recorded in the light. Furthermore, in some statistically non-significant cases visual inspection still indicated presence of a clear modulation. Even in M42, the monkey with the least frequent occurrence of extraretinal modulation, 18% (6/34) of the cells had significant extraretinal modulation following fixational saccades in the light. However, the time courses of individual responses were more variable

Figure 6. Comparison of firing rates in response to flashes and to saccades. Peak firing rate values are based on the mean of the three highest 10-ms bins in the 0- to 120-ms period of the initial transient of the response. (A) Peak firing rates were normalized by flash response (horizontal dotted line). Crossing saccades were grouped as small (*s*, amplitude ≤1.5°), medium (*m*, 1.5° to 3°), and large (*l*, from 3° to 6°). Error bars denote standard error. (B) Fixational saccade- and flash-triggered population averages of spike probability for two monkeys. Spike probability was estimated in 1-ms time bin and averaged across 10-ms bin. Shaded regions represent $\pm SEM$ of each time course. Note that the apparent difference in "flash-to-saccade" ratios between panels A and B is explained by larger variations in the latency of the post-saccadic peak as compared to flash responses (see S1). This results in "smearing" the post-saccadic peaks in panel B but not in panel A, where the post-saccadic firing rate was estimated for each cell individually using the three highest 10-ms bins rather than averaging with a fixed latency before normalizing and averaging across cells.
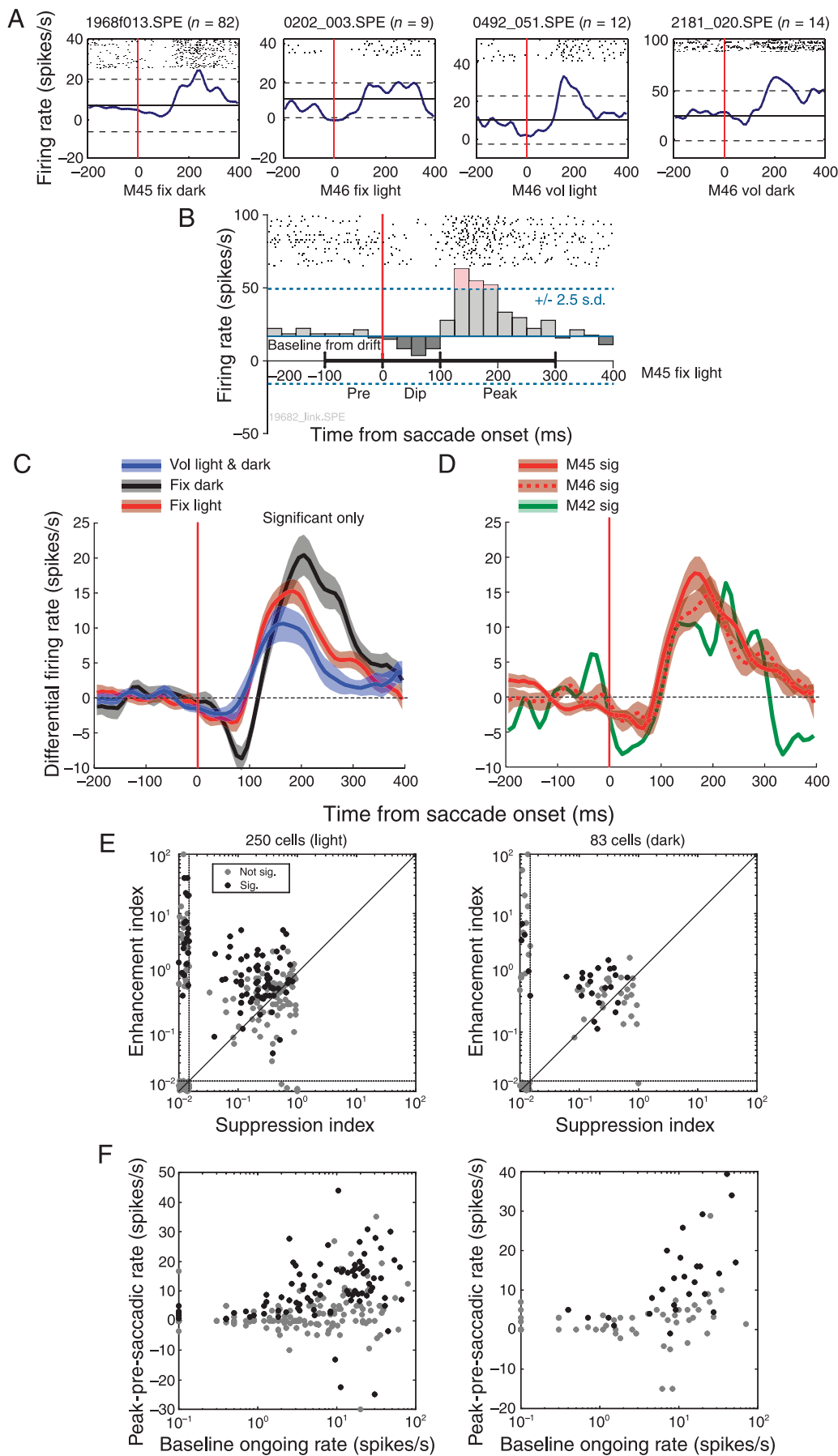
and inconsistent, which may be related to the higher frequency of fixational saccades in this monkey (see later). For voluntary saccades, the majority of the cells studied

Figure 7. Extraretinal modulation of neuronal firing associated with saccades. (A) Saccade-triggered averages of firing rate of 4 single neurons in two monkeys, binned at 10 ms and smoothed with Gaussian kernel (σ = 15 ms). An example histogram from another cell in panel B shows parameters used to quantify the extraretinal modulation and judge its statistical significance. Time 0 is saccade onset (vertical dashed line). Nerve impulses recorded from one cell during 30 fixational saccades in the light are indicated in the raster display at the top of the panel. The ongoing firing rate (the baseline) was estimated from 600-ms epochs in drift periods extending backward from 50 ms before the saccade (not shown). For the test of modulation significance, a saccade-triggered histogram was constructed using 25-ms bins. Firing rate increments above the baseline are shaded light gray, decrements—dark gray. Modulations of firing rates were considered significant if they exceeded ±2.5 *SD* of the ongoing rate (horizontal dashed lines). Three characteristic periods describing the extraretinal modulation are labeled below the horizontal axis as the *pre*-saccadic period, the period showing a *dip* in the firing rate, and the period containing the *peak* post-saccadic enhancement. (C) Population saccade-triggered averages of extraretinal modulation by fixational and voluntary saccades for cells with a statistically significant modulation. The ongoing firing rate has been subtracted. There was a statistically significant difference between the latency of the post-saccadic enhancement in the dark (170 ± 31 ms, median 162.5 ms) and in the light (142 ± 34 ms, median 137.5 ms) for M45 (*p* < 0.05, one-way ANOVA for multiple comparisons), but no difference between these measures and latency in the light for M46 (153 ± 50 ms, 137.5 ms). (D) Population saccade-triggered averages of extraretinal modulation by fixational saccades in the light, for significantly modulated cells plotted separately for each of three monkeys. For M42 significantly modulated cells (*n* = 6), shaded area representing *SEM* is omitted for clarity. (E) Scatter plots of enhancement vs. suppression indices for light (left) and dark (right) conditions. Black dots indicate cells with significant modulation; gray dots indicate cells whose modulation did not reach the criterion for statistical significance. Data points with one or two zero indices were jittered on zero-index axis between 0 and 0.005 (dashed horizontal and vertical lines) for presentation purposes. Scatter plots comparing the magnitude of the "peak" and "dip" of the extraretinal modulations are shown in S3. (F) Correlation between the enhancement strength and the ongoing rate for light (left) and dark (right) conditions. Each dot represents data from a single cell as in (E). Note that because of low ongoing firing rates in many cells, a correlation is inherent for the strength of suppression, since cells with no ongoing rate cannot exhibit a decrease in firing and thus will be "less suppressed" than cells with higher ongoing activity. However, such a correlation is not inherent in the strength of enhancement, which was nevertheless correlated with the ongoing firing rate.

showed significant extraretinal modulation (14/21 cells in the light and 3/3 cells in the dark).

Figure 7C plots the average time course of the extraretinal modulation by fixational and voluntary saccades in the light and in the dark for all cells in M45 and M46 for which there was a significant effect. For fixational saccades in the light, Figure 7D compares average data for all cells showing a significant modulation from M45, M46, and M42. Clearly, the modulation is very similar in all three monkeys.

Figure 7E presents scatter plots demonstrating the predominantly *enhancing* effect of extraretinal modulation, separately for fixational saccades in the light and in the dark. Most significantly modulated cells (black circles) show more enhancement than suppression. Based on the average perisaccadic time course of the modulation (Figure 7C), we calculated mean firing rates for three time periods as shown in Figure 7B: a pre-saccadic interval 200 ms before saccade onset, a post-saccadic interval from 0 to 100 ms after saccade onset encompassing the *dip* in neuronal firing and an interval 100 to 300 ms after saccade onset designed to include the *peak* of the enhanced firing. Next we tested whether the strength of the modulation was related to the ongoing rate. We considered two possible outcomes: an additive contribution that would be invariant of the ongoing rate; or the modulation could depend on the ongoing rate, implying a more complex interaction with a multiplicative component. In favor of the latter, Figure 7F reveals a statistically significant correlation between the ongoing rate and the enhancement strength expressed as the difference between mean firing rates in the *peak* post-saccadic periods and the *pre*-saccadic periods (note that the enhancement *strength* is not normalized like the enhancement index). Similarly, the percentage of cells showing significant extraretinal modulation (either suppression or enhancement) was higher in cells with higher ongoing rates. In the light, only 26% of the cells with an ongoing rate <7 spikes/s showed significant modulation (39/149 cells), as compared to 46% (47/101) of cells with an ongoing rate ≥7 spikes/s. Analogous values were found for the dark condition (17% vs. 47%). Interestingly, 29/71 cells (light) and 11/25 cells (dark) with ongoing rates ≤1 spikes/s did fire a few spikes in the post-saccadic period, indicating that the enhancement may include an additive component (Supplemental Figure S4).

We did not find a significant relationship between strength of extraretinal modulation and receptive field class (simple, complex or monocontrast; Kagan et al., 2002), the type of eye movement activation (saccade, position/drift, mixed), or the size of the saccade (Supplemental Results). Instead, the detection of the extraretinal modulation may depend on the individual patterns of fixational eye movements. Figure 8 compares metrics of fixational eye movements in two monkeys that showed the robust extraretinal effect (M45 and M46) and one that showed only a weak effect (M42), demonstrating that
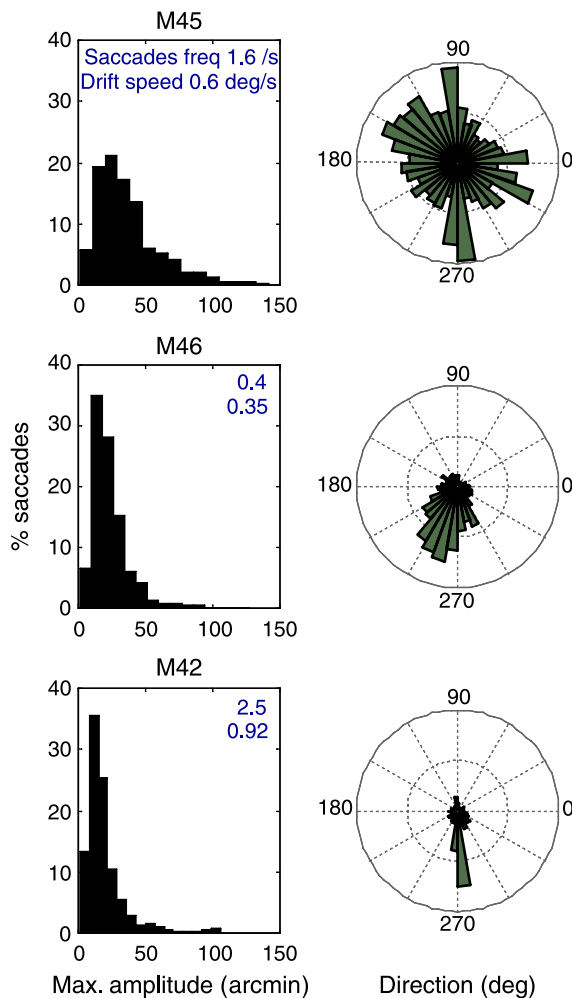
Figure 8. Metrics of fixational eye movements of the three monkeys studied for this paper, recorded while viewing a light screen. M42 shows a weaker extraretinal effect associated with fixational saccades than the other two monkeys. *Left column:* Distributions of maximal displacement amplitudes of fixational saccades. The blue numbers in each panel denote the frequency of fixational saccades (saccades/s) and the drift speed (°/s). *Right column:* Distributions of saccade directions. See S2 for other metrics, and S7 and S14 for detailed metrics of fixational saccades and drifts in monkeys M45 and M46.

frequent saccades accompanying faster drifts may diminish the apparent modulation (Supplemental Results).

Differences between extraretinal influences and stimulus-driven effects

Figure 9 illustrates how the slow biphasic time course of the extraretinal influence differs from the responses evoked by near-optimal visual stimuli. Figure 9A compares an averaged perisaccadic histogram for fixational

saccades that caused increased firing in the presence of a steadily illuminated stimulus with averaged responses from the same cells with no stimulus present. Figure 9B presents the corresponding comparison for voluntary landing saccades. The stimulus-evoked response in both situations is larger and faster than the extraretinal modulation. However, we note that the stimuli were chosen to be near-optimal for the cells, so in a natural environment, there will be many cells encountering suboptimal stimuli that may not be so much more effective than the extraretinal modulation.

We tested whether extraretinal influences play a role in shaping the saccade-evoked activity even when a strong visual stimulus is present. We looked specifically for differences in the time course of transient responses to voluntary landing saccades and to flashes. Figure 9C shows that in transient cells with $TI_{flash} > 0.6$ the response to the landing eye movement lasts longer than the response to a flash (TI_{flash} 0.91 ± 0.13, TI_{land} 0.75 ± 0.18; $n = 13$, $p < 0.05$), which suggests a post-saccadic enhancement. Moreover, the difference between time courses of flash and saccadic responses, plotted in Figure 9D, was similar to the form of the biphasic extraretinal modulation of ongoing activity (cf. Figures 7C and 7D). These modulations in neuronal activity associated with saccadic eye movements are candidate mechanisms for contributing to changes in stimulus visibility associated with saccades (see Discussion).

