Title:

Eye-position error influence over "open-loop" smooth pursuit initiation

Abbreviated title:

Transient disruption of smooth pursuit initiation

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Abstract

The oculomotor system integrates a variety of visual signals into appropriate motor plans, but such integration can have widely varying time scales. For example, unlike with rapid saccades, it has been suggested that initiating a smooth pursuit eye movement to follow a moving target involves an obligatory open-loop interval, in which new visual motion signals presumably cannot influence the ensuing motor plan for up to 100 ms after movement initiation. However, this view runs directly contrary to the idea that the oculomotor periphery has privileged access to short-latency visual signals. Here we show that smooth pursuit initiation is highly sensitive to visual inputs, even in open-loop intervals. We instructed male rhesus macaque monkeys to initiate saccade-free smooth pursuit eye movements, and we injected a transient, instantaneous eye position error signal at different times relative to movement initiation. We found highly robust short-latency modulations in eye velocity and acceleration, starting only ~50 ms after transient signal occurrence, and even during open-loop pursuit initiation. Critically, the spatial direction of the injected position error signal had predictable effects on smooth pursuit initiation, with forward errors increasing eye acceleration and backwards errors reducing it. These effects persisted during sustained pursuit, and also affected catch-up saccades in a similar manner. Importantly, catch-up saccade kinematics were altered ~50 ms after transient signal occurrence, much like microsaccade kinematic alterations during simple fixation. Our results demonstrate that smooth pursuit initiation is highly sensitive to visual signals, and that catch-up saccades behave like regular microsaccades and saccades.

Introduction

A highly intriguing property of motor circuits is that they have privileged access to sensory signals. For example, neck muscles and related upper body motor synergies, which are normally recruited to re-orient the upper body towards behaviorally-relevant stimuli, exhibit very early stimulus-locked responses after visual flashes (Corneil, 2004; Corneil et al., 2013; Corneil and Munoz, 2014). These stimulus-locked responses, arriving <100 ms after stimulus onset, occur almost as early as neural responses in primary visual areas (Corneil, 2004; Boehnke and Munoz, 2008; Corneil and Munoz, 2014). Similarly, oculomotor control circuitry in the brainstem are riddled with eye-movement related neurons exhibiting both visual and eye-movement related discharge in the very same neurons. In fact, at least in the superior colliculus (SC), it is only such visual-motor neurons in particular that exhibit the most perceptual and behavioral relevance to task success in some cases (Wurtz and Mohler, 1976; Chen and Hafed, 2017). This concomitance of visual and motor signals in the very same neurons occurs even in pre-motor nuclei (Evinger and Kaneko, 1982; Everling et al., 1998; Missal and Keller, 2002), a mere single synapse away from the motor neurons innervating the ocular muscles.

Intuitively, access to the sensory periphery by motor systems makes sense. For example, in the case of vision and the oculomotor system, such access can allow very rapid resetting of oculomotor plans in order to make appropriate orienting reflexes (Reingold and Stampe, 1999, 2002; Buonocore and McIntosh, 2008; Rolfs et al., 2008; Hafed and Ignashchenkova, 2013). Such resetting becomes evident in eye movement behavior after a stimulus flash, and it can also have large repercussions on eye movement kinematics (Edelman and Xu, 2009; Guillaume, 2012; Buonocore et al., 2016; Buonocore et al., 2017), even before most higher-level cortical visual areas finalize processing of the appearing stimuli (Schmolesky et al., 1998). That is,

when one carefully analyzes the properties of eye movements a mere ~50-100 ms after stimulus onset, one discovers that a full processing cycle from visual input all the way to eye movement output has taken place (Fischer and Boch, 1983; Boch et al., 1984; Fischer and Ramsperger, 1984; Tian et al., 2018). This rapid access to visual inputs imparts on the oculomotor system a highly efficient pathway for behavioral re-orienting.

However, even within a single motor output modality, different classes of behaviors can have very different time scales. For example, saccadic eye movements occur on a much more rapid time scale when compared to much slower smooth pursuit eye movements used to track a moving target. Even though recent research has shown that saccades and smooth pursuit eye movements do share neural resources (Krauzlis, 2004), the different time scales between these two types of eye movements have led to descriptions of categorical differences between them. One such difference concerns the initiation of the two types of eye movements. While saccade initiation may be independent of visual stimulation, smooth pursuit eye movements require motion signals (Rashbass, 1961). More importantly, smooth pursuit eye movements are overwhelmingly described as possessing an “open-loop” interval of ~100 ms, before which new visual motion signals cannot influence the motor behavior (Lisberger and Westbrook, 1985). This directly contradicts the idea that the oculomotor periphery, which ultimately influences both saccades and smooth pursuit, has privileged access to visual signals.

Here we found that smooth pursuit initiation is indeed sensitive, and with short latencies, to visual stimuli within the same time window as saccades. We introduced instantaneous visually-defined eye position errors to the smooth pursuit system during the initiation of tracking eye movements. We found a highly systematic directional influence of such signals on smooth pursuit initiation, and with latencies of

only 50-80 ms. These results, directly paralleling saccadic ones (Reingold and Stampe, 2002; Buonocore and McIntosh, 2008; Buonocore et al., 2017), suggest that the concept of “open-loop pursuit” may be too coarse to accurately describe the mechanisms of smooth pursuit initiation.

Materials and Methods

Animal preparation and laboratory setup

We recorded eye movements with high spatial and temporal precision from two male rhesus macaque monkeys (monkey M and monkey A) aged 7 years. The monkeys were implanted, under general anesthesia and using sterile surgical techniques, with a titanium head-post attached to the skull with titanium skull screws (all implanted under the skin and muscle surrounding the skull). The head-post allowed stabilizing the animals' head position during the experiments. In a subsequent surgery, one eye in each animal was implanted with a scleral search coil to allow tracking eye movements using the magnetic induction technique (Fuchs and Robinson, 1966; Judge et al., 1980). The animals are part of a larger neurophysiology experiment for which the behavioral training needed for the current study was a necessary prerequisite. The experiments were approved by ethics committees at the regional governmental offices of the city of Tuebingen, and these experiments were in accordance with European Union guidelines on animal research, as well as the associated implementations of these guidelines in German law.

During the experiments, the animals were seated in a primate chair 74 cm from a CRT computer monitor in an otherwise dark room. The monitor had a pixel resolution of 34 pixels/deg and a refresh rate of 120 Hz, and stimuli were presented over a uniform gray background (29.7 Cd/m^2). A small white spot (5 x 5 min arc square) having 86 Cd/m^2 luminance was used as either a fixation spot or the moving target for smooth pursuit eye movements (see *Behavioral tasks* below). Other stimuli that we used consisted of white flashes of different sizes and positions, as described in *Behavioral tasks* below; they all had the same luminance as the spot (86 Cd/m^2). The CRT monitor viewed by the monkeys was controlled by a computer running Matlab's Psychophysics Toolbox (Brainard, 1997; Pelli, 1997; Kleiner et al., 2007).

This computer was in turn receiving commands from a real-time I/O system from National Instruments (Austin, USA), which was maintaining control of the display on a frame-by-frame basis. The system was programmed in-house and described recently (Chen and Hafed, 2013; Tian et al., 2016).

Behavioral tasks

Experiment 1: Saccade-free smooth pursuit initiation with transient position error signals

We employed a paradigm eliciting smooth (i.e. saccade-free) ocular following of a moving target while we introduced a transient eye position error signal in a specific direction. Trials started with a white fixation spot presented at the center of the display over a uniform gray background (spot and background luminance were described above). After the monkey aligned gaze with the fixation spot by 700-1000 ms, the spot jumped by 4 deg either to the right or left of fixation and simultaneously started moving horizontally (at a speed of 20.4 deg/s) in the opposite direction (i.e. there was a leftward step followed by rightward motion or a rightward step followed by leftward motion). The step size was chosen such that by the time a smooth pursuit eye movement (in the target motion direction) was initiated, no initial catch-up saccade was needed (Rashbass, 1961). This allowed us to achieve saccade-free smooth pursuit initiation velocities and, therefore, to explore how instantaneous eye position error signals could modify such smooth velocity initiation.

Instantaneous eye position error signals were introduced by interleaving trials in which the target motion above was accompanied, for a single display frame (~8 ms), by a white flash of the same luminance as the pursuit target spot. The flash, appearing 50-167 ms (in steps of ~8 ms) after target motion onset, could be a full-screen flash or a square of 1 x 1 deg dimensions centered on a position either 2.1

deg in front of or 2.1 deg behind the instantaneous pursuit target position (along the motion trajectory; in monkey M, we also ran the experiment with the square appearing at 6 deg, instead of 2.1 deg, in front of or behind the instantaneous pursuit target position). Thus, with gaze being near the pursuit target, the “forward” flash was introducing an instantaneous eye position error signal in the direction of the smooth pursuit eye movement that was being initiated, and the “backward” flash was introducing an instantaneous eye position error signal in the opposite direction. The full-screen flash was expected to act more like the backward flash, since there was more stimulus energy behind the pursuit target than ahead of it given flash timing in the trials (the flashes often happened when the eye already started to follow the pursuit target, leaving more than half of the display width behind the direction of gaze at the time of the flashes).

The monkeys were rewarded for maintaining their gaze within 3 deg from the pursuit target position until trial end (occurring 500 ms from target motion onset). This virtual gaze window for monitoring monkey behavior was relatively relaxed because we found that the flash could momentarily “attract” gaze position (with both smooth velocity and saccadic effects; see Results), and also because the monkeys were highly trained on accurate gaze alignment on the pursuit target anyway. Across multiple sessions, we collected 3624 trials from monkey M and 1996 trials from monkey A in this task (with the forward and backward flashes being 2.1 deg in front of or behind the pursuit target). We also collected 3705 trials from monkey M in the variant with forward and backward flashes being 6 deg in front of or behind the pursuit target. The four experimental conditions (normal saccade-free pursuit initiation without a flash, initiation with a full-screen flash, initiation with a forward flash, or initiation with a backward flash) were randomly interleaved within a session. The monkeys typically completed ~300 trials of this task per session, with the

remainder of the session dedicated to the other tasks below, or to other neurophysiology-related tasks not described in this study.

Experiment 2: Saccadic smooth pursuit initiation with transient position error signals

We repeated the same experiment above, except that there was no initial step in target position before the target motion onset. That is, in the present experiment, the monkeys steadily fixated the fixation spot at the center of the display, and the spot then started moving at a constant speed directly from the center. Because there is an unavoidable latency in reacting to the spot's motion onset, the monkeys had to perform an initial catch-up saccade as part of initiating their smooth tracking behavior, and our purpose here was to explore the influence of instantaneous eye position error signals introduced by flashes on such an initial catch-up saccade. All other details were identical to those described above for Experiment 1. We collected 1824 trials from monkey M and 1511 trials from monkey A in this task, and the forward and backward flashes in this experiment were only at a 2.1 deg eccentricity relative to the pursuit target.

Experiments 3 and 4: Steady-state pursuit or fixation accompanied by a transient flash stimulus

To compare the effects of transient instantaneous eye position error signals on smooth pursuit initiation to effects during steady-state pursuit or during steady-state gaze fixation, we ran two additional versions of the above experiments, which we pool here because of their conceptual similarity to each other. In the first variant (Experiment 3), the flashes could appear during steady-state smooth pursuit as opposed to during smooth pursuit initiation. Trials started with the fixation spot presented at +/- 7 deg horizontally from display center, to allow for a long period of

sustained smooth pursuit spanning large portion of the horizontal extent of our display area. After 300-500 ms of fixation, the target started moving towards (and past) the center of the display at a speed of 12 deg/s. The motion lasted for ~1500-1800 ms, and at a random time during steady-state smooth pursuit (0-1500 ms from motion onset; in steps of ~8 ms), a flash could appear as in Experiments 1 and 2 above (forward and backward flashes were at 2.1 deg). We explored the effects of the flash and its type (forward, backward, or full-screen) on both smooth velocity effects as well as on catch-up saccades. We collected 5085 trials from monkey M and 2849 trials from monkey A in this experiment. In the second variant of the experiment (Experiment 4), the flashes could happen during steady-state fixation, and we explored the effects of the flashes on microsaccades (Hafed and Ignashchenkova, 2013; Buonocore et al., 2017). The flashes could happen 75-999 ms after steady fixation of a central fixation spot. We collected 5222 trials from monkey M and 3160 trials from monkey A in this experiment.

Data analysis

Detecting catch-up saccades and microsaccades

We numerically differentiated our digitized eye position signals to obtained estimates of eye velocity and eye acceleration. We used velocity and acceleration criteria to detect microsaccades and catch-up saccades, as described previously (Krauzlis and Miles, 1996a; Hafed et al., 2009; Chen and Hafed, 2013). Briefly, a radial velocity threshold identified candidate saccadic events in the eye position traces, and acceleration thresholds were then used to refine the start and endpoint of a given movement. For a subset of data, we used a machine learning approach for saccade detection (Bellet et al., 2018). All detected microsaccades and catch-up saccades were manually inspected to correct for misses or false detections.

Measuring smooth eye velocity effects

In Experiment 1, our aim was to isolate the influence of transient instantaneous eye position error signals on smooth pursuit initiation. Our task design minimized the occurrence of catch-up saccades. In the analysis, we replaced all saccades and catch-up saccades from the eye trace with Not-a-Number (NaN) labels that extended in duration to +/- 50 ms around each detected saccade. This way, we made sure that smooth pursuit averages were not influenced by high velocity changes due to the rare saccades that may have occurred. This procedure avoided catch-up saccades during initiation, and, equally important, our exclusion of microsaccades also avoided the potential influence of these fixational eye movements (occurring near target motion onset) on visual sensitivity and behavior (Hafed and Krauzlis, 2010; Hafed, 2013; Chen et al., 2015; Hafed et al., 2015; Tian et al., 2016; Chen and Hafed, 2017; Tian et al., 2018). We then averaged the radial eye velocity traces aligned on motion onset and/or flash onset for the different trial types (e.g. no flash, full-screen flash, forward flash, or backward flash). The no flash eye velocity traces acted as our baseline, to which we compared velocities when there was a flash during pursuit initiation.

To perform such comparisons, we first plotted radial eye velocity after motion onset for the no flash trials as well as for either full-screen, forward, or backward flash trials. Plotting the averages side by side (e.g. see Fig. 1A in Results) allowed us to establish that there was indeed an influence of early transient flashes on “open-loop” smooth pursuit initiation. We then plotted the difference in eye velocity between flash and no flash trials. To do this, we subtracted within each session the average radial eye velocity in no flash trials from the radial eye velocity of every flash trial of a given type (e.g. for forward flashes only); we then repeated this subtraction for all

sessions from a given monkey. This allowed us to achieve a population of velocity "difference" curves isolating the influence of a given flash type (e.g. Fig. 1B in Results); we estimated variance in this population by calculating the 95% confidence interval of the difference curves across all trials in all sessions.

To summarize the impact of a given flash type on eye velocity, we identified a measurement interval (either 50-100 ms after full flash onset or 50-80 ms after backward/forward flash onset) in the velocity difference curves described above; this interval was chosen because it was the interval during which the flash exerted the influence over eye movements that we were particularly interested in investigating (see Results). For all trials of a given type (e.g. forward flash, backward flash, or full-screen flash), we averaged the eye velocity difference in this post-flash interval, and this gave us a population of measurements across trials. We then compared populations of measurements across the different flash conditions in our trial types (e.g. forward versus backward flashes) and also for different flash times relative to smooth pursuit initiation.

To compare the influence of transient instantaneous eye position error signals on smooth pursuit initiation to their influence during steady-state smooth pursuit, we repeated the above analyses but for data containing steady-state pursuit (Experiment 3). For all flash times occurring >200 ms after target motion onset (to ensure that smooth pursuit was already ongoing) and >200 ms before trial end, we plotted radial eye velocity from -100 ms to 200 ms relative to flash onset. Importantly, we removed from the eye traces all catch-up saccades (along with flanking +/- 50 ms intervals) to avoid effects of high velocity changes on our measurements. We averaged trials from each type (e.g. full-screen flash, forward flash, or backward flash) to demonstrate the influence of the flash on smooth eye velocity.

Measuring effects on catch-up saccades and microsaccades

We analyzed catch-up saccade times, amplitudes, and peak velocities. Since the absolute majority of catch-up saccades occurred in the direction of ongoing smooth pursuit (e.g. 97% in monkey A and 98% in monkey M from the pursuit initiation trials in Experiment 2; and 90% in monkey A and 97% in monkey M in Experiment 3), we restricted our catch-up saccade analyses to saccades in the direction of pursuit. We summarized catch-up saccade frequency of occurrence, amplitude, and peak velocity with and without flashes, and as a function of forward or backward flashes. We also analyzed the influence of flash time during smooth pursuit initiation.