Discussion

We investigated the activation of V1 neurons by fixational and voluntary (visually guided) eye movements, and we dissociated effects fast retinal image motion and changes in retinal position caused by saccades from effects of slow motions occurring in drift periods. A summary of our findings and their relationship to previous work is given in Table 1. The effects of voluntary and fixational saccades were largely equivalent. For both fixational and voluntary saccades, neurons could be grouped into three response classes: complementary saccade-activated and position/drift-activated cells responding exclusively to fast or to slow motion, and mixed cells responding to both. Consistent with their perceptual importance, strong sustained position/drift responses were present in large numbers of neurons. We also established that eye movement activation was strongly correlated with multiple response properties of cells, including transiency of response to flashes, speed tuning, response latency, and spatial selectivity. We found that eye movements that activate the cells elicited responses that were similar in strength, time course, and tuning to responses evoked by flashed and by externally moving stimuli. In addition to these strong stimulus-driven
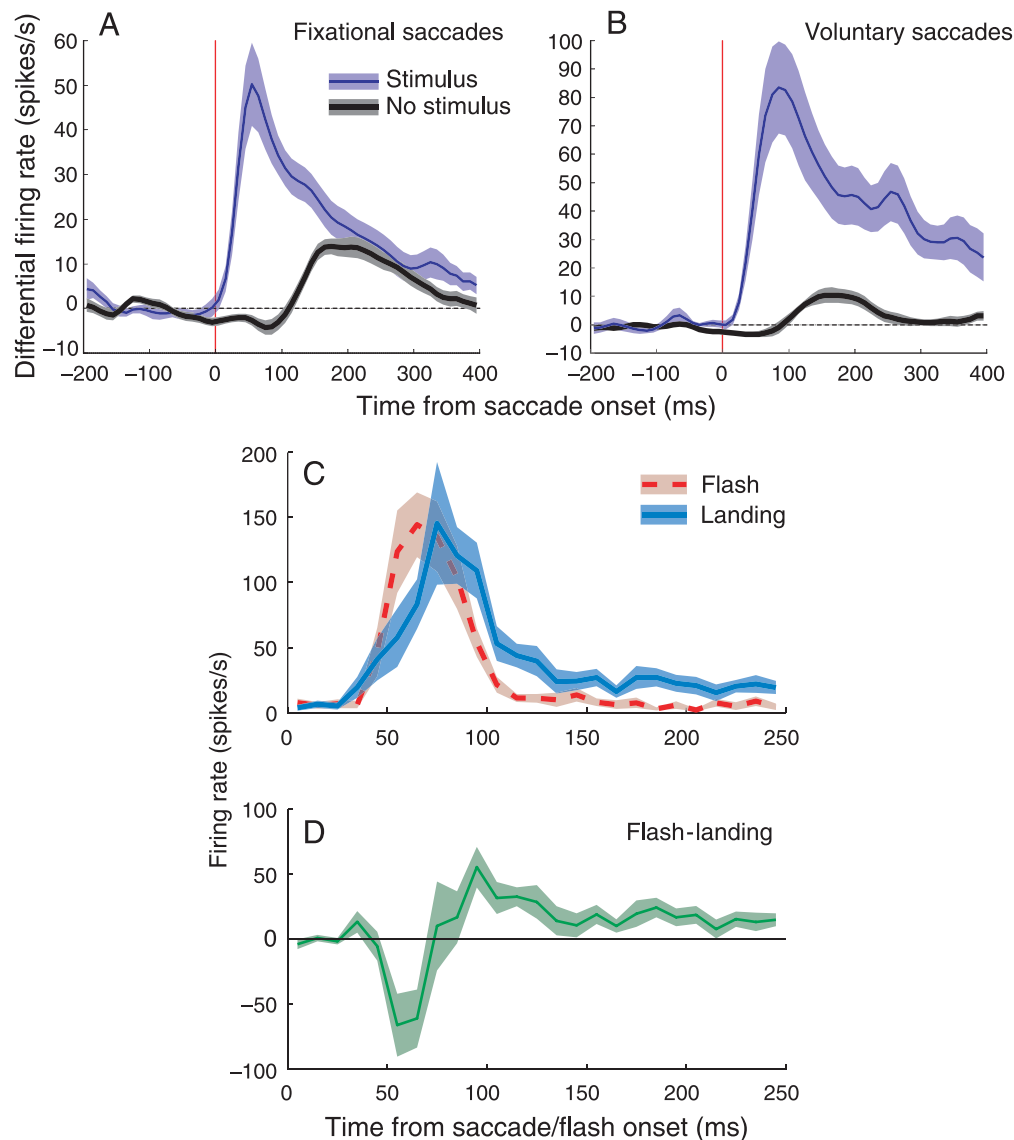
Figure 9. (A–B) Comparison of stimulus-driven and extraretinal influences. Population saccade-triggered averages for fixational increasing (A) and voluntary landing (B) saccades with and without a stimulus. The ongoing rate has been subtracted. For this figure, extraretinal influences measured in the light and in the dark have been combined. Bin width 10 ms, convolved with Gaussian of σ 15 ms. Shaded regions represent $\pm SEM$ (A) $n = 27$ significantly modulated cells out of 73 tested with and without stimulus; 25 in light, 10 in dark. (B) $n = 15/22$ cells, 14 light, 3 dark. See S5 for cell-by-cell analysis of "peak" firing rates with and without visual stimulus. (C–D) Comparison between time courses of responses to prolonged flashes of an optimal stationary bar (flash duration at least 250 ms) and responses to voluntary landing saccades. (C) Mean post-saccadic and post-flash activity for a subset of 13 transient (sac and mix) neurons with $TI_{flash} > 0.6$. (D) Mean differential (post-saccadic − post-flash) activity for the same 13 neurons. The differential activity was calculated separately for each neuron and then averaged across neurons. Note the similarity of the differential time course to the time course of ongoing activity modulation (cf. Figure 7). Latency differences between flash and landing conditions in the onset of the visual stimulus on the CRF cannot account for the observed difference in the time course because the mean response latency difference (L_{land} − L_{flash}) for all 13 cells was only 3 ± 12 ms, and only in 5/13 cells was L_{land} longer than L_{flash} by 16 ± 7 ms. See Supplemental Methods for evidence that eye position sampling rates were not important factors in determining response latency values.

effects, we identified extraretinal modulation in about a third of our sample. In the rest of this section, we relate our results to previous neurophysiological and psychophysical findings and propose how different components of eye movement activation may contribute to visual processing.

The high prevalence of position/drift responses in V1

Approximately two-thirds of V1 neurons, the saccade cells and the mixed cells, discharge bursts of spikes whenever the CRF crosses, leaves, or lands on a stationary

Results	Major conclusions and comparisons with previous work
Three firing patterns result from dynamic interactions between stimulus and spatiotemporal CRF caused by saccades and drifts:	Cells activated only by saccades are a minority. Sustained drift responses present in many cells. Fixational and voluntary saccades evoke similar firing patterns.

Saccade cells	Position/drift cells	Mixed cells	Previous work: Did not identify drift-related activation. No comparison of fixational and voluntary saccades.
Transient bursts after crossing, landing, and leaving saccades. No response during drifts.	Sustained firing during drifts while CRF on the stimulus. Weak or no response to crossing saccades.	Bursts after saccades. Weaker sustained firing during drifts.	

Activation by eye movements correlates with responses to external stimuli that provide similar spatiotemporal stimulation of the CRF:	Sustained drift responses may provide basis for encoding fine spatial detail. In V1, effects of external and self-imposed retinal image motions can be described in the same spatiotemporal framework.

Saccade cells	Position/drift cells	Mixed cells	Previous work: Did not compare spatiotemporal properties of neurons to eye movement effects.
Transient, short-latency response to flashes. Prefer high speeds.	Sustained, longer-latency flash response. Prefer slow speeds. Often have smaller, sign-of-contrast selective CRFs.	Intermediate flash response properties and speed preferences.	

Strength of saccade-evoked activity is similar to flash responses. Fixational increasing saccades: 50–90% of flash response strength. Voluntary landing saccades: 90–100% of flash response strength.	Post-saccadic responses constitute large components of visually evoked activity, even during fixation. Previous work: Similar results for large voluntary saccades. Much weaker activation by fixational saccades than by flashes.
Extraretinal modulation accompanies both fixational and voluntary saccades in 1/3 of neurons. Modulation is biphasic: initial weak suppression is followed by stronger enhancement.	Extraretinal modulation is widespread in V1, even during fixation. It may contribute to saccadic suppression and post-saccadic updating of visual inputs. Previous work: Similar results for larger voluntary saccades. Did not detect extraretinal modulation with fixational saccades.

Table 1. Summary of results and relationship to previous work (for references, see Discussion).

stimulus. These burst of spikes have been the exclusive focus of several physiological investigations of fixational eye movements (Bair & O'Keefe, 1998; Livingstone et al., 1996; Martinez-Conde et al., 2000, 2002). However, during the drift periods, a comparable number of cells, the mixed cells and the position/drift cells, give sustained discharges that continue as long as the stimulus remains on the CRF (Figure 2); these selective sustained responses have been ignored by other investigators. A critical requirement for eliciting the maximal sustained response is to place the stimulus accurately on the CRF. We satisfied this requirement by obtaining a precise measure of the CRF size and retinal location while compensating for fixational eye movements before assessing the effects

of the eye movements themselves. Another important step is to separate the drift periods into ones following saccades that cause an increase in firing from those that do not. When no increase occurs, it means one of three things:

1. the saccade has moved the CRF off the stimulus entirely;
2. the saccade has moved the CRF so that the stimulus falls on a less sensitive part of the field; or
3. the CRF has not encountered the stimulus at all.

If these conditions are included in the overall average, the true drift response is diluted (Figure 6B) and it may be

missed altogether. Importantly, the activity during drift periods is not caused by undetected saccades (as has been proposed by Martinez-Conde et al., 2002), because (1) even with a 100-Hz eye position sampling rate, short saccades are detected because most saccades are "step-like" and result in a net change in position; (2) most of the data for this paper were collected with 1–2 arcmin resolution at 200 Hz, which is more than adequate to detect fixational saccades (cf. Horowitz, Fine, Fencsik, Yurgenson, & Wolfe, 2007; Leopold & Logothetis, 1998); (3) even assuming that very small saccades might occasionally go undetected, this would not explain the fact that position-specific sustained responses are associated with some neuronal classes, and not others. An extensive treatment of sampling rate and saccade detection issues is provided in Supplemental Methods, including detailed metrics of saccades and drifts.

Comparison of eye movement activation with external stimulation

We found a close correspondence between neuronal responses to external stimuli such as flashing and sweeping bars, and the responses evoked by eye movements. The most similar situations are a stimulus flashed on the CRF and a saccade that abruptly lands the CRF on the stimulus. When voluntary saccades were elicited to produce this comparison, the peak responses were nearly identical to peak flash responses (Figure 6A, landing saccades). Differences in the time course of the responses (Figure 9C) may be due to the extraretinal influences discussed later. These results agree with a recent study that found equal or even slightly higher responses to voluntary saccades than to flashes, especially in the later part of the response (MacEvoy, Hanks, & Paradiso, 2008).

The comparison between effects of fixational saccades and the effects of flashed stimuli is less precise because of the diversity of fixational saccades, but peak responses evoked by saccades that caused increases in firing were 60–95% as strong as responses to flashes (Figure 6A). Given that the majority of fixational saccades (62%) led to increased firing, our results underscore the significant contribution of fixational eye movements to visual responses, making the eye movements a major source of variability in responses to moving bars (Gur, Beylin, & Snodderly, 1997; Gur & Snodderly, 2006) and to drifting gratings (Kagan et al., 2002).

Our results are not consistent with the conclusions of Martinez-Conde et al. (2002), who reported that responses to flashing bars are *7 times larger* than responses to stationary bars present during fixational saccades. Part of the discrepancy can be ascribed to their inclusion of all saccades, whether they increased or decreased neuronal firing. However, even when we performed the same analysis, using averaged spiking probability following

any fixational saccade as the measure of response strength, we still obtained *at most* a 2-fold difference between post-saccadic bursts and peak flash responses (Figure 6B). The reasons for the much lower estimates of saccadic effectiveness by Martinez-Conde et al. are unclear, but they may include the small sample size (6 cells) used for the comparison, fixational saccade/receptive field sizes, and their behavioral paradigm. In our experiments, the monkey was required to attend to a fixation target and press a lever while the CRF stimulus was presented extrafoveally. Under our conditions, mean eye position and visual responses are very stable from trial to trial (Tang et al., 2007). In the experiments of Martinez-Conde et al., the monkey was not required to perform a specific task, but data were recorded whenever the monkey's eye position was within a predefined window. Under those conditions, flashed stimuli might evoke a bias in eye position or acquire a salience that would enhance the flash response relative to the activation by stationary, unchanging stimuli. Enhanced responses to flashed stimuli as compared to static stimuli displayed during saccades have been demonstrated in parietal area LIP (Gottlieb, Kusunoki, & Goldberg, 1998).

The other externally controlled stimulus that we used was a smoothly moving bar that crossed the CRF. The speed preferences of neurons for the moving stimulus were consistent with the patterns of activation by eye movements. Neurons that preferred faster moving stimuli were more likely to be activated by saccades. Conversely, cells strongly activated by drifts were likely to prefer more slowly moving external stimuli. These results confirm predictions made previously that cells termed "velocity low pass" would be especially sensitive to the slow speeds imparted to retinal images by fixational eye movements (Orban, Kennedy, & Bullier, 1986).

Comparison of fixational saccades and voluntary saccades of different sizes

One of the robust effects in our data was a decrease of response amplitude to crossing saccades of increasing size. We interpret this as another effect of stimulus speed. Limitations of our video stimulation system did not allow speeds comparable to large crossing saccades (200–300°/s, see Supplemental Figure S12); the maximal external bar speed that we used was 36°/s. Nevertheless, our findings are consistent with results from early studies of the effects of eye movements on V1 neurons that used classical optics to produce high stimulus speeds (Judge et al., 1980; Wurtz, 1969a). Many cells responded poorly or were suppressed by rapid motion of the retinal image across the CRF, whether it was caused by movement of the eyes or by external motion of the stimulus (Fischer, Boch, & Bach, 1981; Judge et al., 1980). These results led to the conclusion that large saccades caused suppression due to

the rapid image motions (Fischer et al., 1981; Wurtz, 1969b), which is consistent with our interpretation.

Functions of fixational saccades and drifts

Transient firing after fixational saccades is common and is easy to detect, but the function of the fixational saccades that generate these bursts has been hotly debated (Ditchburn, 1980; Kowler & Steinman, 1979, 1980) and remains controversial. One point of view is that fixational saccades function to prevent image fading, but at present, there is no causal evidence that fading elicits counteracting fixational saccades. It has been shown that there is a correlation between the rate of fixational saccades and the probability of stimulus re-appearance during prolonged fixation in conditions of Troxler fading, but saccades preceded only a fraction of re-appearances (Martinez-Conde et al., 2006). In similar experiments, monkeys signaled the perceptual or physical disappearance of a stimulus under conditions of the General Flash Suppression illusion (Jie Cui, Wilke, Logothetis, Leopold, & Liang, in press). The saccade rate was lower during *both* perceptual disappearance and physical target removal conditions as compared to a "no disappearance" condition, suggesting a potential motor preparation/attentional component in the suppression of fixational saccades. Similarly, the suppression of fixational saccades in humans following salient cues has been reported by several authors and interpreted to reflect motor preparation and/or attention (e.g., Engbert & Kliegl, 2003; Hafed & Clark, 2002; Horowitz et al., 2007; Valsecchi, Betta, & Turatto, 2007). Other experiments with humans found no effect of *externally imposed* stimulus fading on the frequency of fixational saccades (Poletti & Rucci, 2007). Moreover, typical natural fixation periods are short (<0.5 s). Taken together, these results suggest that in the absence of a significant retinal motion/change, in particular during conditions of prolonged fixation, fixational saccades may help to refresh a faded image in the periphery. However, fixational saccades are not part of a feedback system to regulate image visibility. Otherwise saccade frequency should increase when the contrast of images fades, either as a consequence of external manipulations or of internal neural dynamics.

The frequency and distribution of fixational saccades need to be considered together with the characteristics of drifts, because a portion of fixational saccades may be triggered to correct the retinal displacements caused by drifts (Cornsweet, 1956; Engbert & Mergenthaler, 2006; Gur & Snodderly, 1997; Nachmias, 1959; Skavenski et al., 1975). For example, in our monkeys, the prevalence of *downward* saccades was related to the incidence and speed of *upward* vertical drift (Figure 8; Supplemental Table S2; also cf. Supplemental Figures S7 and S14). In the foveal region, slow drifts may provide enough changing input for enhancing the visibility during fixation

bouts (Rucci & Desbordes, 2003; Rucci et al., 2007). Under natural viewing conditions, fixational saccades are less frequent and they are interspersed with small and large voluntary saccades (e.g., Malinov, Epelboim, Herst, & Steinman, 2000; Poletti & Rucci, 2007; Steinman, Pizlo, Forofonova, & Epelboim, 2003). Although fixational and voluntary saccades have been usually treated as separate phenomena, our results show that fixational and small voluntary saccades produce similar effects in V1, including extraretinal effects (see below). Therefore, the question whether fixational saccades are mostly a "laboratory phenomenon" (as argued by Steinman and colleagues) becomes less critical. Moreover, whether a fixation bout was initialized by a fixational or a voluntary saccade, the ensuing drift would activate a subpopulation of V1 cells, providing a neuronal basis for continuous visual experience. In the absence of a clearly defined fixation target, drifts can be more substantial and, combined with head movements, may contribute even more to the perception of a scene during natural viewing conditions. Together, voluntary saccades, fixational saccades, and drifts shape neuronal activity, generating and reducing spatiotemporal correlations among neurons to allow for efficient representation of natural scene statistics (Ahissar & Arieli, 2001; Desbordes & Rucci, 2007).

Extraretinal modulation

The presence of extraretinal modulations of the ongoing discharge of V1 neurons was initially demonstrated by studying the effects of large voluntary saccades made in the dark (Duffy & Burchfiel, 1975) or in a uniform field (Kayama et al., 1979). Our results confirm those observations and extend the analysis to the more difficult case of fixational saccades. In our previous averaged data, the extraretinal effect accompanying fixational saccades was not apparent in one of two monkeys studied (Snodderly et al., 2001), and another laboratory observed no extraretinal modulation with fixational saccades (Martinez-Conde et al., 2002). To resolve this issue, we analyzed data from three monkeys on a cell-by-cell basis to determine whether differences among animals were due to differences in the relative numbers of cells that show an effect. We found that extraretinal influences accompanying both fixational and voluntary saccades were present in about a third of our total V1 sample, but cells showing the effect were less frequently encountered in a monkey with a high saccade rate than the other two monkeys. The extraretinal influences were much more prolonged than saccades, indicating that they are not just mirror images of short-lived motor commands (corollary discharges). Furthermore, the strength of the extraretinal modulation increased with the cells' ongoing rate, suggesting that this modulation is "activity dependent." The predominantly enhancing effect is consistent with fMRI data from human lateral geniculate nucleus (LGN) and V1 (Sylvester,

Haynes, & Rees, 2005), both of which showed a positive BOLD signal in response to saccades made in the dark.

Figure 10 compares the time course of extraretinal modulation in monkey V1 with extraretinal modulation in the monkey LGN (panel A) and extraretinal modulation in human perceptual sensitivity (panel B). All these functions are biphasic, with suppression followed by enhancement. The time course of the enhancement component of the extraretinal modulation in V1—both in the light and in the dark—is much slower than the time course in LGN studies that used full-field flash stimulation (Ramcharan et al., 2001; Reppas et al., 2002) and cannot be explained by latency differences in visual responses (~25 ms LGN, ~50 ms V1). Another LGN study using a dark background reported much slower enhancement, similar to our V1 data (Royal et al., 2006). The difference
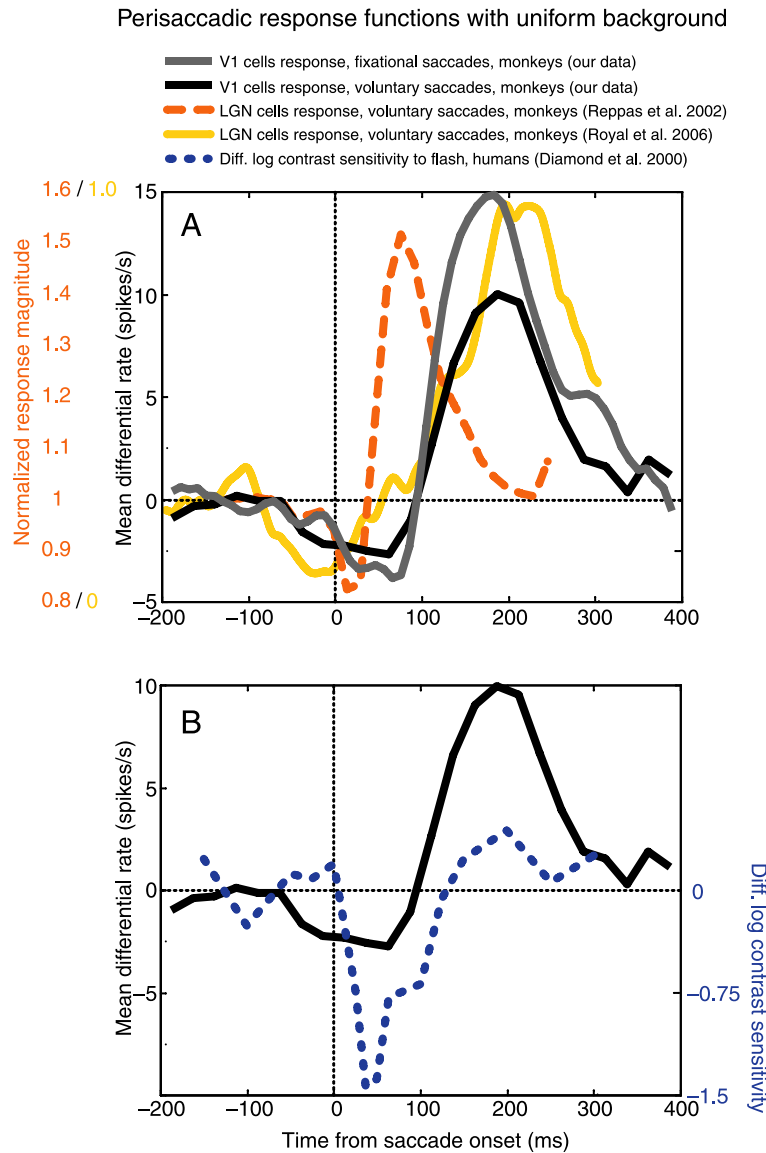

Figure 10. Comparison of extraretinal perisaccadic modulation at different levels of visual processing. Saccade onset-triggered averages with a uniform lighted background in each case. (A) Modulation of neuronal firing by saccades in the presence of a blank field. Gray curve, V1 cells, our data, differential firing rate associated with fixational saccades, mean values from 2 monkeys. Black curve, differential firing rate associated with voluntary saccades. Dashed orange line, monkey LGN normalized firing rate (data from Reppas et al., 2002). Solid yellow line, monkey LGN normalized firing rate (data extracted from Figure 9A of Royal et al., 2006, using WinDig software). All neuronal data are for cells showing significant modulation. (B) Comparison of monkey V1 data with extraretinal modulation of human perceptual thresholds. Black curve, V1 extraretinal modulation associated with voluntary saccades copied from A. Dashed blue line, human psychophysical data: differences between log contrast sensitivity at times relative to a real saccade and relative to simulated saccadic motion for a flashed grating (2 human subjects averaged, data from Diamond et al., 2000). The human data have been shifted rightward by 50 ms to account for visual processing delay.

between LGN visual stimulation regimes (full-field flash vs. dark background) may have contributed to the discrepancy between these reports. Given these rather striking differences, it is difficult to say how the LGN modulations are transformed in V1. At least some of the long-latency activation in V1 may be driven by descending inputs to V1 from higher-order cortical areas (cf. Toyama, Komatsu, & Shibuki, 1984), but both the initial suppression and later enhancement could include LGN influences as well. In area LIP, the averaged time course of perisaccadic response functions without a stimulus in the CRF is remarkably similar to V1 (Heiser & Colby, 2005).

Analogous modulations of human perceptual sensitivity associated with saccades have been reviewed by Ross et al. (2001), and the most pertinent example is reproduced in Figure 10B (dotted blue curve). These data show the human contrast sensitivity for a flashed grating presented at various times relative to the onset of a large voluntary saccade compared to fluctuations in sensitivity evoked by externally imposed motion that simulated the retinal image motion produced by the saccade (Diamond, Ross, & Morrone, 2000). However, since the neurophysiological data represent a modulation of sensitivity at the cortex, and the psychophysical data represent a modulation of sensitivity relative to the time that a stimulus falls on the retina, an approximate retina-to-cortex processing delay of 50 ms has been added to the psychophysical data. A qualitative correspondence between suppression and enhancement in two types of data is apparent. One cannot conclude from this comparison whether the extraretinal modulations in V1 are major determinants of the perceptual effect, but they probably play a role, and at least they must affect the activity of downstream cortical areas that may also be involved.

Fixational saccades in V1: Enhancement or suppressive effects?

Finally, we address an apparent discrepancy between results from different labs regarding the sign of effect of fixational eye movements in V1: Livingstone at al. (1996) and Martinez-Conde et al. (2000, 2002) show only enhancement of the firing following fixational saccades, but Leopold and Logothetis (1998) reported mostly suppression of V1 activity. Our findings include both effects and suggest that the discrepancy can be explained by different visual stimuli and possibly by different neuronal samples. Leopold and Logothetis used continuously viewed grating stimuli that produced high *sustained* firing of V1 cells and restricted their analysis to very small saccades (10′ median) that caused relatively little change in light flux within the receptive field. Consequently, they reported a time course very similar to our extraretinal effect on the ongoing firing (cf. Figure 7). Moreover, the long latency (∼100 ms, cf. our Figure 5) and sustained

time course of responses to a flashed stimulus (their Figure 2A) suggest that their V1 sample may have been mostly position/drift and mixed cells that would show suppression with little post-saccadic enhancement when the CRF was moving *within* the stimulus. Consistent with this suggestion, we showed that firing of some low-pass position/drift cells was transiently interrupted if the saccade was small enough so that the receptive field stays on the stimulus (Snodderly et al., 2001). These considerations suggest that the Leopold and Logothetis (1998) results do not contradict the presence of an enhancement following fixational saccades under most stimulus conditions.

Conclusions

The two types of visual activation in V1 by eye movements are mediated by different neuronal populations that can encode complementary information. Post-saccadic bursts accompanying fixational and voluntary saccades signal abrupt change or motion in the CRF and can be utilized to detect salient features like edges irrespective of sign of contrast and current spatial position. When viewing complex stimuli such as natural scenes, widespread neuronal subsets throughout the retina will discharge synchronously after saccades, signaling the occurrence of an eye movement as distinguished from local responses that signal object movement. During natural vision, the post-saccadic transient effects are interspersed with maintained firing during the ubiquitous inter-saccadic drift periods. Depending on the amplitude of saccades and drifts, the functional significance of these activations will be a function of eccentricity. In the periphery, fixational saccades may help to refresh the scene and contribute to visibility. The slower drifts activate cells with complementary characteristics, maintaining continuous visual experience and providing selective information about location and fine spatial detail that is especially important in retinal regions nearer the fovea (Rucci et al., 2007). Biphasic extraretinal modulations that accompany both voluntary and fixational saccades suppress inputs near the times of the saccades, and slightly later enhance sensitivity, which could facilitate post-saccadic updating of the visual input.

Acknowledgments

Footnote

[1]"Fixational saccades" vs. "microsaccades" and "voluntary saccades." We use the term "fixational saccades" because the alternative term "microsaccades" is ambiguous. Different labs define "microsaccades" in the amplitude range from <12′ (e.g., Steinman et al., 2003) to <60′ (e.g., Engbert & Mergenthaler, 2006) or as much as 120′ (Martinez-Conde et al., 2006). The amplitudes of small voluntary saccades can overlap with the range of amplitudes of fixational saccades (e.g., Malinov et al., 2000), and the amplitude of fixational saccades can significantly differ between subjects, training history, and task/stimulus conditions (for details, see Figure 8 and Supplemental Results). We use the term "voluntary" saccades to distinguish instructed saccades from the non-instructed saccades that occur during fixation.

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Supplemental Material for "Saccades and drifts differentially modulate neuronal activity in V1: effects of retinal image motion, position, and extraretinal influences" by Kagan, Gur and Snodderly

Supplemental Methods, Supplemental Results, 3 Supplemental Tables, 14 Supplemental Figures, 3 Supplemental Movies

Supplemental figures are numbered from S1-S14 and supplemental tables are numbered from S1-S3. Figures S1-S5 are referred to in the main body of the paper. Other supplemental figures are discussed below.

Supplemental Methods

Sustained responses are not caused by undetected fixational saccades

Sustained position/drift responses to fixational eye movements (Snodderly et al., 2001) have been previously questioned on grounds of a potential failure to detect small saccades (Martinez-Conde et al., 2002). This is primarily an issue for a smaller subset of our data that was collected with a 100 Hz sampling rate. The suggestion that undetected saccades are the source of activity during drifts is implausible for several reasons:

1. A 100 Hz sampling rate is sufficient to detect most fixational saccades and map their end-points. A simple calculation shows that a 10°/s velocity threshold will allow detection of >6' (minarc) position shift within 1 sample interval (10 ms). The percentage of detectable small saccades depends on the saccade duration and approaches 100% for amplitudes >10' (Figure S6). Comparison of data with 100 Hz and 200 Hz sampling rates shows that *at the maximum 4%* of all saccades have amplitudes <6' and would be missed in 100 Hz recordings (Figure S7, *cf.* main sequences for M45 and M46). A real number for monkey M45 is probably even lower because she tended to make larger saccades than M46. The majority of the fixational saccades in our recordings (both with 100 Hz and 200 Hz) lasted at least 20 ms or more and had amplitudes >10' (Figure S7). The shortest 5 ms saccades, detected with 200 Hz rate (first red 5-ms bin in the duration distribution for M46), comprised only 2% of all saccades. But it does not mean that we failed to detect all <10 ms duration saccades with 100 Hz. Most fixational saccades are "step-like", with a net change in position (Figures S8, S9, S10), so even if we missed the exact onset of a short saccade, we would detect a position change in the next sample. Examples of raw eye position records of fixational saccades with 100 Hz and 200 Hz sampling are shown in Figure S9 and Figure S10, and metrics of fixational saccades and drifts are shown in Figure S7 and Figure S14.

2. Most data for the present paper was collected with a new experimental system, using an eye coil and sampling frequency of 200 Hz, which is adequate for studying small fixational eye movements.

A number of previous studies of fixational eye movements used a 200 Hz sampling rate (*e.g.* Horowitz et al., 2007; Leopold and Logothetis, 1998), 250 Hz (Horwitz and Albright, 2003) and many human studies used video-based eyetrackers with sampling rates close to 200 Hz, *e,g,* 250 Hz (Moller et al., 2002; Engbert and Kliegl 2003), 240 Hz (Hafed and Clark, 2002). Monkey M46 had a low frequency of only 0.4 saccades/s with eye position records sampled at 200 Hz, but exhibited strong sustained position/drift responses in completely saccade-free intervals, in both fixational (Figure S10) and voluntary (Figure 2) saccade trials.

3. The overall pattern of our findings clearly demonstrates that even if we missed some rare saccades, it still would not invalidate any of our results. First, there is a clear difference between neuronal classes in cells collected with same sampling rate. Saccade cells do not respond at all during drift periods, even when the CRF is clearly staying on the stimulus, while position/drift cells that exhibit sustained activation during drift periods do not respond at all to crossing saccades. If we missed some saccades, we would have missed them in both neuronal classes, and it would not explain why position-specific sustained responses occurred for some cells but not others. Second, for the potentially undetected tiny saccades to generate the observed levels of the sustained firing during "presumed" saccade-free drift periods, we would have to assume a completely unrealistic occurrence of saccades approximately every 150 ms.

These arguments show that undetected saccades are not the basis for activation during drift periods.

Estimation of neuronal response latency to saccades is not compromised by "insufficient" (100 Hz / 200 Hz) eye position sampling rate

A potentially important effect of a lower eye position sampling rate is a higher variation in estimation of the saccade onset time (up to 5 or 10 ms, Figure S6). This jitter could presumably result in a temporal blurring of post-saccadic response peaks determined by saccade-triggered averaging. Yet, one of our results is that we find very strong post-saccadic responses, almost equal to flash responses (where the onset is known with 1 ms precision), while another lab that used 1000 Hz sampling rate found 7-times weaker post-saccadic responses than flash responses (Martinez-Conde et al., 2002). Another study that employed an even lower, 60 Hz, sampling rate of eye position also reported near-equal response peaks for post-saccadic and flash responses (MacEvoy et al., 2008). These findings suggest that our temporal jitter in determining saccade onset is not sufficient to blur the post-saccadic response. We also report a high correlation (r=0.78) between response latencies to saccades and flashes, on a cell-by-cell basis, and a systematic relationship of post-saccadic and flash response latency to other independent characteristics, such as eye movement activation class, transiency and speed tuning. This additionally demonstrates the validity of our saccade response latency estimation. To summarize, the temporal jitter of the saccade onset times up to 5 or even 10 ms is small compared to the combined variations in saccade duration (Figures S7 and S12) and neuronal response latency (~40-70 ms, Table S1), and it does not introduce a systematic bias because positive and negative "shifts" average out (Figure S6).

Supplemental Results

Correspondence between results obtained from fixational and voluntary saccades during visual stimulation

In the subset of cells tested both with fixational and voluntary eye movements (n=33), there was a good correspondence between transiency index TI (r=0.82, p <0.01), SDD (r=0.88, p <0.01) and cross-covariance function estimates derived from the two conditions. When the classification of eye movement activation based on saccade-drift difference was performed independently for fixational and voluntary eye movements, only 5/33 cells received different assignments (pos<->mix, mix<->sac; there was never a confusion between saccade- and position/drift grouping. These 5 cells were recorded in monkey M46, which had a very low frequency of fixational saccades, so the number of fixational saccade events for these cells was smaller (7±2) than the number of voluntary landing saccades (10±3). Given the small number of fixational saccades and the more controlled conditions with voluntary saccades, we used assignments derived from the voluntary landing saccades for these 5 cells.