We sometimes referred to saccade metrics and kinematics in relation to the eye position error that was present at the time of triggering a saccade. Eye position error was defined as the instantaneous retinotopic position of the pursuit target 10 ms before catch-up saccade onset (i.e. instantaneous target position minus eye position).

To compare changes in catch-up saccade frequencies and metrics/kinematics as a function flash occurrence to changes in other types of saccades, we performed similar analyses on microsaccades during steady-state fixation (Experiment 4). In this case, microsaccades could happen in all directions, so we classified them according to flash direction (for the non-full-screen flash trials); that is, we classified microsaccade directions according to their horizontal component as being in the same direction of the flashed square or opposite it, similar to earlier work (Hafed and Ignashchenkova, 2013; Tian et al., 2016; Tian et al., 2018). Our motivation for comparing catch-up saccades to microsaccades is that microsaccades were recently shown to exhibit kinematic changes after transient visual signals similar to larger saccades (Buonocore et al., 2017); thus, microsaccades are a viable reference for

comparing catch-up saccade effects during smooth tracking to saccades without such smooth tracking (Heinen et al., 2016; Heinen et al., 2018)

To investigate saccade kinematic changes, we followed recent findings that specific post-flash saccades have a relationship between saccadic peak velocity and saccadic amplitude that deviates from the expected saccadic main sequence curve (Guillaume, 2012; Buonocore et al., 2016; Buonocore et al., 2017). To summarize this behavior in our own data, we computed the ratio between saccade peak velocity and amplitude. This value was then compared between a baseline interval before flash onset, or in some cases from no flash trials, and an interval during the appropriate post-flash period.

Experimental design and statistical analysis

Each experiment underwent within-subject analyses, and we reported summaries and statistics from each subject individually. We used two male monkeys, as stated above, to ensure replicability of results while being constrained by the 3R (Refine, Reduce, Replace) principles for animal research. Combined with congruence of several key ingredients of our results with several prior studies in monkeys (e.g. (Krauzlis and Miles, 1996b; Hafed and Ignashchenkova, 2013; Buonocore et al., 2017) and humans (e.g. (Schutz et al., 2008; Edelman and Xu, 2009; Buonocore et al., 2016; Braun et al., 2017), we have high confidence that our results from the 2 monkeys are a general property of the smooth pursuit eye movement system.

In each experiment, we randomly interleaved the relevant conditions for statistical comparisons (e.g. no flash, full-screen flash, forward flash, and backward flash in equal proportion). We also ensured a minimum of 300 repetitions per condition for the great majority of our analyses. Such a large number of repetitions made using parametric statistical comparisons in our analyses well justified. For

some analyses of catch-up saccades or microsaccades during a “saccadic inhibition” post-flash phase of trials (Reingold and Stampe, 1999, 2000, 2002; Buonocore and McIntosh, 2008; Hafed and Ignashchenkova, 2013; Buonocore et al., 2017), there was (by definition of the phenomenon) a relative rarity of saccades to analyze. However, we ensured statistical robustness even for these observations by increasing the number of repetitions per condition that we collected overall (see above), such that there was still statistical confidence in the relatively fewer inhibition-phase saccades that we were analyzing (we show 95% confidence intervals for all of our analyses).

Further statistical validation was obtained by performing, for each animal, two-way ANOVAs (factor “flash time” by factor “flash direction”, with the appropriate number of factor levels depending on the specific design within the experiment) on the summary statistics described above (e.g. amplitude of saccades and ratio of velocity over amplitude in a post-flash period). Post hoc comparisons were followed up with two-sample t-tests. Either simple or two-sample t-tests were also performed to compare different conditions when the ANOVA was not necessary; alpha levels were corrected following Bonferroni methods. We performed all of our statistical tests using standard Matlab (MathWorks, Inc.) functions.

Results

Smooth pursuit initiation is thought to involve an “open-loop” interval, of ~100 ms, during which eye velocity is presumably not yet under visual feedback error control (Lisberger and Westbrook, 1985). However, visual signals can enter into the processing hierarchy of the oculomotor system significantly earlier than 100 ms. For example, transient visual responses in the brainstem can occur with latencies shorter than ~40-50 ms (Rizzolatti et al., 1980; Boehnke and Munoz, 2008; Chen et al., 2018). We thus hypothesized that there exist mechanisms in which even “open-loop” smooth pursuit initiation can be significantly modified by visual inputs, and in a spatially-specific manner. We tested this by introducing a brief (~8 ms) transient visual stimulus onset that effectively added a transient instantaneous eye position error signal to the smooth pursuit system (Materials and Methods). We found that such a signal was highly effective in modifying smooth pursuit initiation. To demonstrate this, we first present cases in which the transient visual onset consisted of a full-screen white flash, and we then show results when spatially-precise position error signals were introduced by smaller transient flashes.

Transient visual signals perturb smooth pursuit initiation velocity even during “open-loop” intervals

Our monkeys initiated smooth pursuit eye velocity without saccades through our use of a Rashbass step-ramp target trajectory (Experiment 1; Materials and Methods) (Rashbass, 1961). After a period of steady-state fixation, the pursuit target stepped in a direction opposite to the motion trajectory and instantaneously moved at constant speed, such that by the time at which smooth pursuit was initiated, there was no need for a catch-up saccade (de Brouwer et al., 2002). At a random time during the initiation phase of smooth pursuit, we presented a full-screen flash. Figure 1A shows

398 an example of saccade-free smooth pursuit initiation with and without a full-screen
399 flash from monkey M. The gray curve shows average radial eye velocity aligned on
400 the onset of target motion across no-flash trials (i.e. control trials), and error bars
401 denote 95% confidence intervals. As expected from the task design, eye velocity
402 increased smoothly to approach target velocity, and the onset of smooth velocity
403 acceleration occurred after a latency of ~100 ms from target motion onset. However,
404 when a full-screen flash occurred 50 ms after target motion onset (blue curve in Fig.
405 1A), the initiation of smooth pursuit was transiently disrupted (indicated by the arrow
406 and label "Dip" in the figure); there was a transient reduction in eye acceleration
407 starting ~50 ms after flash onset, and almost coincident with the very earliest phase
408 of smooth pursuit initiation. This effect can be better visualized in Fig. 1B; here, we
409 subtracted, for every flash trial, the average radial eye velocity of the no-flash
410 condition from the given trial's radial eye velocity, and we then averaged all velocity
411 "difference" curves (Materials and Methods). The average velocity difference curve
412 showed a clear transient dip starting ~50 ms after full-screen flash onset (error bars
413 denote 95% confidence intervals). Importantly, we ensured that this data did not
414 contain any microsaccades or catch-up saccades (Materials and Methods). Thus,
415 early smooth pursuit initiation (so-called "open-loop" pursuit) is reliably modified by
416 transient visual signals.

417 The transient change in saccade-free pursuit initiation that we observed after
418 full-screen flashes was always time locked to flash onset. For example, Fig. 1C, D
419 shows results similar to those from Fig. 1A, B above (and from the same monkey),
420 except that the flash now happened 83 ms after target motion onset rather than after
421 50 ms. A similar transient decrease in eye acceleration during pursuit initiation was
422 evident, but it now happened slightly later in the initiation phase of smooth pursuit.
423 Similarly, Fig. 1E, F shows results with flash onset 117 ms after target motion onset,

424 and Fig. 1G, H shows results with flash onset 150 ms after target motion onset. In all
425 cases, there was a transient reduction in eye acceleration (marked by the label “Dip”
426 in each figure panel), such that the eye velocity difference curves (Figs. 1B, D, F, H)
427 always showed a negative-going transient ~50 ms after flash onset. These transients
428 are reminiscent of reductions in saccade and microsaccade frequency with short
429 latency after full-screen visual flashes (Reingold and Stampe, 1999, 2000, 2002;
430 Buonocore and McIntosh, 2008; Rolfs et al., 2008; Edelman and Xu, 2009; Bompas
431 and Sumner, 2011; Hafed and Ignashchenkova, 2013; Tian et al., 2018). However, in
432 our case, the changes to smooth pursuit initiation happened in a more “analog”
433 fashion for smooth eye velocity as opposed to a more “digital” or “all-or-none” fashion
434 for saccade frequency.