Correlation between receptive field size and eye movement activation

The correlation between SDD and CRF size was strong in one monkey where we collected most data within a relatively narrow range of eccentricities (r=0.48, p <0.001, all but one cell within 1.3-6.2°, mean±s.d. 5.3±1.4°, M45), but not significant in another monkey where we sampled a broader eccentricity range (0.6-10°, mean±s.d. 3.9±1.7°, M46).

condition/cells	pos	mix	sac
fixational increasing post-saccadic response latency	72±20 (31)	50±16 (33)	38±8 (21)
flash response latency in corresponding cells	62±16 (31)	59±12 (33)	49±8 (21)
voluntary landing post-saccadic response latency	74±15 (17)	54±16 (17)	54±14 (10)
flash response latency in corresponding cells	71±17 (17)	57±9 (17)	52±11 (10)

Supplemental Table S1. Neuronal response latency following saccades and flashes, mean±s.d. in ms (number of cells in parentheses).

Extraretinal effects

Statistics of fixational eye movements influences detection of extraretinal modulation

The lower incidence of extraretinal modulation in cells recorded from M42 compared to the other two monkeys ($p<0.05$, Fisher's exact test) prevented detection of its presence in our previous study (Snodderly et al., 2001), because for each monkey we averaged data across all cells, thereby diluting the effect. Nevertheless, the question remains why the monkeys differ. To illustrate the differences among monkeys, the metrics of their fixational eye movements are summarized in Figure 8 and Table S2. M42 had a higher velocity of upward vertical drift that had to be compensated with frequent small downward saccades; whereas the drift velocity was lower and saccades less frequent in other two monkeys. It is unlikely that the saccade direction determined the occurrence of modulation, since most saccades in both M46 and M42 were downward, but they were more frequently accompanied by detectable extraretinal modulation in M46 than in M42. These results are consistent with the suggestion that saccade frequency and drift rate affect the detectability of the extraretinal modulation and are the likely determinants of the individual differences.

Pre-saccadic suppression

On the population level, using averages across cells, we find little evidence of pre-saccadic suppression. The elevated baseline noticeable in the averaged data recorded in the light in M45 is most likely the artifact of overlapping influence from preceding saccades (Figure 7D). This monkey had a higher frequency of fixational saccades (Figure 8), therefore we re-did the analysis with more stringent conditions of no previous saccade in the 500 ms epoch prior to any selected saccade, and the pre-saccadic baseline became flat (data not shown). Very weak pre-saccadic suppression is also noticeable in monkey M46 for voluntary saccades. In general, some individual neurons did appear to show a weak suppression before the saccade onset, but this was not a consistent pattern across neurons (*cf.* Reppas et al., 2002). However, random fluctuations in the ongoing firing and low levels of ongoing activity in many cells could have contributed to the masking of this potential weak effect.

Effects of different saccade sizes on extraretinal modulation

We explored the possibility that saccade size influences the strength or the time course of the modulation. Fixational saccades were divided into three groups: amplitude <20', between 20' and 40', and >40'. Voluntary saccades were divided into four groups: <40', between 40' and 170', between 170' and 300', and >300'. In no case did we detect a statistically reliable distinction between modulation signals caused by various saccade sizes. This lack of dependence of extraretinal influences on saccade size is consistent with the results of Reppas et al. (2002) for LGN neurons. These results make it unlikely that that gaze shifts (Rosenbluth and Allman, 2002), rather than saccades *per se*, modulated the ongoing firing, since the effect was not dependent on saccade size and could be observed for movements less than 20'. Moreover, even after controlled voluntary saccades, when the gaze direction remains constant until the next saccade, the modulation signal returns to the baseline approximately in 300 ms, arguing against the possibility that gaze-dependent variations could account for the observed transient perisaccadic modulations.

monkey	mean saccade frequency (sac/s)	mean smoothed drift speed (°/s)	saccade amplitude mean (minarc)	saccade amplitude median (minarc)
M45	1.6	0.60	37	31
M46	0.4	0.35	24	20
M42	2.5	0.92	20	15

Supplemental Table S2. Metrics of fixational eye movements of three monkeys recorded while viewing a light screen (no stimulus present). Note that monkey M42 had a higher frequency of fixational saccades (corresponding to the mean drift duration 419 ms, median 330 ms) and higher drift speeds. Please refer to Figures S7 and S14 for more metrics of fixational eye movements in monkeys M45 and M46, and details of drift velocity calculations.

parameter	condition	mean	median	s.d.
saccade amplitude (minarc)	light	37	31	24
	dark	32	29	16
saccade velocity (°/s)	light	51	49	25
	dark	40	37	18

Supplemental Table S3. Light vs. dark condition, fixational saccade metrics, monkey M45.

Supplemental References (not listed in the main text)

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Supplemental Movies

These movies (Flash *.swf format, please open in the browser) show a single behavioral trial recording (~5 s) for each of three eye movement activation classes: saccade (movie1), position/drift (movie2), and mixed (movie3) cells. In each movie, the dynamic display on the left shows the stationary stimulus (green bar), moving CRF (blue ellipse) and instantaneous firing rate in 100 ms time bins (color-coded vertical bar). Audio stream represents the neuronal firing – each "click" corresponding to a spike. The lower panel shows the corresponding trial record in the same format as used in Figures 1 and 2.

Supplemental figure legends

Figure S1. Relationship between transiency index (TI) and saccade-drift difference (SDD) for V1 neurons. Each point represents data from one cell averaged for 5-143 (mean 19±20) saccades. **(A)** Fixational saccades (118 cells). **(B)** Voluntary landing saccades (44 cells). Symbols' color denotes cell classification (black: "saccade-activated", gray: "mixed", white: "position/drift-activated"). Histograms on x and y axes show frequency distributions of SDD and TI, respectively. Vertical scale bars correspond to 20 cells in A and 8 cells in B.

Figure S2. Cross-covariance analysis. **(A)** Scatter plot of eye velocity – firing rate cross-covariance function peak vs. SDD, for fixational (n=118, circles) and voluntary (n=44, squares) eye movements. Symbol colors denote cell classification (black: "saccade-", gray: "mixed", white: "position/drift-activated" cells). The parameters derived from the cross-covariance analysis correlated well with the SDD: r=0.7, fixational; r=0.68, voluntary; p <0.01. The peak of the cross-covariance function did not reach p<0.05 significance in 40 position/drift and mixed cells, because of the strong firing in drift periods, which did not co-vary with eye velocity (*i.e.* saccades) but rather with retinal image position. **(B)** Means and s.d. of cross-covariance peak values, minimal latency (first significant lag), and peak latency (the lag corresponding to the peak of cross-covariance function) for sac, mix and pos cell classes derived from cross-covariance analysis.

Figure S3. Scatter plots for "peak" and "dip" minus pre-saccadic firing rates for light **(A)** and dark **(B)** "extraretinal" conditions. In the light, mean dip minus pre-saccadic rate for significantly modulated cells was -3.3±9.3 spikes/s, mean peak minus pre-saccadic rate 10.2±10.4 spikes/s; in the dark, -4.5±5.8 and 12.9±10.2 spikes/s, correspondingly.

Figure S4. Saccade-triggered averages from cells with ≤1 spikes/s ongoing activity that had a non-zero positive peak of extraretinal modulation. **(A)** 29 cells recorded in the light condition (mean ongoing firing 0.3±0.3 spikes/s; **(B)** 11 cells recorded in the dark (0.3±0.3 spikes/s). The shaded area indicates s.e.m.

Figure S5. Extraretinal modulation with no stimulus is much weaker than near-optimal stimulus-evoked responses. Scatter plots of post-saccadic mean "peak period" firing rates with and without a stationary stimulus for fixational saccades (**A,** n=27 significantly modulated cells out of 73 tested in both conditions, with and without stimulus; 25 in light, 10 in dark) and voluntary saccades (**B,** n=15/22 cells, 14 light, 3 dark). Each circle represents a datum from one cell with significant extraretinal modulation, either in the light (gray) or in the dark (black), or in both conditions. Two points are shown for cells with both light and dark conditions. Same cells as in [Figure 9](#)A, B.

Figure S6. Simulations of the effect of eye position sampling rate on estimation of saccade onset and detection of small saccades. (**A)** Examples of simulated "actual" eye position ("sampled" at 1000 Hz) and 100 Hz samples acquired at 9 different temporal offsets around the "saccade", and corresponding velocity profiles. Threshold for saccade detection 10°/s. Note the temporal jitter introduced by the 100 Hz sampling rate to the estimated saccade onsets (as compared to "actual" onsets for 1000 Hz traces). This jitter can be negative (saccade onset delayed) or positive (saccade onset advanced). The distribution of these shifts for the upper row example in A is shown in **B**. Since the shifts are both positive and negative, the net inaccuracy in the estimation of the saccade onset times across many saccades would be small. Note that depending on the temporal offset and the duration of the saccade and the sampling intervals, very small saccades can be detected or missed (**A**, rows 2 and 3). Missed instances are marked by solid orange threshold lines in the velocity panels. Panel **C** shows the percentage of detected saccades as a function of saccade amplitude, for 5-15 ms saccade durations and for 100 Hz and 200 Hz sampling rates. Mean detection curves calculated across all saccade durations as a function of amplitude are shown in gray (200 Hz) and black (100 Hz). All saccades >6' and >12' will be detected with 200 Hz and 100 Hz rates, respectively.

Figure S7. Eye position signals sampled at 100 Hz allow reliable detection of small fixational saccades of monkeys. Metrics of fixational saccades for eye position sampled at 100 Hz (M45; top two rows) or 200 Hz (M46; bottom two rows). Data in both cases were collected during presentation of a stationary bar. In both monkeys, saccade **directions** were distributed unevenly (bin size 10°, *cf.* Bair and O'Keefe 1998). The 10°/s velocity threshold for saccade detection resolves >6' saccades at 100 Hz and >3' saccades at 200 Hz. Thus even very small and short saccades, occurring within one sample (10 ms and 5 ms, red 5 ms bins in **duration** distribution histograms) were detected in both cases. The dense lines in the **main sequence** scatter plots (right column, rows 1 and 3) are the result of the sampling quantization – they represent the fixed relationship between the amplitude and the velocity for saccades whose duration was one sample. We did not use up-sampling of eye position traces as we wanted to present the original raw data for inspection. Note the similarity of main sequence slopes in both monkeys (see also Figure S13). The saccades in M46 were on average smaller than in M45 (see distributions of **overall and maximal amplitudes**, column 1, row 2 and row 4, bin size 2'); but smaller saccades are not the outcome of a faster 200 Hz sampling rate (*cf.* monkey M42 data at 100 Hz in Figure S13: mean amplitude 20'). Most saccades had equal overall (from start- to end-point of the trajectory) and maximal (from start- to farthest-point of the trajectory) amplitudes, indicating that circular or away-and-back saccades were rare (see also Figure S8). See Figures S9, S10, S11 for examples of raw eye position traces. Figure 8 shows a subset of similar saccade metrics for these monkeys in the experimental conditions when no stimulus was present on the screen (fixation in the light), used for the analysis of the ongoing activity.

Figure S8. Most fixational saccades in our study had a step-like trajectory, not circular or away-and-back spike-like waveforms. The histograms and cumulative plots in **(B)** show the distribution of the difference

between maximal 2D vector amplitude during the saccade and the overall 2D saccade amplitude measured at the end-points of the saccadic trajectory, expressed in % of the maximal amplitude. For an ideal step-like saccade **(A, top)**, the difference should be 0%, for an ideal circular or spike-like (away-and-back) saccade **(A, bottom)** – the difference should be 100%. In both monkeys, with both eye tracking systems (M45 – 100 Hz Purkinje eye tracker; M46 – 200 Hz eye coil) most saccades have 0% difference, and only a small fraction has values different from 0%, mostly within 10% to 50% (similar to **A, middle)**. These data show that at least 95% of saccades under our experimental conditions result in a substantial change in eye position that would be detected even if the onset of the saccade escaped detection.

Figure S9. Examples of raw eye position traces, velocity calculation and 2D trajectory during fixational saccades, in monkey M45 (100 Hz sampling rate) and in monkey M46 (200 Hz sampling rate). For each monkey, 4 saccades are shown. The first two columns illustrate very small saccades (<10'). Columns 3 and 4 illustrate larger straight and looping saccades, also shown in complete trial recordings in Figure S10. Time stamp 250 ms – saccade onset, ±100 ms epochs around the saccade onset are shown. Amplitude and maximal amplitude (in minarc) are shown above each 2D trajectory plot.

Figure S10. Examples of raw eye position traces and automatic saccade detection, in monkey M45 (100 Hz sampling rate) and in monkey M46 (200 Hz sampling rate). For each monkey, two complete 5-s fixational eye movement trials are shown, one for "position/drift-activated" and one for "saccade-activated" cells. A stationary bar was placed in the center of the CRF, and fixational eye movements moved the CRF over and around the bar. Green curves – horizontal eye position, magenta curves – vertical eye position. Orange inverted triangles denote "increasing" saccades (saccades that resulted in increased firing rate, see Methods), yellow – "decreasing", empty – "no effect" saccades. Long vertical dashed lines denote saccade onsets as found by the automatic saccade detection algorithm. Saccades denoted by *S9* labels are shown in Figure S9 on a more expanded scale. Drifts denoted by *S11* labels and light gray rectangles are shown in Figure S11 on an expanded scale. Short blue vertical lines along time-axis represent neuronal spikes. Note the contrasting neuronal firing patterns in the two cell types (*cf.* Figure 2A). In particular, note the sustained firing during slow drift periods, which is not associated with any saccade. In the example saccade cell in M46 (lower panel), the initial burst marked by **(*)** at the beginning of the trial is caused by the acquisition saccade (prior to trial onset, not shown), and the few spikes not associated with saccades are attributed to ongoing firing (5 spikes/s as assessed by separate recordings of spontaneous activity in the light without a visual stimulus).

Figure S11. Examples of raw eye position traces, velocity calculation and 2D trajectory during fixational drifts, in monkey M45 (100 Hz sampling rate) and in monkey M46 (200 Hz sampling rate). For each monkey, two drift periods are shown. Left column (panels 1) for each monkey illustrates drift periods during which strong sustained responses were recorded from "position/drift" cells – see Figure S10, light gray epochs. Right column (panels 2) shows additional drift examples. Three different drift velocity estimates are
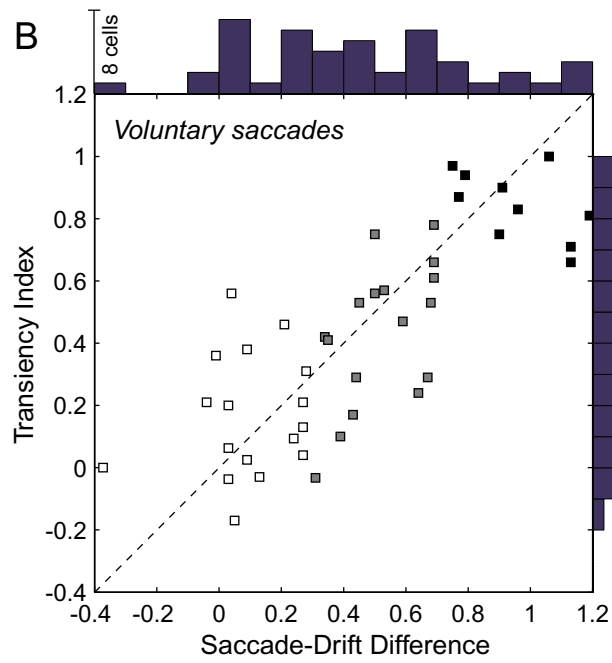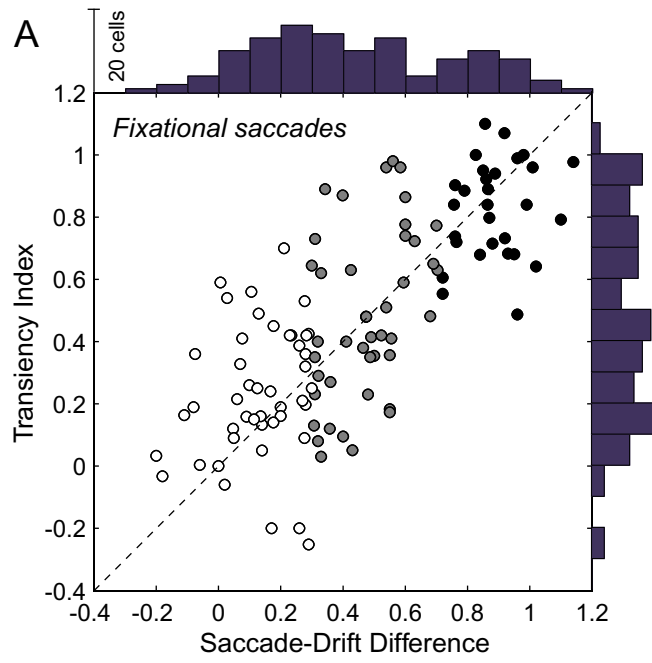
illustrated: "per sample" – using raw horizontal and vertical eye position records, "smoothed" - raw horizontal and vertical eye position records were linearly interpolated to 1 KHz and smoothed with a Gaussian filter of 10 ms σ before taking derivatives to obtain a smoothed velocity estimate, and "dir hor/ver" – per sample "directional" velocity calculated separately for horizontal and vertical eye position.
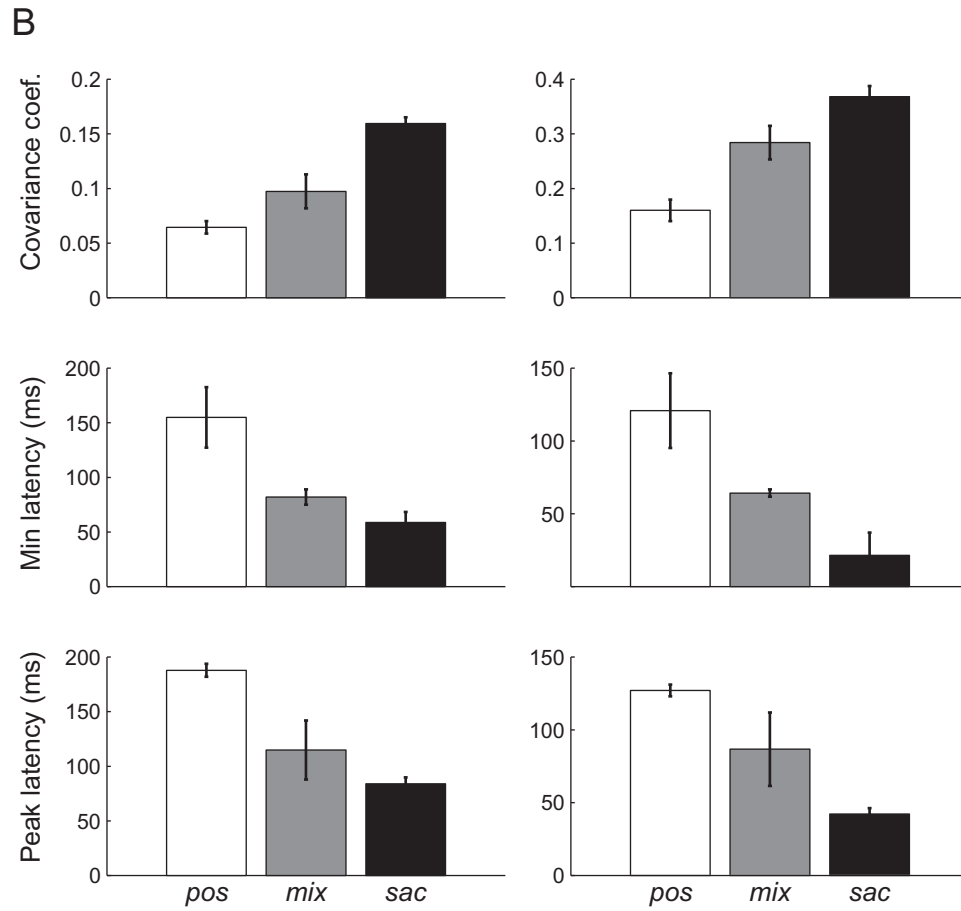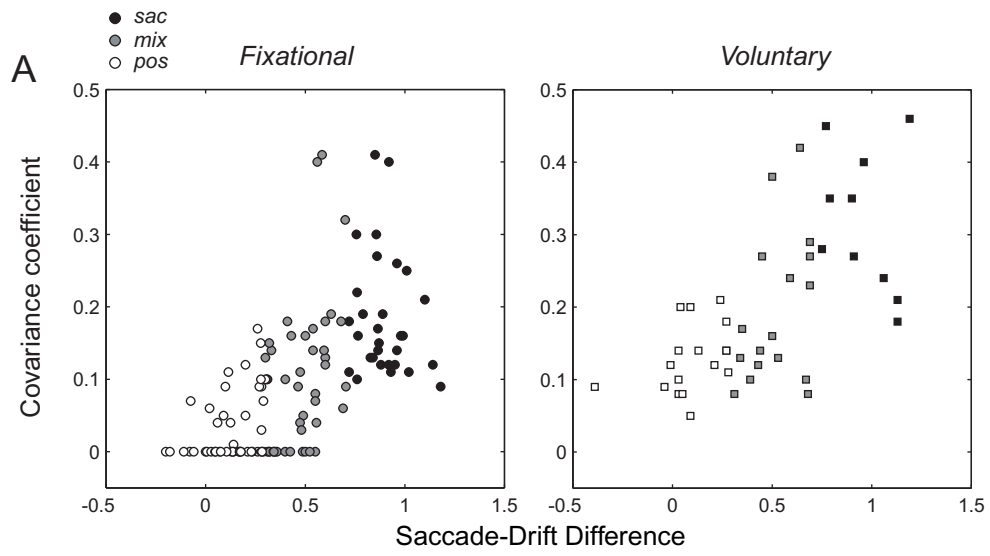
Figure S12. Voluntary saccade metrics collected with a 200 Hz eye position sampling rate (monkey M46). This figure shows the distribution of saccade directions (row 1, column 1 and 2; chosen to be orthogonal to the optimally oriented bar for each neuron), the main sequence (row 1, column 3), and the distributions of saccade amplitude, mean velocity, and duration (row 2, columns 1-3).
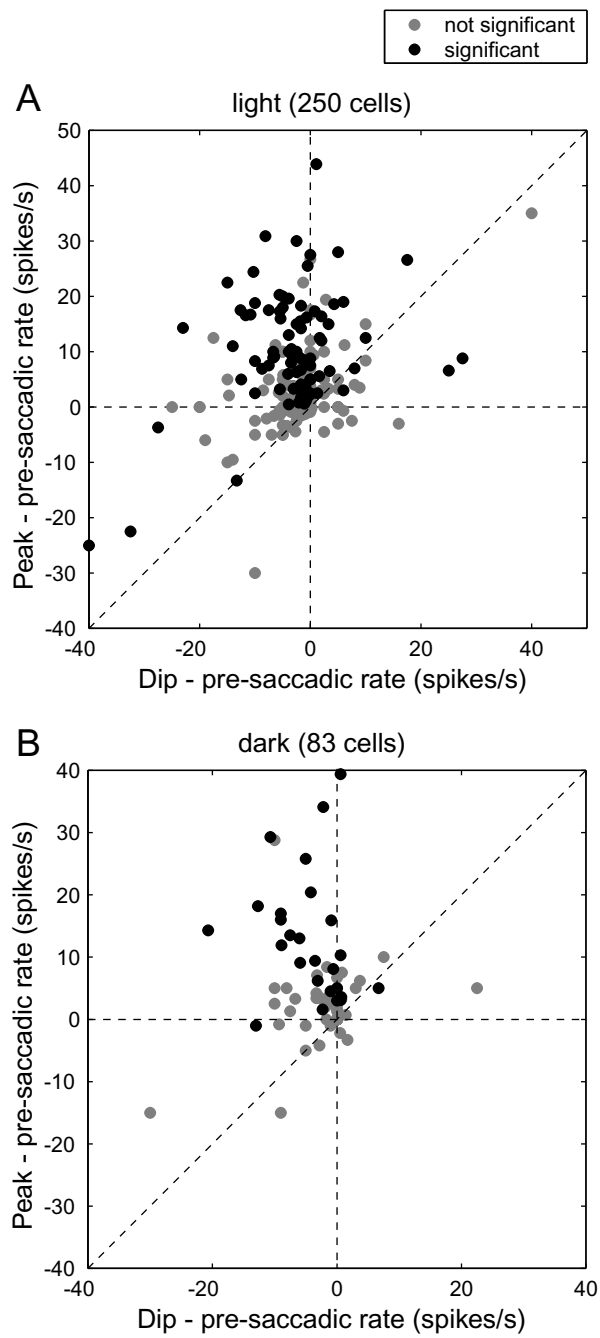
Figure S13. Saccadic main sequence (peak velocity as function of amplitude) for fixational and voluntary saccades for the 3 monkeys used in these experiments. Data from monkey M42 were collected at 100 Hz in the light without any stimulus present and used for the analysis of extraretinal effects (also used in Figure 8). Data from monkeys M45 and M46 are the same as in Figures S7 and S12. Note that fixational and voluntary saccades lie on a similar slope, and that amplitudes of larger fixational and smaller voluntary saccades overlap. See the legend to Figure S7 for explanation of sampling quantization effects manifested as dense lines of points with a fixed relationship between the amplitude and the velocity.
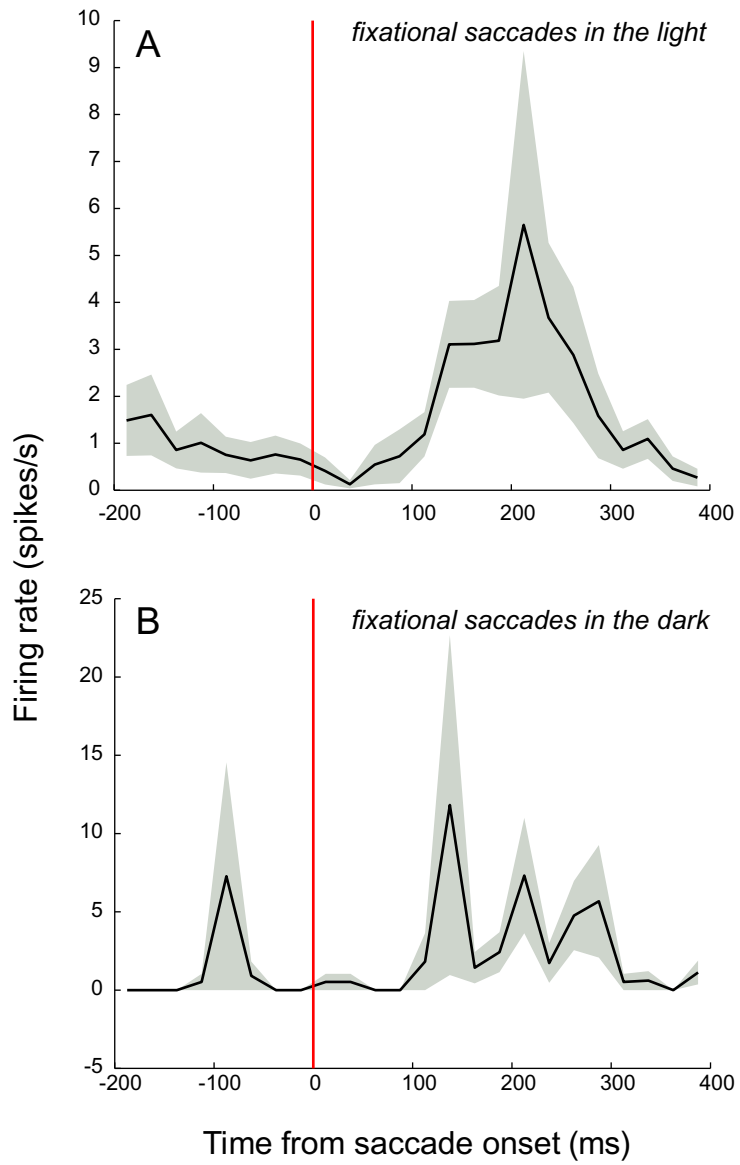
Figure S14. Fixational drift metrics in 2 monkeys acquired at 100 Hz eye position sampling rate (M45, top two rows) and 200 Hz sampling rate (M46, bottom two rows). The intersaccadic drift periods were recorded from the onset of a saccade until the next saccade. The 250 ms delay was imposed to ensure that all neuronal activity analyzed during drift periods was not attributed to modulations accompanying the preceding saccade. **Direction:** Overall drift direction was measured as the difference between eye position at the start- and end-points of the drift period (column 1, rows 1 and 3). Note the upward drifts in M46 (column 1, row 3), which were compensated by infrequent, predominantly downward, fixational saccades whose distribution was shown in Figure 8 and in Figure S7. **Duration:** Since we used 250 ms delay after each saccade for collecting neuronal data during drifts, and used exactly the same data epochs for calculating drift metrics, the distributions and the numbers shown in column 2, rows 1 and 3 should be increased by 250 ms to represent the full intersaccadic drift interval. **Position:** Eye position during most drift periods was relatively stable, with s.d. <5' (column 3, rows 1 and 3). **Amplitude:** Amplitude was estimated in three ways (column 1, rows 2 and 4): 1) *Overall amplitude*, the 2D displacement from start- to end-point of drift period. 2) *Max from onset*, the 2D displacement from start- to the farthest point of the trajectory (this was done to account for potential looping or oscillating curved trajectories). 3) *Max per sample*, the maximal 2D displacement from one sample to the next (5 or 10 ms). The similarity between "overall" and "max from onset" distributions shows that most drifts were directionally monotonic. **Velocity:** We report the following measures of velocity during drift periods (columns 2 and 3, rows 2 and 4): 1) Mean velocity from one sample to the next (*i.e.* per sample). 2) Peak velocity per sample. To a large degree these velocities are caused by the noise in the experimental system (2-3' for Purkinje eyetracker, 1-2' for
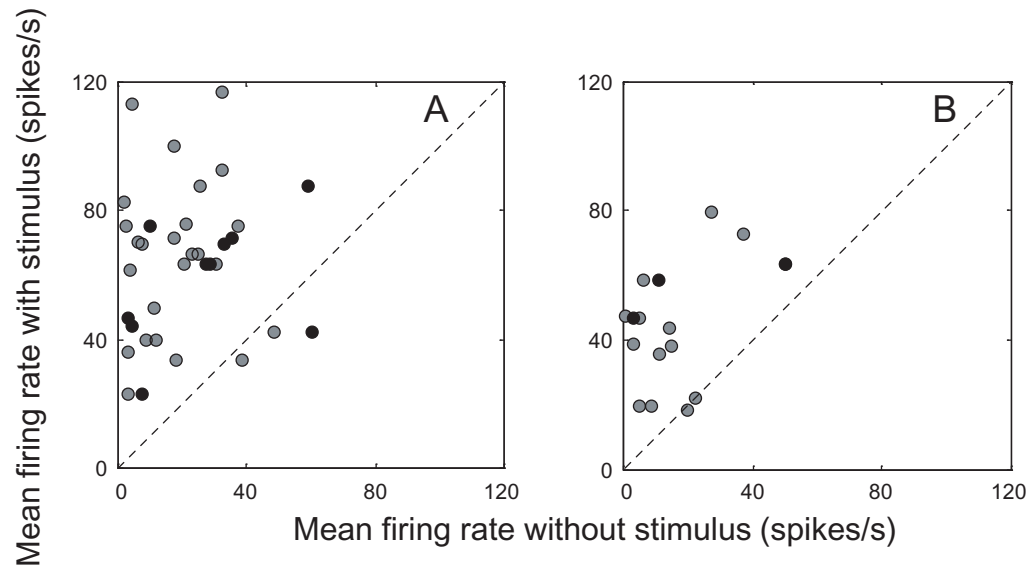
eye coil). For example, a displacement of 1' will have 1.7°/s speed, 2' – 3.3°/s, 3' - 5°/s at 100 Hz. Importantly, this noise is random and uniformly fluctuates around the mean signal, therefore it does not cause any significant change in the position (see Figures S10, S11). These calculations are shown primarily to illustrate that the criterion used to detect saccades (*i.e.* velocity <10°/s) is outside the velocities calculated in the same way during the drift periods. We used two additional approaches to get a more realistic estimate of drift velocities. 3) *Mean directional velocity* was calculated separately for horizontal and vertical eye position, from start- to the farthest point of the trajectory. This procedure averages random positive and negative per-sample fluctuations of the position signal, but retains any consistent directional component of the drift. Again, averaging until the farthest point of the trajectory was chosen to ensure that the velocity was not underestimated in looping trajectories. Most of the directional velocity estimates fell below 1°/s (largely overlapping green and magenta curves, bin size 0.25°/s). 4) *Mean and peak smoothed drift velocity* was calculated by linearly interpolating eye position records to 1 KHz and then smoothing them with a Gaussian filter of σ=10 ms (see Figure S11). This procedure eliminated most of the instrumental noise from the records but kept the position fluctuations intact.