435 To demonstrate that the transient changes in smooth eye velocity were time
436 locked to full-screen flash onset in both of our monkeys, we computed the velocity
437 difference curves described above (e.g. Fig. 1B), but we did so for all flash times that
438 we tested, and also after aligning the curves to flash onset as opposed to target
439 motion onset. The left panel of Fig. 1I shows the results of this analysis for monkey
440 A, and the right panel shows the results for monkey M. Note that for presentation
441 purposes, the figure shows curves from every other flash time, but the results were
442 identical for all flash times: regardless of flash time, its influence on smooth pursuit
443 initiation appeared in each monkey ~50 ms after flash onset. To statistically verify this
444 observation, we averaged the value of the curve for each flash trial in the interval 50-
445 100 ms after flash onset and tested against zero. As expected, for both monkeys,
446 there was reliable “inhibition”, or a decrease in the difference velocity signal, across
447 flash onset times (results of the statistical tests are reported in the caption of Fig. 1).

448 The above results indicate that even open-loop smooth pursuit initiation is
449 modifiable by visual signals. We confirmed that such modification is similar to what

might happen during sustained, steady-state smooth pursuit, as was recently reported in humans (Kerzel et al., 2010). Specifically, in a subset of our experiments (Experiment 3; Materials and Methods), we presented the full-screen flash during sustained steady-state smooth pursuit. We picked an interval of smooth pursuit eye movements around the time of full-screen flash onset, and we removed any catch-up saccades present in such interval (Materials and Methods). Like described above, we also plotted radial eye velocity aligned on flash onset time (Fig. 2; error bars denote 95% confidence intervals). Full-screen flashes had a very similar effect to their effect during smooth pursuit initiation: steady-state smooth pursuit eye velocity started being transiently reduced ~50 ms after flash onset. Therefore, our results so far indicate that transient visual onsets reliably modify even open-loop smooth pursuit initiation, and in a manner similar to what happens during steady-state pursuit.

Transient instantaneous eye position error signals modify smooth pursuit initiation velocity in a direction-dependent manner

Given the results above, might it be possible that transient spatially-specific position error signals can also modify open-loop smooth pursuit initiation, and in a spatially-specific manner? To answer this question, we turned back to our saccade-free smooth pursuit initiation experiments of Fig. 1. We analyzed trials in which we presented, for only one display frame (~8 ms), a small white square of 1 deg dimension either 2.1 deg ahead of or 2.1 deg behind the smooth pursuit target. The square could appear at different times relative to target motion onset; therefore, the flash “in front” of the pursuit target could sometimes happen either “behind” or “in front of” instantaneous gaze position depending on flash timing. For example, if target motion was rightward and a “forward” flash (i.e. 2.1 deg to the right of the pursuit target) happened ~50 ms after motion onset, then the flash was actually still

476 to the left of instantaneous gaze position because the target motion had not yet
477 crossed gaze position (with a step of 4 deg leftward followed by 20.4 deg/s rightward
478 motion, the target would still be leftward relative to gaze at ~50 ms). On the other
479 hand, if the same flash happened ~150 ms after motion onset, then the flash was
480 already ahead of instantaneous gaze position. According to our hypothesis of a
481 spatially-specific position error signal introduced by the flash, this means that both
482 "forward" and "backward" flashes relative to pursuit target location should transiently
483 reduce smooth pursuit acceleration if the flash happened ~50 ms after target motion
484 onset, but "forward" flashes should increase (rather than decrease) smooth pursuit
485 acceleration if the flashes happened ~150 ms after target motion onset.

486 This is indeed what we found. In Fig. 3A, B, we plotted data in a format similar
487 to Fig. 1A, B above, but only for "forward" flashes happening 50 ms after motion
488 onset (from monkey M); similarly, in Fig. 3C, D, we plotted the same data for
489 "backward" flashes happening 50 ms after motion onset (from the same monkey). In
490 both cases, transient reductions in eye acceleration happened. On the other hand,
491 when the flash happened 150 ms after motion onset, the transient stimulus increased
492 eye acceleration for forward flashes (Fig. 3E, F), while it reduced it for backward
493 flashes (Fig. 3G, H). This is because for forward flashes relative to target location, a
494 flash occurring 150 ms after target motion onset was also "forward" with respect to
495 instantaneous gaze position.

496 We summarized the observations above across both monkeys in Fig. 4A, B.
497 For each monkey, we measured the value of the velocity difference curve between
498 flash and no-flash trials (e.g. Fig. 1B, D, F, H and Fig. 3B, D, F, H). We did so by
499 averaging the value of the curve for each flash trial in the interval 50-80 ms after flash
500 onset; this gave us a population of measurements for any given flash time and flash
501 location, and we first performed statistics by estimating 95% confidence intervals on

such a population. We then plotted the velocity difference for a given flash type (e.g. forward or backward relative to pursuit target position) as a function of flash onset time relative to pursuit target motion onset (Fig. 4A, B; error bars denote 95% confidence intervals). The pink curve for each monkey shows the effect of a forward flash (relative to pursuit target position) on smooth eye velocity, and the green curve shows the effect of a backward flash. Clearly, there were effects of both forward and backward flashes on even open-loop pursuit initiation. Specifically, and consistent with Fig. 3, both monkeys showed reliable influences of spatially-localized flashes on smooth pursuit initiation, with a transition from inhibition (negative y-axis values) to enhancement (positive y-axis values) in the forward flash conditions (relative to no flash pursuit initiation) according to the observations of Fig. 3. We further validated this result by performing a two-way ANOVA (flash time by flash location) for each of the monkeys. As expected, we found a significant interaction between the two factors of flash time and flash location (monkey A: $F(14, 753) = 4.1$, $p = 6.4414*10^{-7}$; monkey M: $F(14, 1693) = 3.47$, $p = 1.3060*10^{-5}$), supporting the hypothesis that for longer delays, the two flashes were differentiating inhibition from enhancement of eye acceleration. In particular, starting from flashes happening at around 100 ms after motion onset (monkey A: 116 ms; monkey M: 92 ms), forward flashes led to a smaller inhibition that turned into an enhancement when compared to backward ones (Fig. 3). Therefore, open-loop smooth pursuit initiation is actually modifiable, and it reliably reflects the influence of transient instantaneous position error signals introduced by spatially localized flashes.

To show that enhancement of smooth pursuit initiation acceleration can still happen even for flashes as early as 50 ms after target motion onset, we repeated the experiment of Fig. 3 one more time, but this time, the forward or backward flashes happened 6 deg in front of or behind the pursuit target position. This means that even

528 if the flash happened ~50 ms after target motion onset, the flash in front of the pursuit
529 target was also in front of instantaneous gaze position, unlike in Fig. 3. In this case,
530 summary plots like those shown in Fig. 4A, B should show enhancement for forward
531 flashes even for early flashes; this was indeed the case (Fig. 4C). In this case, a two-
532 way ANOVA (flash location by flash time) showed a predominant main effect of the
533 factor flash location (monkey M: $F(1, 1707) = 121.03$, $p = 3.0190*10^{-27}$), statistically
534 supporting the interpretation that forward flashes induced enhancement while
535 backward flashes led to inhibition. We also observed a main effect of flash time
536 (monkey M: $F(14, 1707) = 2.53$, $p = 0.0014$) suggesting that later flashes had a lower
537 inhibitory component (in Fig. 4C, it is possible to appreciate this effect as a positive
538 slope across time for both flash directions) and a small interaction between the two
539 factors (monkey M: $F(14, 1707) = 1.8$, $p = 0.0341$). Therefore, even the inhibitory
540 influences seen for early forward flashes in Fig. 3A, B above were not a result of a
541 generalized inhibitory effect of flashes on eye movements, as sometimes suggested
542 from the saccadic inhibition literature with similar flashes (Reingold and Stampe,
543 1999, 2000, 2002; Buonocore and McIntosh, 2008). Instead, the flash was
544 introducing a spatially-defined geometric addition to the input of the oculomotor
545 system, as is also the case with the saccadic system (Hafed and Ignashchenkova,
546 2013; Buonocore et al., 2017). What is most intriguing is that such a spatially-specific
547 input was able to modify even so-called open-loop smooth pursuit initiation.
548 Finally, we also analyzed the effects of spatially localized flashes during
549 sustained pursuit. In the limit of steady-state smooth pursuit, a forward flash was also
550 effective in increasing saccade-free smooth pursuit eye velocity, and a backward
551 flash was effective in reducing smooth eye velocity (Fig. 5A, B; also see Fig. 2 for an
552 analog of this result with full-screen flashes). Therefore, transient instantaneous eye
553 position error signals have spatially-specific effects on both open-loop and sustained