Supplementary Figure S1
Kagan, Gur and Snodderly

Eye movements and neuronal firing in macaque V1

Supplementary Figure S2
Kagan, Gur and Snodderly

Eye movements and neuronal firing in macaque V1

Supplementary Figure S3
Kagan, Gur and Snodderly   Eye movements and neuronal firing in macaque V1

Firing rate (spikes/s)

A *fixational saccades in the light*

B *fixational saccades in the dark*

Time from saccade onset (ms)

Supplementary Figure S4
Kagan, Gur and Snodderly   Eye movements and neuronal firing in macaque V1

A

(1) saccade amplitude 12.1 minarc, duration 10 ms, 100 Hz eye position sampling rate - 100% detection

Eye position (minarc)

Velocity (deg/s)

-*shift*

+*shift*

• sampled at 100 Hz
— "actual" (1000 Hz)

(2) saccade amplitude 6.1 minarc, duration 5 ms, 100 Hz eye position sampling rate - 60% detection

Eye position (minarc)

Velocity (deg/s)

(3) saccade amplitude 8.1 minarc, duration 10 ms, 100 Hz eye position sampling rate - 50% detection

Eye position (minarc)

Velocity (deg/s)

Saccade onset relative to sampling (ms)

B

data from panel A(1)

Onset estimation shift (ms)

+*shift*

-*shift*

Saccade onset relative to sampling (ms)

C

% detected saccades

200 Hz (mean)
5 ms
7.5 ms
10 ms
12.5 ms
15 ms

100 Hz (mean)
5 ms
7.5 ms
10 ms
12.5 ms
15 ms

Saccade amplitude (minarc)

Supplementary Figure S6
Kagan, Gur and Snodderly

Eye movements and neuronal firing in macaque V1

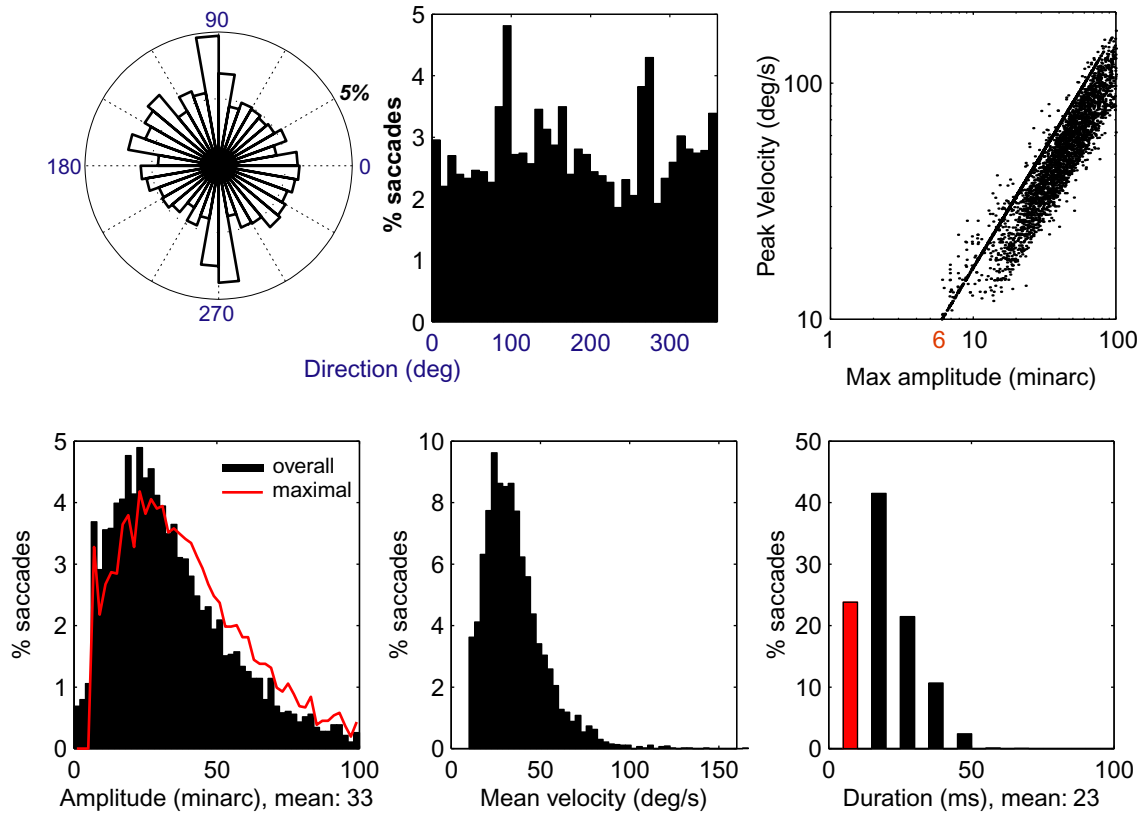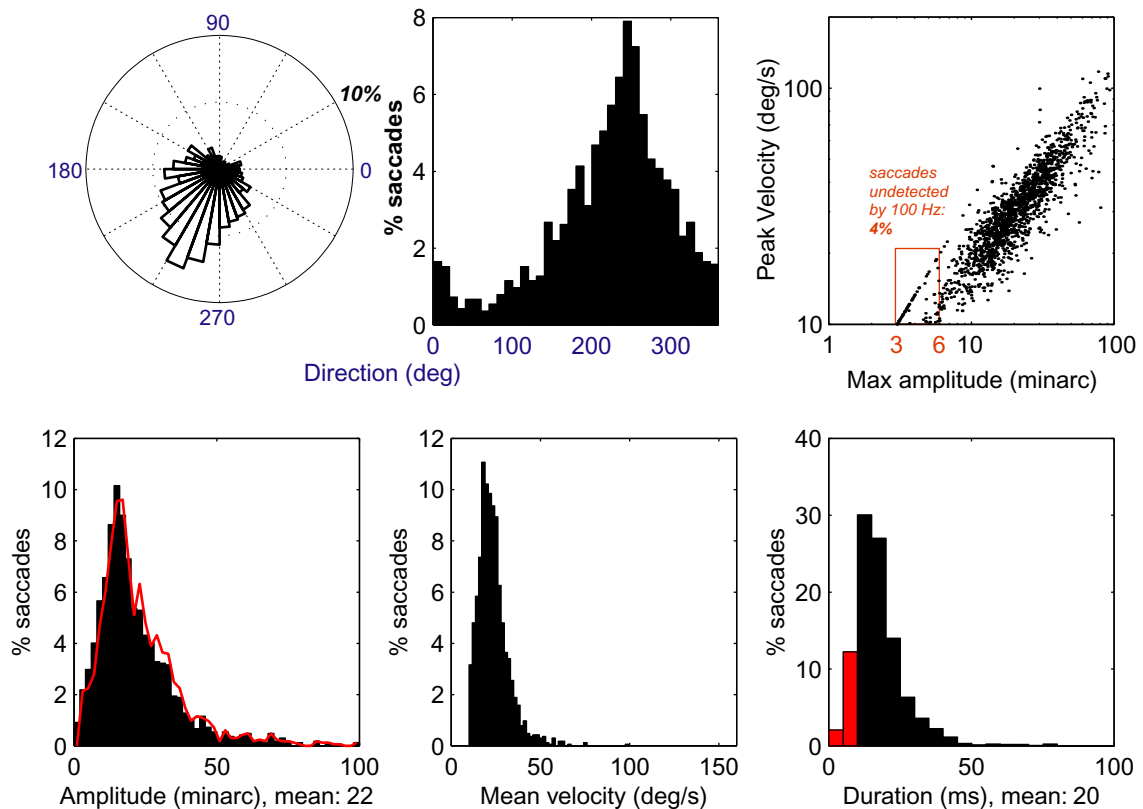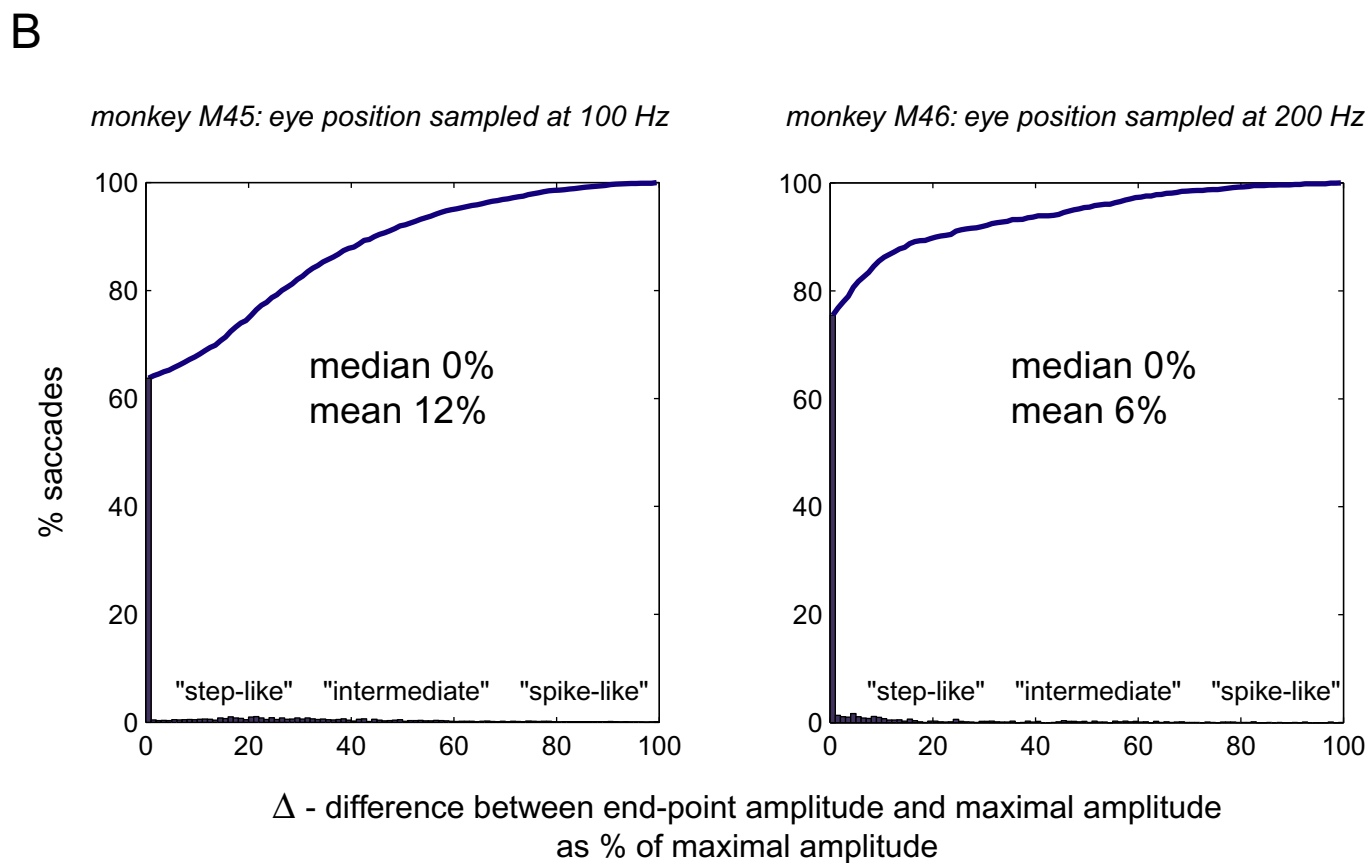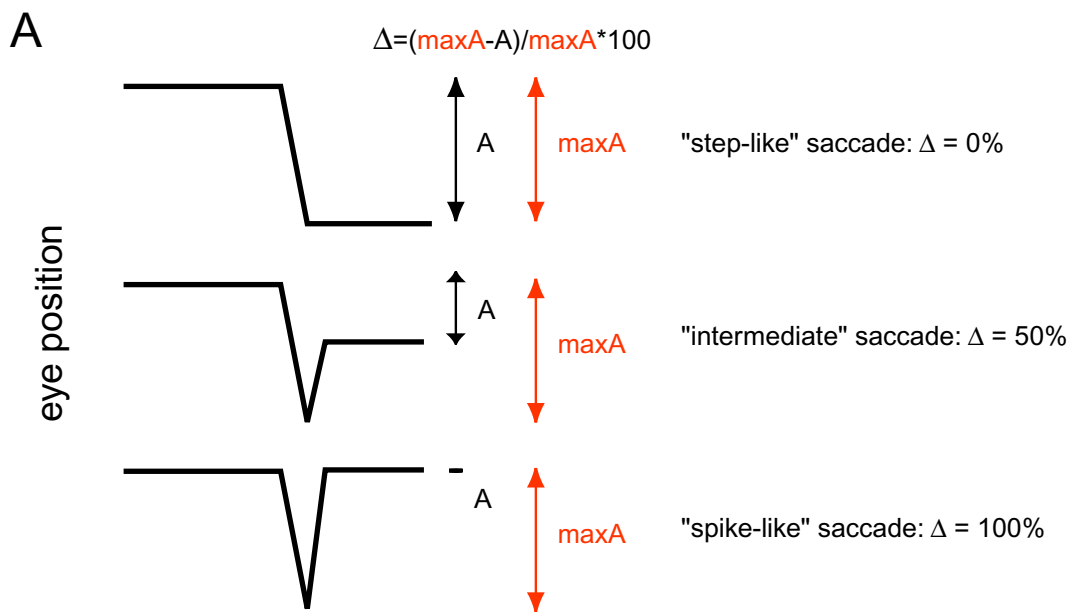monkey M45: eye position sampled at **100 Hz**, 53 neurons - FIXATIONAL SACCADES

monkey M46: eye position sampled at **200 Hz**, 65 neurons - FIXATIONAL SACCADES

Supplementary Figure S7
Kagan, Gur and Snodderly

Eye movements and neuronal firing in macaque V1

A

$\Delta=(maxA-A)/maxA*100$

A | maxA | "step-like" saccade: $\Delta = 0\%$

A | maxA | "intermediate" saccade: $\Delta = 50\%$

A | maxA | "spike-like" saccade: $\Delta = 100\%$

eye position

B

monkey M45: eye position sampled at 100 Hz

monkey M46: eye position sampled at 200 Hz

% saccades

median 0%
mean 12%

"step-like"   "intermediate"   "spike-like"

median 0%
mean 6%

"step-like"   "intermediate"   "spike-like"

Δ - difference between end-point amplitude and maximal amplitude
as % of maximal amplitude

Supplementary Figure S8
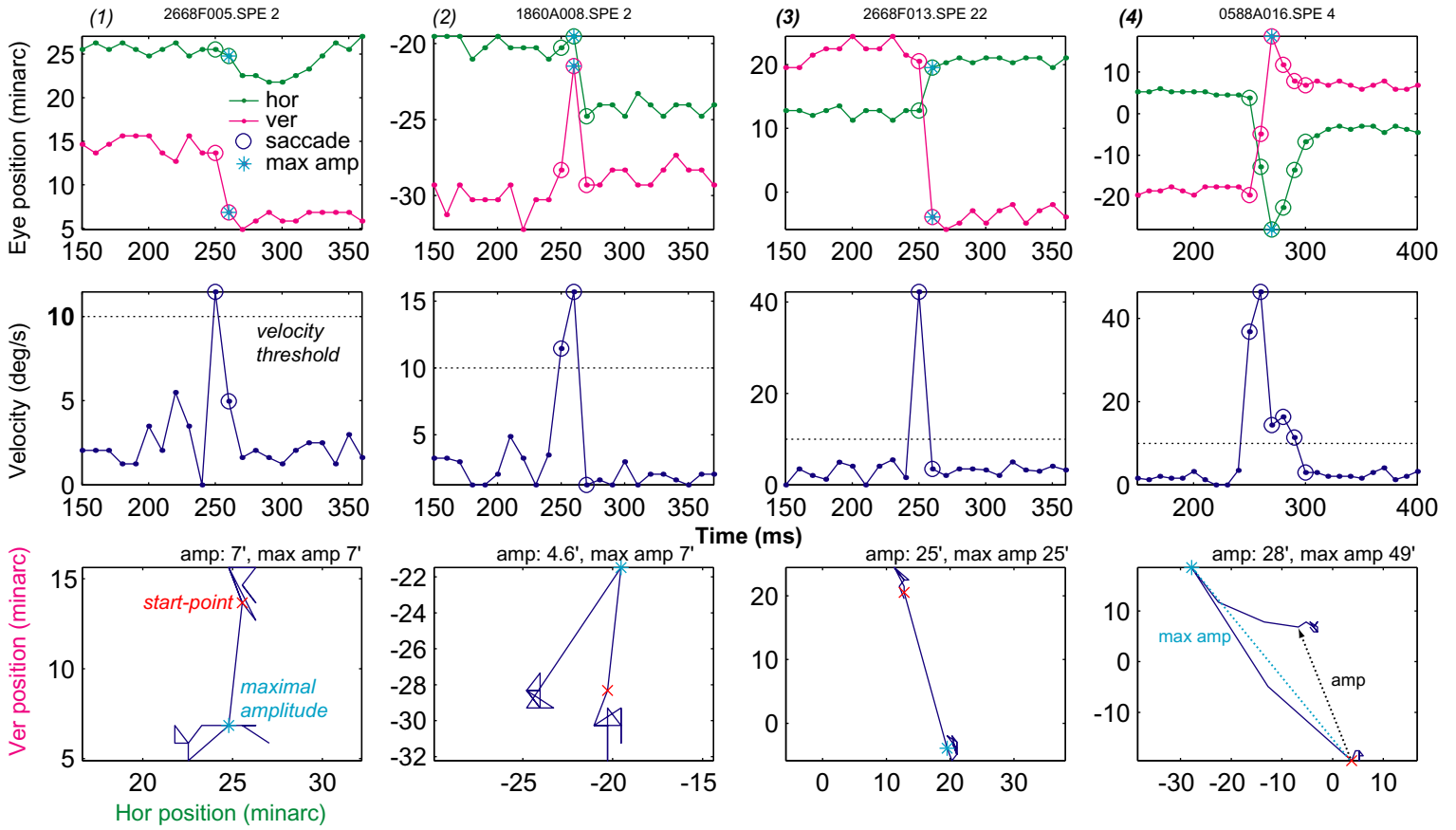*Kagan, Gur and Snodderly*   Eye movements and neuronal firing in macaque V1