554 smooth pursuit eye movements, and the results of Fig. 1 above are not merely a
555 generalized inhibitory effect of flashes on eye movements.
556
557 *Catch-up saccades are influenced by transient instantaneous position error signals*
558 *similarly to smooth pursuit eye velocity*
559 We explored the analogy between smooth pursuit initiation effects and saccade
560 effects alluded to above further. Specifically, we recently hypothesized that at the
561 time of flash-induced action potentials in oculomotor circuits, like the superior
562 colliculus (SC), a saccade that is triggered would have an amplitude reflecting the
563 addition of visually-induced action potentials to the already-preprogrammed saccade-
564 related action potentials (Buonocore et al., 2017). Thus, saccades would violate the
565 expected monotonic relationship (Zuber et al., 1965; Bahill et al., 1975) between
566 saccade amplitude and peak velocity, because these saccades would integrate
567 visually-driven action potentials with already-programmed motor-related action
568 potentials into the total eye displacement. Our smooth pursuit effects above (Figs. 1-
569 5) are consistent with this mechanism, and further support for this would emerge if
570 one were to analyze the trials in which catch-up saccades did occur after flash onset
571 in our smooth pursuit experiments (as opposed to only saccade-free trials in Figs. 1-
572 5). Therefore, during steady-state smooth pursuit and the 1x1 deg flashes
573 (Experiment 3; Materials and Methods), we took only trials in which a catch-up
574 saccade was triggered 50-150 ms after flash onset (Fig. 6A, G). Such a saccade
575 putatively happened at the same time at which visually-induced action potentials from
576 flashes exerted an influence over the oculomotor system in Figs. 1-5.
577 We aligned all catch-up saccades together and classified them as having a
578 direction either congruent or incongruent with “forward” or “backward” flashes relative
579 to the pursuit target in Experiment 3. For comparison, we also analyzed catch-up

saccades on no-flash control trials (using a virtual timing of fictive flashes that did not occur in reality). Figure 6B, C shows eye position (Fig. 6B) and eye velocity (Fig. 6C) for catch-up saccades from monkey A congruent with the flash location; the gray curves show catch-up saccades during steady-state smooth pursuit with no flashes, and all eye position curves were aligned to eye position at catch-up saccade onset before averaging. Similarly, Fig. 6D shows eye position and Fig. 6E eye velocity for catch-up saccades incongruent with flash location, and gray curves show no-flash catch-up saccades. Error bars in all cases denote 95% confidence intervals. As can be seen, for forward flashes, catch-up saccades triggered 50-150 ms after flash onset were larger than those without a flash (two-sample t-tests: $t = 6.992$; $df = 1707$; $p < 0.0001$) (Fig. 6B). Peak eye velocity seemed to also increase correspondingly for these movements (Fig. 6C). For backward flashes (Fig. 6D), we observed a small reduction in amplitude, as it might have been expected (Buonocore et al., 2016; 2017) and also a small reduction in peak velocity. In Figure 6F, we plotted the ratio between peak velocity and saccadic amplitude as an index of main sequence violation. Conceptually, if the peak velocity is not appropriately scaled to the larger/smaller saccades generated during the post-flash interval, the ratio between velocity and amplitude should lead to values bigger/smaller when compared to the ratio computed when no flash was presented (Buonocore et al., 2017). In monkey A, we observed that the peak velocity was in fact scaling with saccadic amplitude when backward flashes were presented. On the other hand, there was a trend for the ratio to be smaller than baseline for forward flashes (two-sample t-tests: $t = 1.571$; $df = 1707$; $p = 0.1163$) (Fig. 6F), suggesting the idea that main sequence violation can reliably happen during the post-flash period even with smooth pursuit. Similar results in amplitude modulation were observed for monkey M, but with a stronger and more significant trend for the saccades in violating the main sequence. (Fig. 6G-L). All the

statistics comparing catch-up saccades triggered in the no-flash conditions against catch-up saccades triggered when either forward or backward flashes were presented are reported in the caption of Fig. 6.

We also confirmed that during smooth pursuit initiation, there was again an effect on saccades and not just on smooth eye velocity. In Experiment 2 (Materials and Methods), target motion started directly from fixation, requiring an initiation saccade during smooth pursuit initiation. We analyzed the kinematics of such an initiation saccade (first saccade occurring after motion onset and with latency 30-80 ms from forward or backward flash onset) and compared them to kinematics of saccades when no flash was presented. We observed reliable modulations of saccade kinematics similar to the ones that we observed during steady state pursuit (Fig. 7). In the top row of Fig. 7, saccades were larger than baseline after a forward flash for both monkey A (Fig. 7A pink bar) and monkey M (Fig. 7B) (Monkey A, two-sample t-tests: $t = 9.113$; $df = 255$; $p < 0.0001$; monkey M, two-sample t-tests: $t = 8.411$; $df = 335$; $p < 0.0001$). Backward flashes reliably reduced saccade amplitudes relative to baseline in monkey A (two-sample t-tests: $t = -5.278$; $df = 217$; $p < 0.0001$) and monkey M (two-sample t-tests: $t = -8.205$; $df = 283$; $p < 0.0001$). To explore kinematic violations like in the case of catch-up saccades during sustained pursuit (Fig. 6), we plotted the ratio of peak velocity and amplitude for post-flash saccades (Fig. 7C, D). This ratio was different from baseline behavior in both monkeys. In particular, in monkey A, the increase in amplitude after forward flashes was not accompanied by a corresponding increase in peak velocity, resulting in a smaller ratio than baseline ($t = 2.459$; $df = 255$; $p = 0.0146$), while no significant violation was found for backward flashes. In monkey M, the violation after backward flashes occurred because saccade amplitude decreased without an appropriate scaling down of the peak velocity ($t = -6.053$; $df = 283$; $= p < 0.0001$).

632 Taken together, our results so far indicate that the sudden onset of a visual
633 transient can interfere with the spatial parameters of the motor plan by addition of
634 visual spikes, and even potentially making saccade amplitude relatively larger for a
635 given peak velocity in the case of forward flashes, and the opposite pattern for
636 backward flashes (Buonocore et al., 2016; Buonocore et al., 2017).
637
638 *The temporal structure of catch-up saccades is reset by transient visual onsets*
639 *similarly to the temporal structure of microsaccades during gaze fixation*
640 Finally, we explored the effects of localized visual flashes during stable fixation. The
641 stimuli that we used were identical to the ones of the previous experiments with the
642 only difference being that the fixation target was not moving in the present
643 experiments. In these conditions, we replicated the previous observation that flash
644 onset strongly reduced the probability to make a microsaccade in an interval of 50 to
645 150 ms after flash onset for both monkeys (Fig. 8A, B), a phenomenon known as
646 (micro)saccadic inhibition (Engbert and Kliegl, 2003; Rolfs et al., 2008; Hafed et al.,
647 2011; Hafed and Ignashchenkova, 2013). More interestingly, during such an
648 inhibitory period (very similar to the one observed for catch-up saccades; e.g. Fig.
649 6A, G), the kinematics of microsaccades were also modulated in a way similar to
650 what we observed for catch-up saccades generated during smooth pursuit initiation.
651 In particular, for both monkeys, flashes congruent with microsaccade direction were
652 pulling the amplitude of eye movements higher, but no effect was observed for
653 backward flashes (Fig. 8C, D). We validated this observation by performing a two-
654 way ANOVA (flash time by flash location) on microsaccade amplitude for each of the
655 monkeys. As expected, we found a significant interaction between the two factors
656 (monkey A: $F(1, 159) = 6.43$, $p = 0.0122$; monkey M: $F(1, 251) = 5$, $p = 0.0263$).
657 Follow-up analysis confirmed that flashes congruent with microsaccade direction

were indeed leading to the generation of larger microsaccades during the inhibitory period (monkey A, two-sample t-tests: $t = -5.53$; $df = 72$; $p < 0.0001$; monkey M, two-sample t-tests: $t = -3.113$; $df = 131$; $p = 0.0023$). We then tested if the microsaccades launched during the inhibition interval (50-150 ms after flash onset; Fig. 8A, B) were also violating the main sequence. As predicted from previous research on the topic (Guillaume, 2012; Buonocore et al., 2016; Buonocore et al., 2017), and from the results reported above, a pattern of violation was revealed again in both animals. In particular, we observed a general reduction in the velocity over amplitude ratio for both animals (Fig. 8E, F). A two-way ANOVA (flash time by flash location) highlighted a significant main effect of flash time (monkey A: $F(1, 159) = 6.41$, $p =0.0123$; monkey M: $F(1, 251) = 4.03$, $p = 0.0457$) with no interaction between factors, supporting the hypothesis that the peak velocity was not appropriately scaling with the modulations in amplitude caused by flash onset (Buonocore et al., 2017).

After confirming similarities between the spatiotemporal parameters of catch-up saccades produced during smooth pursuit and microsaccades generated during stable fixation, we turned to analyze the temporal structure of catch-up saccades and microsaccades when no flashes were presented. As a first analysis, we compared inter-saccadic intervals (Fig. 9A, B) and saccade rates (Fig. 9C, D) in the two tasks. In Fig. 9C, D, it is possible to appreciate that both animals produced eye movements with a similar rate, independently of the task being sustained pursuit (orange curve) or stable fixation (violet curve). Moreover, the inter-saccadic interval (i.e. the timing between consecutive movements) was also mainly overlapping between the two tasks, and it was peaking around 200-250 ms in both animals (Fig. 9A, B). Taken together, these results support the hypothesis that the temporal structure of microsaccades and catch-up saccades is similar .

The third observation in our comparison of catch-up saccades to microsaccades regards instead the eye position error (i.e. the Euclidian distance between the current fixation location and the eye position 10 ms before producing a saccade). In Fig. 9E, F, we could observe clearly that there was a substantial difference in the position error between sustained pursuit (orange) and stable fixation (violet). In particular, both monkeys had a higher position error during sustained pursuit, as might be expected. This finding supports the hypothesis that the urge to make a saccade (i.e. when to trigger an eye movement) was relatively independent of the eye position error (i.e. where the eyes are relative to the target/fixation) within the ranges of position error that were present in our study. One might therefore argue that, up to a certain threshold, an internal clock governing the triggering of (micro)saccades is fairly constant and unaltered by the ongoing velocity of the eyes (Tian et al., 2018).

Discussion

We injected brief, perceptually subtle visual flashes around the time of smooth pursuit initiation, and we observed robust modulations in eye acceleration. These modulations were spatially-specific, depending on flash location relative to instantaneous gaze position, and they were therefore not reflecting a generalized inhibitory effect similar to freezing behavior in prey animals. Instead, eye acceleration during pursuit initiation was equally easy to increase or decrease, even for flashes appearing as early as ~50 ms after target motion onset (i.e. even before the onset of smooth tracking itself). We also observed that when smooth pursuit was initiated with the aid of catch-up saccades, these saccades were affected by flashes similarly to how microsaccades (Buonocore et al., 2017) and larger saccades (Buonocore et al., 2016) are affected, and also similarly to how the smooth eye movements themselves were modulated. Our results demonstrate that so-called “open-loop” smooth pursuit is more nuanced than assumed, and they motivate neurophysiological investigations of rapid sensory and motor processing in the oculomotor system.

Beyond saccadic inhibition

Our experiments were motivated by related research investigating the impacts of brief flashes on saccades (Reingold and Stampe, 1999; Buonocore and McIntosh, 2008). Initially, such flashes were discovered to transiently inhibit saccades (Reingold and Stampe, 1999) and microsaccades (Engbert and Kliegl, 2003; Rolfs et al., 2008; Hafed et al., 2011). However, later research demonstrated that the flashes also have directionally-specific influences (Hafed and Clark, 2002; Engbert and Kliegl, 2003; Hafed and Ignashchenkova, 2013; Tian et al., 2016; Tian et al., 2018), as well as effects on saccade kinematics (Buonocore et al., 2016; Buonocore et al., 2017). For example, microsaccades occurring 50-100 ms after flash onset and directionally-

723 congruent with its location are not inhibited as strongly as opposite microsaccades
724 (Hafed and Ignashchenkova, 2013; Tian et al., 2016; Tian et al., 2018), and
725 sometimes even enhanced (as in Fig. 6G for monkey M catch-up saccades).
726 Moreover, congruent microsaccades are enlarged, whereas opposite microsaccades
727 are smaller, as if to reflect averaging of the saccade-vector command with "visual"
728 action potentials induced by flash onset (Hafed and Ignashchenkova, 2013; Tian et
729 al., 2016; Buonocore et al., 2017).

730 We observed the same kinematic alterations in catch-up saccades as in
731 microsaccades/saccades (e.g. Figs. 6-8) even though catch-up saccades rode on
732 high eye velocity. Catch-up saccades were also inhibited after flash onset, and with
733 directionally-specific patterns (e.g. Fig. 6A, G). In fact, we ran our monkeys on a
734 fixation task in the same sessions, and replicated our recent findings (Hafed and
735 Ignashchenkova, 2013; Tian et al., 2016; Buonocore et al., 2017). Our results on
736 catch-up saccades therefore support recent statements that these saccades behave
737 similarly to microsaccades (Heinen et al., 2016; Heinen et al., 2018). We now extend
738 this idea further to include the effects of transient flashes on the saccadic system.

739 Most intriguingly, even when no catch-up saccades occurred, we still observed
740 direct correlates of the saccadic phenomena described above on smooth eye
741 movements. Smooth pursuit initiation was enhanced or inhibited depending on flash
742 location, and sustained pursuit was also affected. The most interesting added
743 advantage of performing the experiments on smooth pursuit rather than saccades
744 was that we could focus on open-loop saccade-free pursuit initiation, demonstrating
745 that it is equally malleable as sustained pursuit. We specifically found that open-loop
746 pursuit is easily modifiable by visual signals injecting an instantaneous transient eye
747 position error. Thus, the term "saccadic inhibition" does not encompass the entire
748 space of impacts of brief flashes on the oculomotor system.

In our experiments, the catch-up saccades of monkey A, in particular, after flash onset were not strongly reduced in amplitude for backward flashes, but they were significantly larger after forward flashes (Fig. 6B, D). We think that observations like these may be interpreted as reflecting the fact that smooth pursuit was ongoing. For example, backward flashes reduced smooth eye velocity itself in addition to influencing saccades (Figs. 2, 5). Therefore, there was a conflict between the increased eye position error due to slower eye velocity (supporting a bigger catch-up saccade) and the backward visually-induced action potentials of the flash (supporting a smaller catch-up saccade). Given the importance of eye position error in smooth pursuit (de Brouwer et al., 2002), it could be the case that the reduction in smooth velocity compensated for effects of the flash on catch-up saccade amplitude. By a similar vein, because monkey A had slightly lower eye velocity than monkey M in tracking (Fig. 2), there was a bigger likelihood that there was positive eye position error at the time of the flash onset. This condition, of a plan to make a corrective eye movement coinciding with a congruent visual flash in the same direction, is known in microsaccades to have maximal effects on the movement being planned (Tian et al., 2016; Tian et al., 2018). That is why monkey A had a larger effect of forward flashes on catch-up saccade amplitude (Fig. 6B).

Smooth pursuit mechanisms

We also observed smooth velocity effects that we think reflect the constant interplay between target position/motion and the instantaneous ongoing eye behavior. For example, we found that smooth velocity effects after forward flash onset had a robust post-enhancement suppression phase. For example, for forward flashes in Experiments 1 and 3, smooth pursuit acceleration or sustained velocity increased (what we termed enhancement in Results) and then decreased later (resulting in a

later dip in the velocity difference curves; e.g. Fig. 5). Such a subsequent decrease could reflect the simple fact that the enhancement causes gaze to overpass the pursuit target, resulting in negative eye position error. A brief deceleration after the enhancement thus brings pursuit back on target.

Therefore, our smooth velocity effects relate, in a sense, to studies of distractor effects on pursuit (Spering et al., 2006). We think that we complement these observations and highlight the relevance of position error for influencing smooth eye velocity. Such an influence even happens during fixation with smooth ocular drifts (Martins et al., 1985; Kowler and Blaser, 1995). In all, this suggests interesting future neurophysiology experiments that not only focus on the specification of target motion signals for pursuit, but that investigate, at the level of the brainstem and other cerebellar and cortical areas, the aggregate interplay between visual and motor signals, with and without transient flashes. One prediction of our work is that SC recordings should reveal simultaneity of flash-induced visual action potentials with alterations in smooth eye velocity, but it would be interesting to know whether other areas down- or upstream of SC play a more critical role.