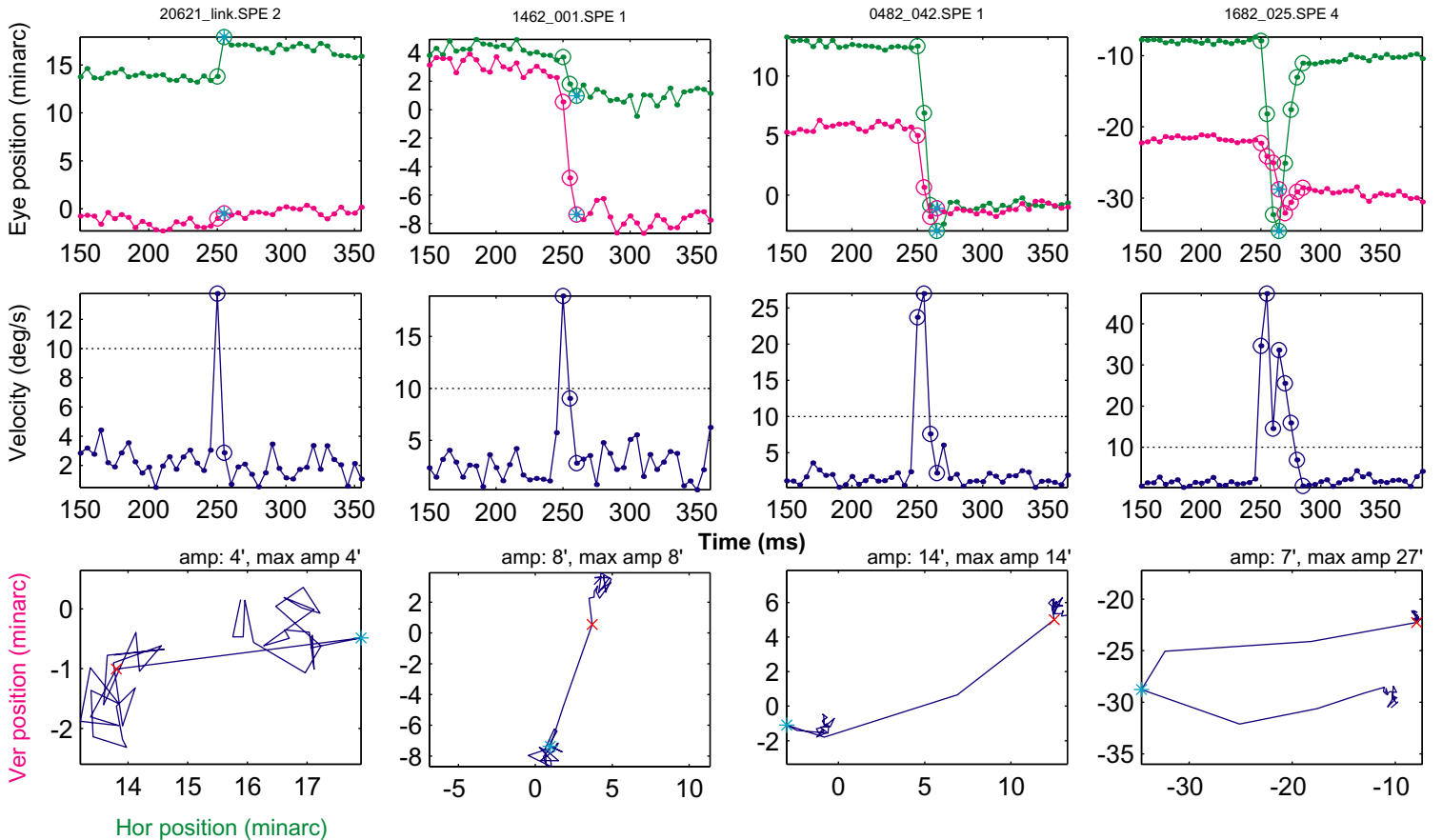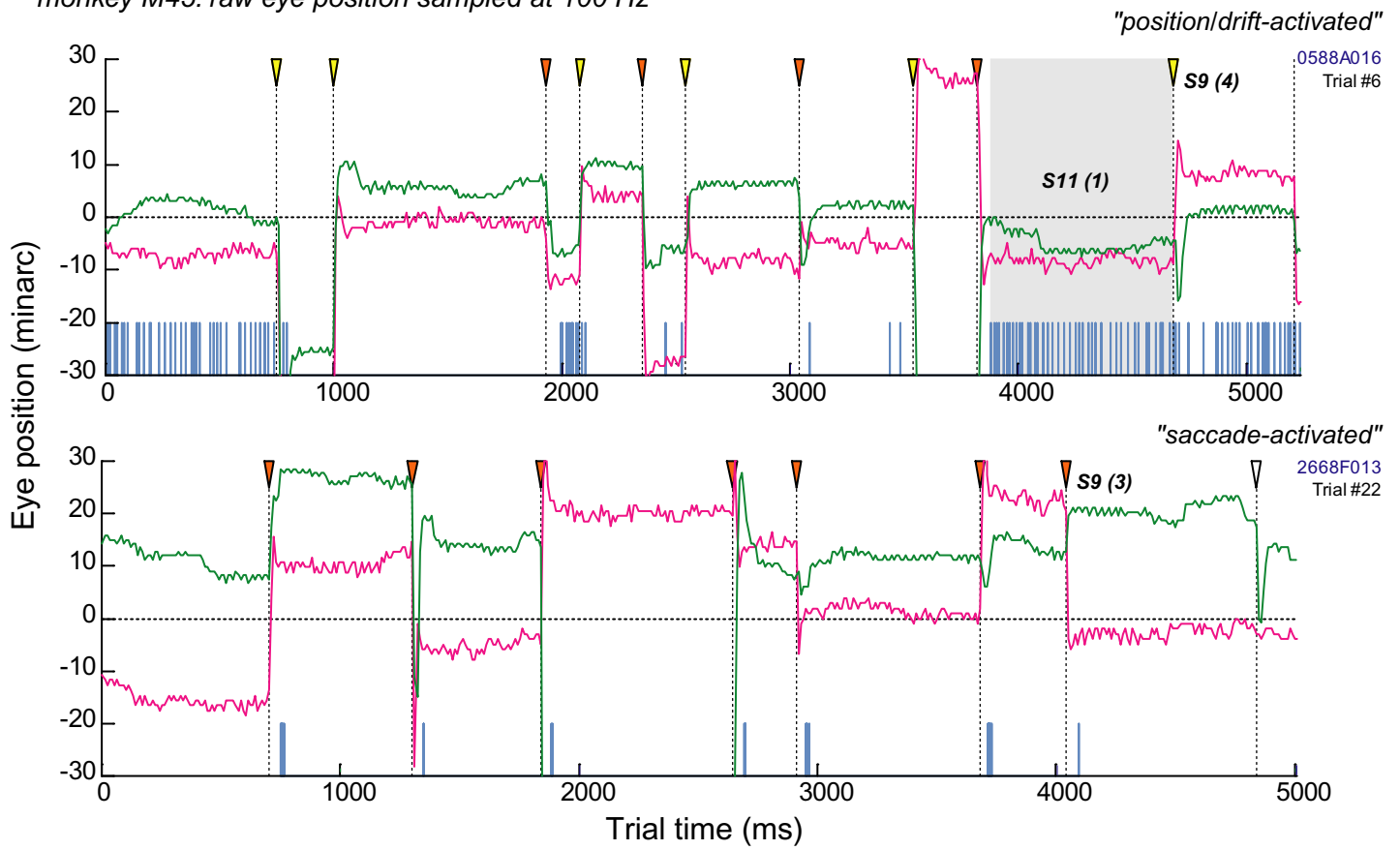*monkey M45: raw eye position sampled at 100 Hz - FIXATIONAL SACCADES*

monkey M46: raw eye position sampled at 200 Hz - FIXATIONAL SACCADES

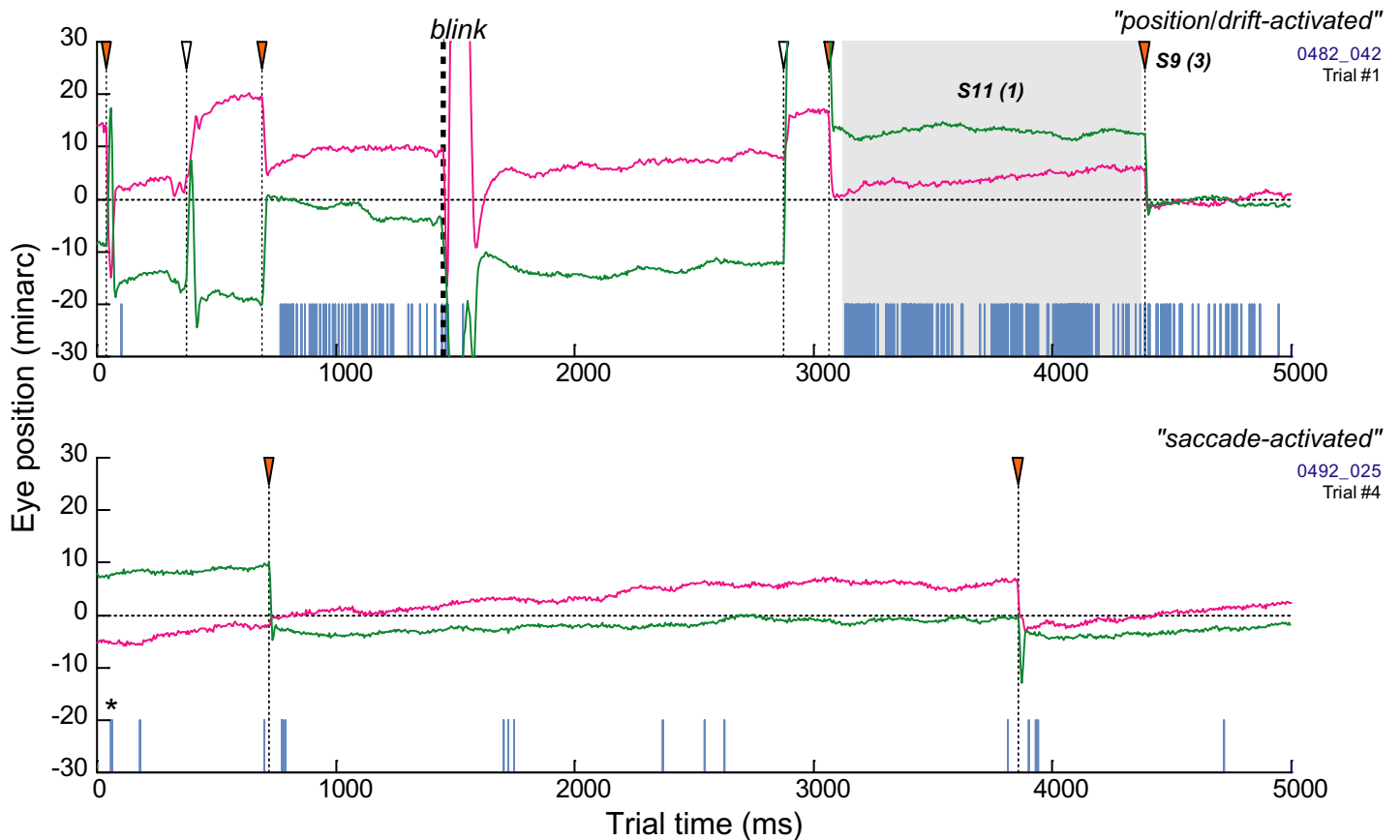**Supplementary Figure S9**
Kagan, Gur and Snodderly

Eye movements and neuronal firing in macaque V1

monkey M45: raw eye position sampled at 100 Hz

"position/drift-activated"

0588A016
Trial #6

S9 (4)

S11 (1)

"saccade-activated"

2668F013
Trial #22

S9 (3)

Eye position (minarc)

Trial time (ms)

monkey M46: raw eye position sampled at 200 Hz

"position/drift-activated"

blink

0482_042
Trial #1

S9 (3)

S11 (1)

"saccade-activated"

0492_025
Trial #4

Eye position (minarc)

Trial time (ms)