What triggers catch-up saccades?

An additional interesting observation from our fixation experiments is that ongoing microsaccade rate and inter-saccadic intervals during sustained fixation were qualitatively similar to those during sustained pursuit, despite different position errors. Even flash-induced modulations in microsaccade or catch-up saccade rate were similar to each other (i.e. there was rate inhibition followed by rate enhancement with similar dependencies on saccade direction relative to flash location; Figs. 6, 8). This raises an intriguing question on what triggers catch-up saccades. It is known that position error and retinal slip (velocity error) are important for triggering these

801 saccades (de Brouwer et al., 2002), but there is also tolerance for these errors such
802 that some combinations of position and velocity errors go briefly uncorrected. This
803 could explain why a “catch-up saccade rate signature” after flash onset looks
804 qualitatively identical to the so-called “microsaccade rate signature” after flash onset.
805 Time since the previous catch-up saccade seems to also matter, and could
806 additionally explain the above tolerance for position and velocity errors. Indeed, there
807 seems to be temporal structure to when catch-up saccades occur during smooth
808 pursuit, similar to that for microsaccades (Fig. 9). Thus, the effect of flashes on the
809 catch-up saccade rate signature during pursuit (Fig. 6A, G) could be explained by the
810 same phase-resetting mechanisms associated with microsaccadic temporal structure
811 (Hafed and Ignashchenkova, 2013).
812 Microsaccades are also sensitive to eye position error (Guerrasio et al., 2010;
813 Ko et al., 2010; Tian et al., 2016), extending the similarities to catch-up saccades
814 mentioned earlier. In the end, a combination of time since the last saccade and
815 oculomotor errors must matter a great deal for what triggers either catch-up or
816 microsaccades (Tian et al., 2018).
817
818 *Implications for experiments*
819 Finally, experiencing our flashes in pilot human experiments revealed a huge
820 contrast between their perceptual appearance to the observer and their impacts on
821 the oculomotor system. These brief stimuli (even full flashes) seem, at face value, to
822 be too transient and inconsequential to affect eye movement behavior (or even
823 perceptual processing). Yet, they do, and in a most robust manner (e.g. Fig. 1). This
824 means that certain experimental manipulations might seem so subtle to task
825 designers but fundamentally systematic for the brain processes being invoked by the
826 tasks. This can influence experimental result interpretations. For example, there is

interest in probing perceptual state during smooth pursuit initiation by presenting brief stimuli around the time of eye movements (Spering et al., 2006; Schutz et al., 2008; Braun et al., 2017). Because our results suggest that pursuit is indeed altered by brief stimuli, even during initiation, ideal stimuli for these kinds of experiments would be ones that would not alter retinal image statistics if smooth pursuit were to be transiently slowed down or accelerated. Otherwise, experimental results may reflect differences in retinal image statistics when pursuit is altered, rather than more interesting perceptual and cognitive factors. A second similar example is the employment of brief stimulus flashes as attentional cues during fixation and assuming that microsaccades are irrelevant, an assumption that we now know to be too simplistic (Hafed et al., 2015).

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982

983

984 **Figure legends**

985 **Figure 1** Transient disruption of open-loop smooth pursuit initiation. (A) We plotted
986 radial eye velocity from monkey M in Experiment 1 when no flashes were presented
987 (gray) and when full flashes were presented 50 ms after target motion onset (light
988 blue). There was a transient dip in eye acceleration relative to the gray curve starting
989 ~50 ms after flash onset. (B) Difference in eye velocity between flash and no-flash
990 trials from A. The flash-induced dip in the difference curve is clearly visible. (C, D)
991 Similar to A, B but for flashes occurring 83 ms after target motion onset. In this case,
992 the transient disruption in open-loop pursuit initiation was slightly delayed relative to
993 A, B because of the slight delay in flash onset time. (E, F) Same for flashes 117 ms
994 after target motion onset. (G, H) Same for flashes after 150 ms from target motion
995 onset. (I) In all cases, and for both monkeys, the disruption was always aligned on
996 flash onset. We plotted eye velocity difference curves as in B, but now aligning the
997 data on flash onset. The different colors denote different flash onset times relative to
998 target motion onset. Regardless of flash time, the velocity difference curves always
999 showed a dip ~50 ms after flash onset. A series of one sample t-tests against zero
1000 showed $p<0.05$ (Bonferroni corrected) for all flash times for both monkeys (with the
1001 exception of flash time 83.33 in monkey A). Error bars in all panels denote 95%
1002 confidence intervals.

1003

1004 **Figure 2** Transient disruption of sustained smooth pursuit. (A) We repeated analyses
1005 similar to Fig. 1 but during sustained smooth pursuit from Experiment 3. In monkey A,
1006 we plotted radial eye velocity with no flashes were presented (gray) or after full flash
1007 onset (light blue). There was a transient reduction on smooth eye velocity ~50 ms
1008 after flash onset. (B) Same for monkey M. Error bars denote 95% confidence
1009 intervals. Note that monkey A had lower sustained pursuit eye velocity than monkey

1010 M (compare gray curves in both panels). This monkey relied more on catch-up
1011 saccades, as will be evidenced by the catch-up saccade rates discussed later (Fig 9.
1012 C, D).
1013
1014 **Figure 3** Directionally-specific modulation of open-loop smooth pursuit initiation by
1015 injected transient eye position error signals. (A, B) Example velocity (A) and velocity
1016 difference (B) traces from monkey M in the same format as in Fig. 1A, B. A flash was
1017 presented 2.1 deg ahead of the pursuit target only 50 ms after target motion onset.
1018 Because pursuit had not yet started at flash onset and because of the geometry of
1019 flash location, the flash was still behind gaze position when it occurred (see text).
1020 This caused a transient dip as in Fig. 1A, B. (C, D) Similar analyses for a flash behind
1021 the pursuit target, again causing a transient dip as in A, B. (E, F) When the forward
1022 flash happened 150 ms after flash onset, it was now ahead of instantaneous gaze
1023 position, and now caused enhancement rather than inhibition of pursuit initiation. (G,
1024 H) A backward flash at the same time caused inhibition. Therefore, pursuit
1025 disruptions are not generalized inhibitory effects, but they reflect spatially-specific
1026 influences of flash position relative to current gaze position. Error bars denote 95%
1027 confidence intervals.
1028
1029 **Figure 4** Spatially-specific transient modulations of open-loop smooth pursuit
1030 initiation depending on flash location. (A, B) For forward and backward flashes 2.1
1031 deg ahead of or behind the pursuit target, we confirmed the observations in Fig. 3 in
1032 both monkeys. Very early flashes caused inhibition and later ones caused
1033 enhancement in smooth pursuit velocity/acceleration. For each flash time, we
1034 measured the eye velocity difference in an appropriate interval after flash onset
1035 (Materials and Methods), and we plotted this measurement as a function of flash