Supplementary Figure S10
Kagan, Gur and Snodderly

Eye movements and neuronal firing in macaque V1

monkey M45: raw eye position sampled at 100 Hz - FIXATIONAL DRIFTS

monkey M46: raw eye position sampled at 200 Hz - FIXATIONAL DRIFTS

Supplementary Figure S11
Kagan, Gur and Snodderly

Eye movements and neuronal firing in macaque V1

monkey M46: eye position sampled at 200 Hz, 44 neurons - VOLUNTARY SACCADES
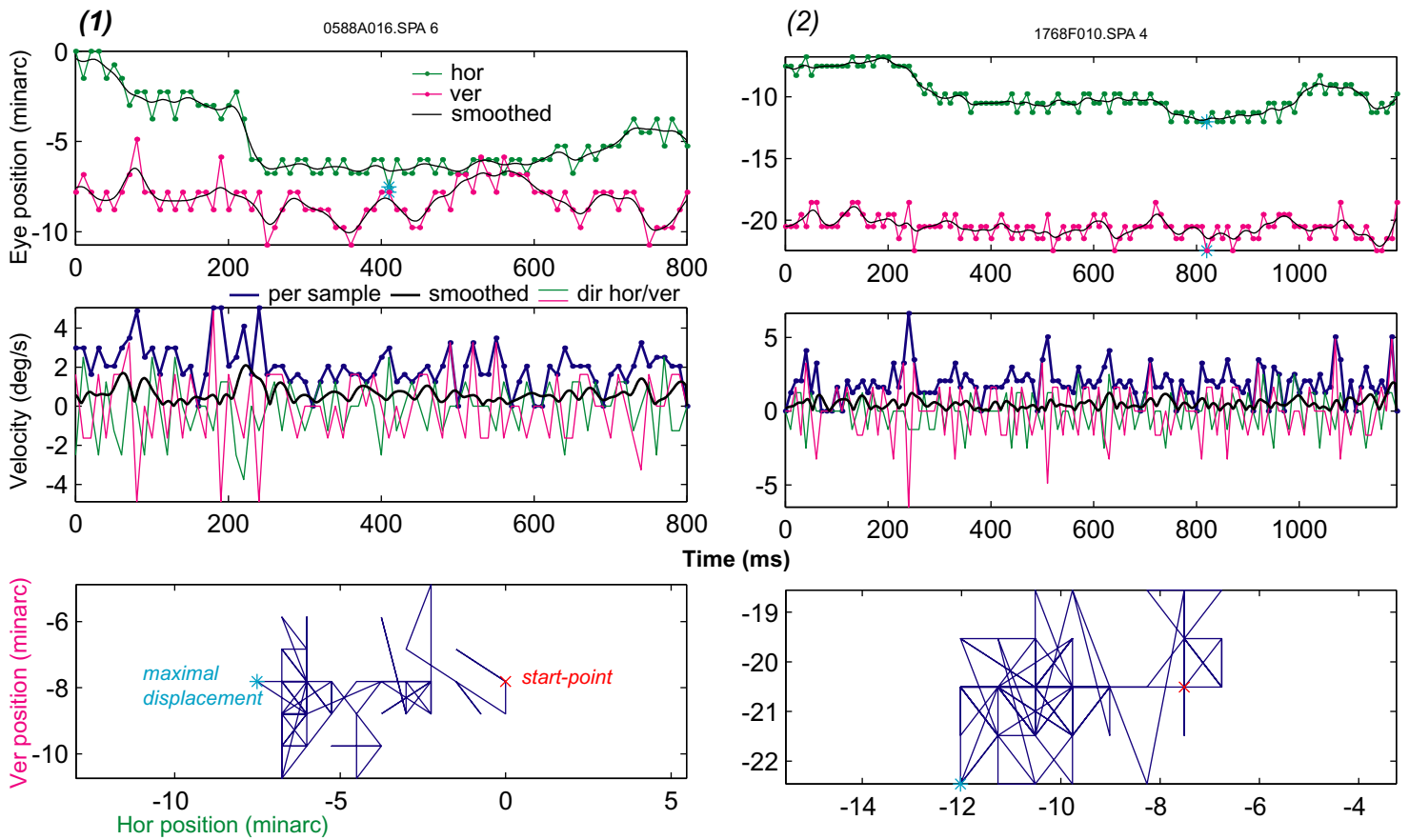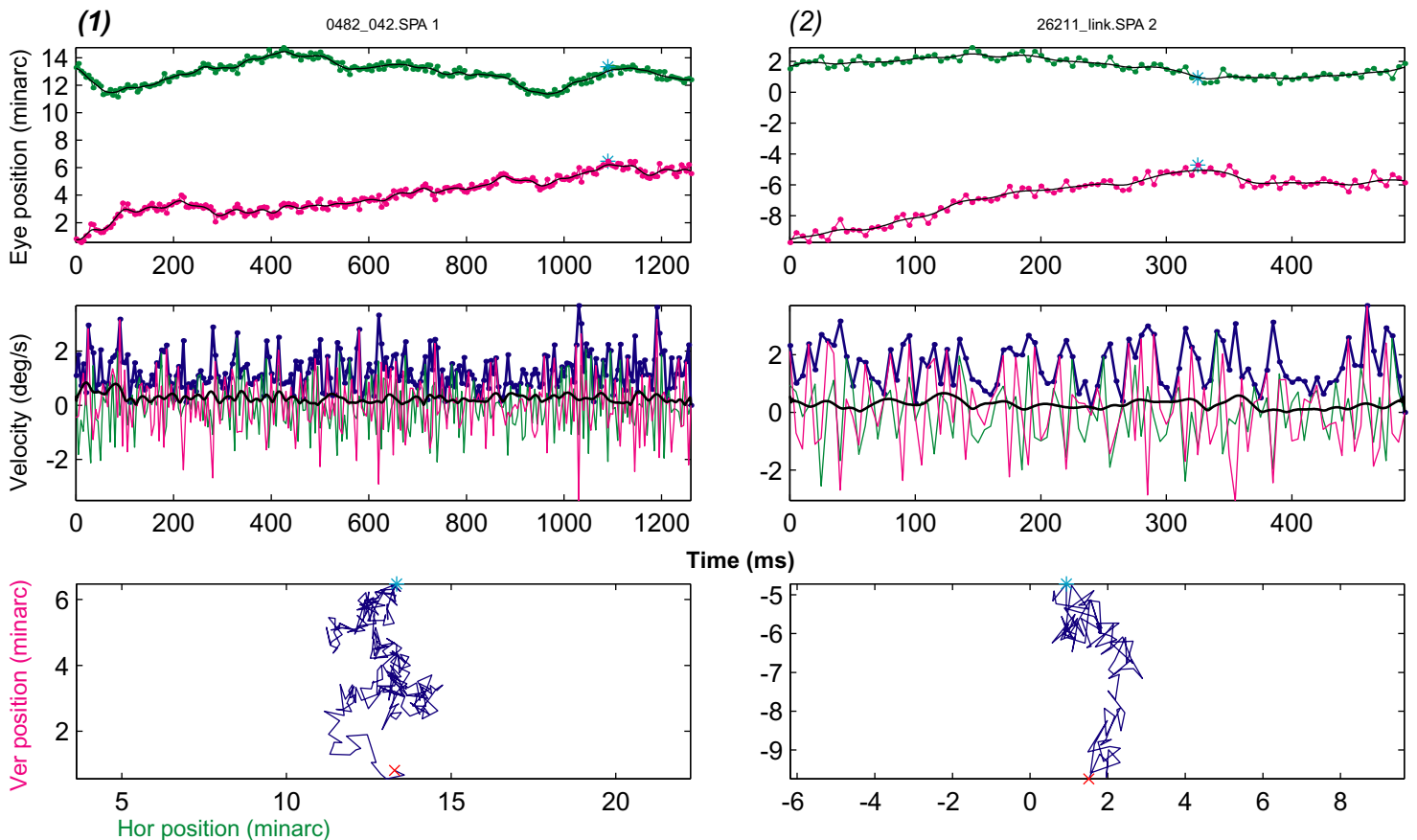
Supplementary Figure S13
Kagan, Gur and Snodderly   Eye movements and neuronal firing in macaque V1

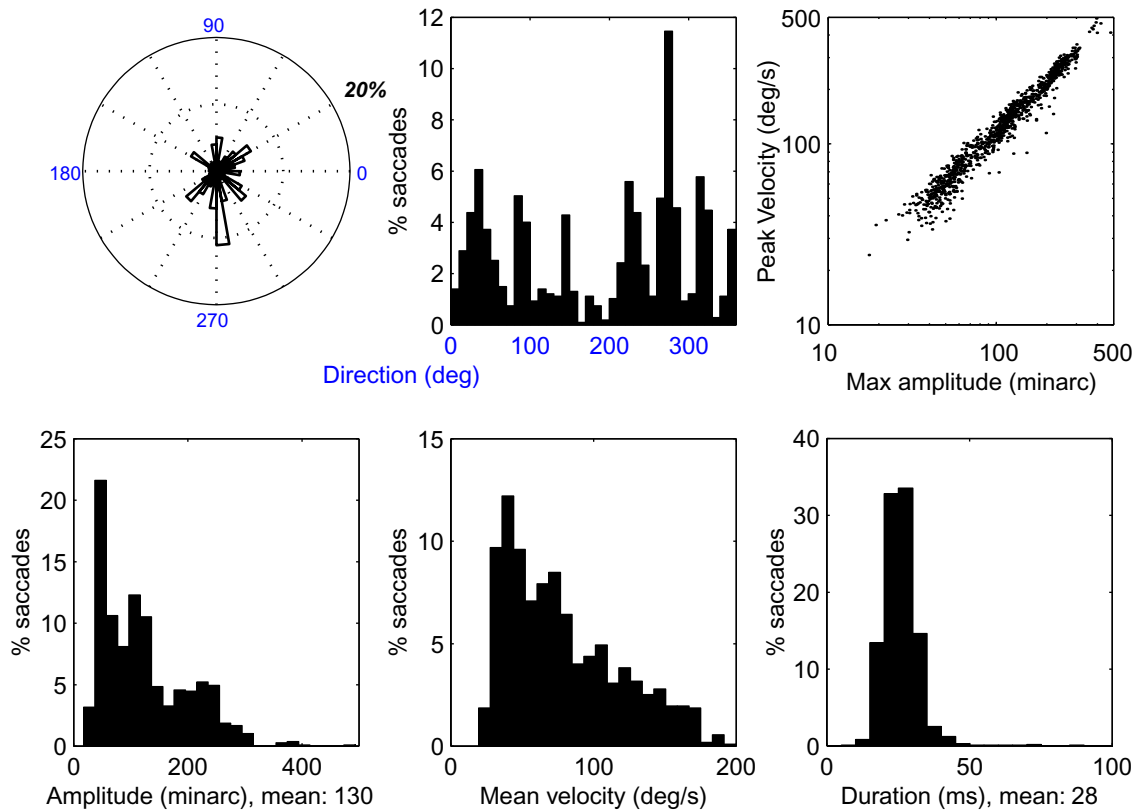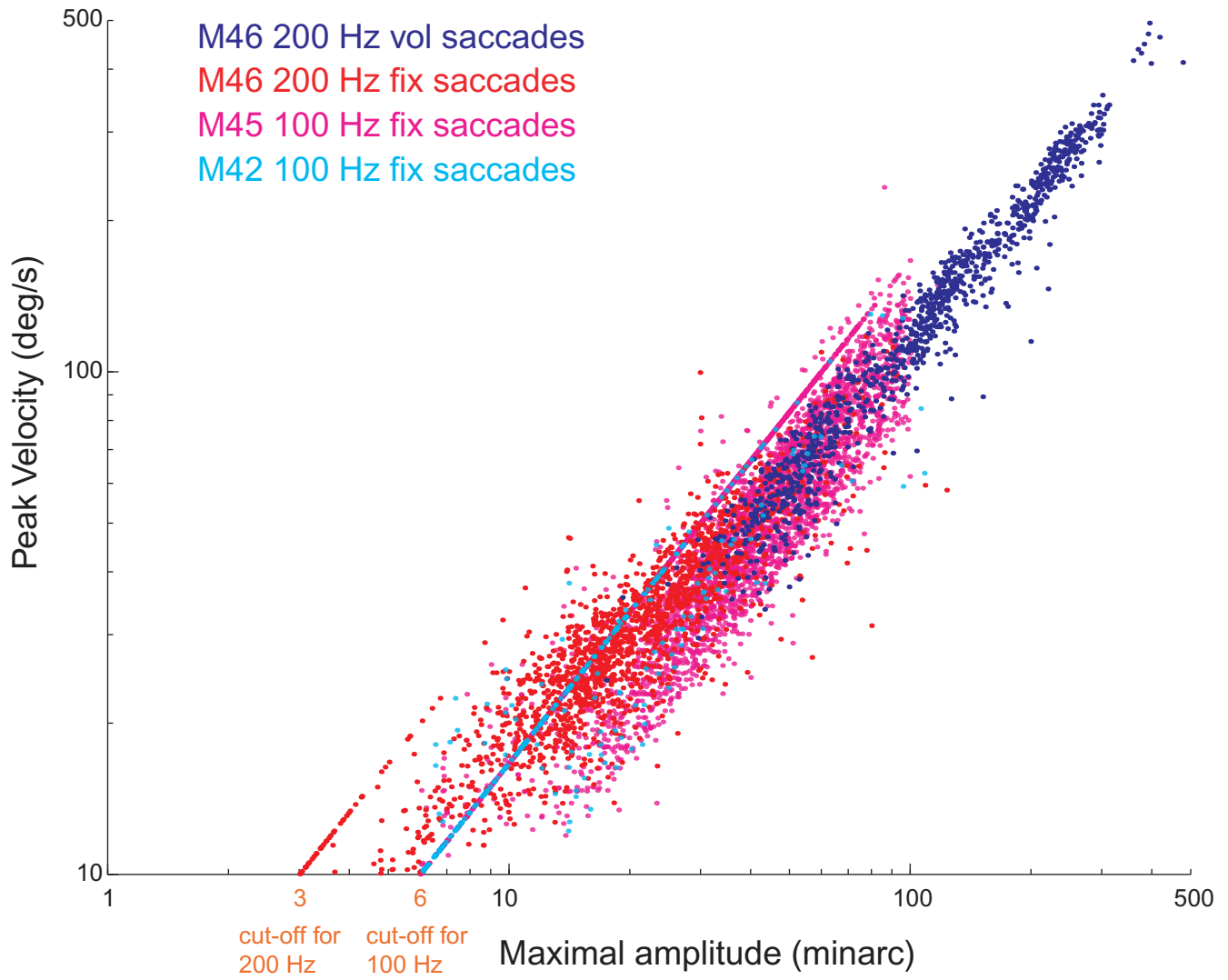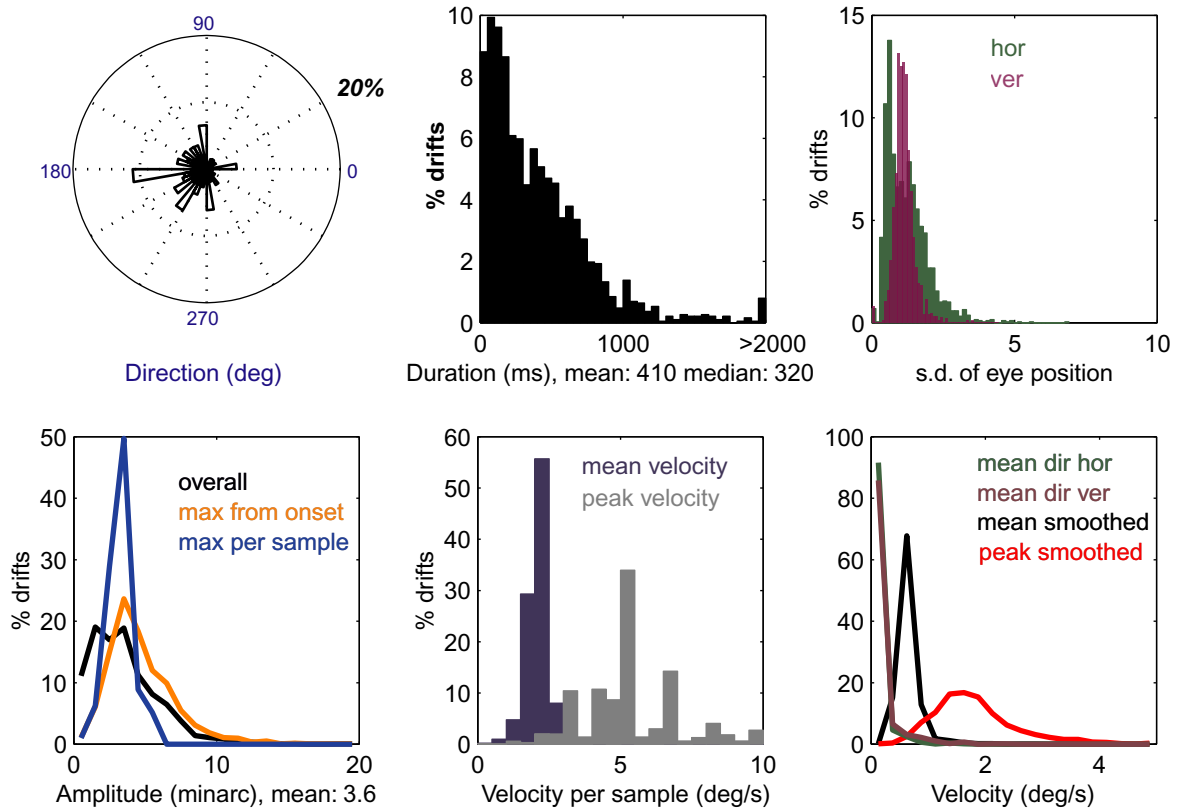*monkey M45: eye position sampled at 100 Hz, 53 neurons - DRIFTS*

Top-left polar plot:
90
180
270
0
20%
Direction (deg)

Top-middle histogram:
10
8
6
4
2
0
% drifts
0   1000   >2000
Duration (ms), mean: 410 median: 320

Top-right histogram:
15
10
5
hor
ver
% drifts
0   5   10
s.d. of eye position

Middle-left plot:
50
40
30
20
10
% drifts
overall
max from onset
max per sample
0   10   20
Amplitude (minarc), mean: 3.6

Middle-middle histogram:
60
50
40
30
20
10
0
% drifts
mean velocity
peak velocity
0   5   10
Velocity per sample (deg/s)

Middle-right plot:
100
80
60
40
20
0
mean dir hor
mean dir ver
mean smoothed
peak smoothed
0   2   4
Velocity (deg/s)

monkey M46: eye position sampled at 200 Hz, 65 neurons - DRIFTS

Polar plot:
90
180
270
0
20%
Direction (deg)

Histogram:
15
10
5
0
% drifts
0   1000   >2000
Duration (ms), mean: 1100 median: 870

Histogram:
10
8
6
4
2
0
hor
ver
% drifts
0   5   10
s.d. of eye position

Plot:
60
50
40
30
20
10
% drifts
overall
max from onset
max per sample
0   10   20
Amplitude (minarc), mean: 6.2

Histogram:
60
50
40
30
20
10
0
% drifts
mean velocity
peak velocity
0   5   10
Velocity per sample (deg/s)

Plot:
100
80
60
40
20
0
mean dir hor
mean dir ver
mean smoothed
peak smoothed
0   2   4
Velocity (deg/s)

Supplementary Figure S14
Kagan, Gur and Snodderly

Eye movements and neuronal firing in macaque V1