1036 onset time relative to target motion onset. For reference, smooth pursuit initiation
1037 typically happens at ~100 ms, meaning that the measurements shown all illustrate
1038 modifications of open-loop smooth pursuit initiation. (C) When the flash was at 6 deg
1039 instead of 2.1 deg, enhancement occurred even for the very earliest flashes,
1040 supporting the conclusion that the early inhibition in A, B was not a generalized
1041 inhibitory effect, but that it depended on instantaneous flash location relative to
1042 instantaneous gaze location. Error bars denote 95% confidence intervals.
1043
1044 **Figure 5** Spatially-specific modulations of sustained smooth pursuit eye movements.
1045 (A, b) In both monkeys, we repeated analyses similar to Fig. 2, but for localized
1046 flashes ahead of or behind the pursuit target (by 2.1 deg). The gray curve shows
1047 sustained pursuit velocity without flashes. The colored curves show velocity
1048 modulations with short latency after flash onset. Forward flashes caused enhanced
1049 smooth eye velocity followed by reduction. Backward flashes primarily caused only
1050 reduction in eye velocity. Note that the reduction in eye velocity after the
1051 enhancement in the forward flash conditions may reflect that the enhancement
1052 caused pursuit to overpass the target. Subsequent slowing of the eye could return
1053 gaze to its normal position relative to the target when no flashes take place. Error
1054 bars denote 95% confidence intervals.
1055
1056 **Figure 6** Modulations in catch-up saccades during sustained smooth pursuit by flash
1057 onsets. (A) Catch-up saccade rate as a function of time from flash onset for forward
1058 and backward flashes. A characteristic inhibition followed by rebound was obvious,
1059 and it was very similar to microsaccadic modulations during fixation (e.g. see Fig. 8A,
1060 B). Note that at the onset of inhibition, the forward flash condition was less effective
1061 in inhibiting saccades as the backward flash condition (also see G for an even

stronger effect in monkey M). These few saccades that did occur, were often dramatically altered by the flash. (B) For example, aligning eye position on catch-up saccade onset for these specific post-flash saccades reveals that they were larger (pink) than normal catch-up saccades (gray) when a forward flash was presented. (C) Their radial eye velocity was also higher than when no flash occurred. (D, E) Backward flashes caused minimal change in saccade amplitude. (F) When we checked saccade kinematics for the specific post-flash saccades occurring near the onset of the inhibition phase in A, we found that the larger saccades in B violated the main sequence curve of peak velocity versus amplitude for saccades: relative to baseline (gray curve), their peak velocity did not scale as much as the scaling of amplitude. This is consistent with microsaccadic effects in the literature (also see Fig. 8C, D). (G-L) Similar analyses for monkey M. Also in this case, forward flashes affected catch-up saccade amplitude (two-sample t-tests: $t = 3.701$; $df = 1742$; $p = 0.0002$) (H, J), and backward flashes made catch-up saccades violate the main sequence curve (two-sample t-tests: $t = 1.884$; $df = 1683$; $p = 0.0598$) (green in L). Error bars denote 95% confidence intervals.

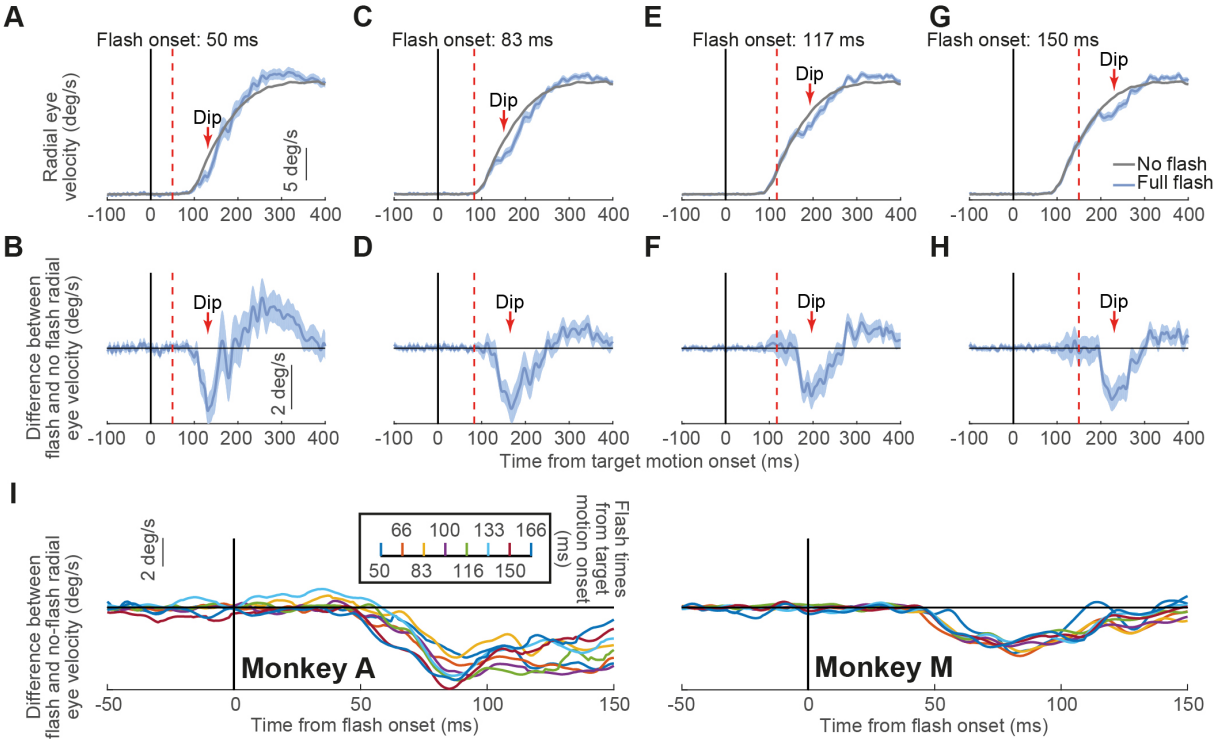
Figure 7 Alterations of catch-up saccade kinematics in pursuit initiation with saccades (Experiment 2). (A, B) Catch-up saccade amplitudes for post-flash saccades were larger than in no flash trials (gray curve) when a forward flash was presented but smaller when a backward flash was presented. (C, D) Similar to Fig. 6, the changes in saccade amplitudes were often associated with altered kinematics (Buonocore et al., 2016). For example, monkey A had lower peak velocity than expected for forward flashes, similar to Fig. 6, and monkey M had higher peak velocity than expected for backward flashes. Thus, kinematic alterations occur during

sustained pursuit (Fig. 6) and also during pursuit initiation (this figure). Error bars denote 95% confidence intervals.

Figure 8 Analyses similar to Figs. 6-7 for microsaccades during gaze fixation. (A, B) Microsaccade rate was robustly affected by flash onset, as expected, and very similarly to the catch-up saccade effects during sustained pursuit. (C-F) Microsaccade amplitude and kinematic effects were also altered, but in an even stronger manner than during smooth pursuit. These analyses all replicate our previous study (Buonocore et al., 2017). Error bars denote 95% confidence intervals.

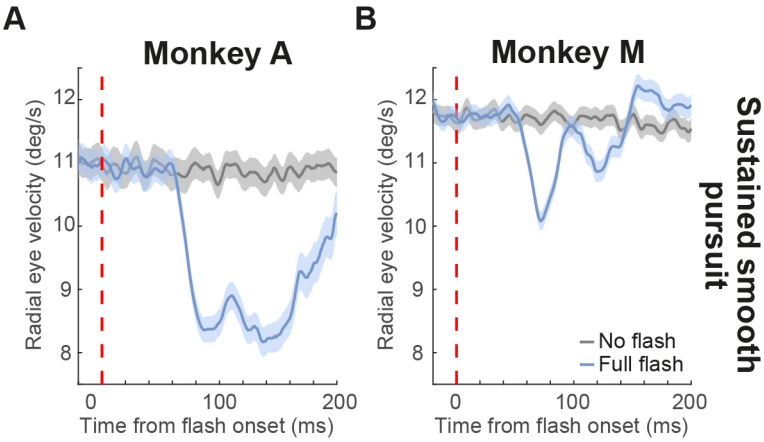
Figure 9 Similar temporal structure of catch-up saccades to microsaccades despite significantly larger eye position errros during pursuit. (A, B) We plotted inter-saccadic intervals during pursuit or fixation. The distributions looked very similar in both tasks, although monkey A often had very rapid back-to-back microsaccades with 100 ms inter-saccadic interval. (C, D) Microsaccade and catch-up saccade rates were also comparable in general. Monkey A made more catch-up saccades because of the lower pursuit eye velocity (Fig. 2). (E, F) Despite similarities in inter-saccadic intervals between the tasks, pursuit was always associated with larger eye position errors at saccade onset than fixation. Error bars denote 95% confidence intervals.

Figure 1


A
Flash onset: 50 ms
Radial eye velocity (deg/s)
Dip
5 deg/s
C
Flash onset: 83 ms
E
Flash onset: 117 ms
G
Flash onset: 150 ms
No flash
Full flash
B
Difference between flash and no flash radial eye velocity (deg/s)
2 deg/s
D
F
H
-100 0 100 200 300 400
Time from target motion onset (ms)
I
Difference between flash and no-flash radial eye velocity (deg/s)
2 deg/s
66 100 133 166
50 83 116 150
Flash times from target motion onset (ms)
Monkey A
Monkey M
-50 0 50 100 150
Time from flash onset (ms)

1113 Figure 2

1114



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1118 Figure 3

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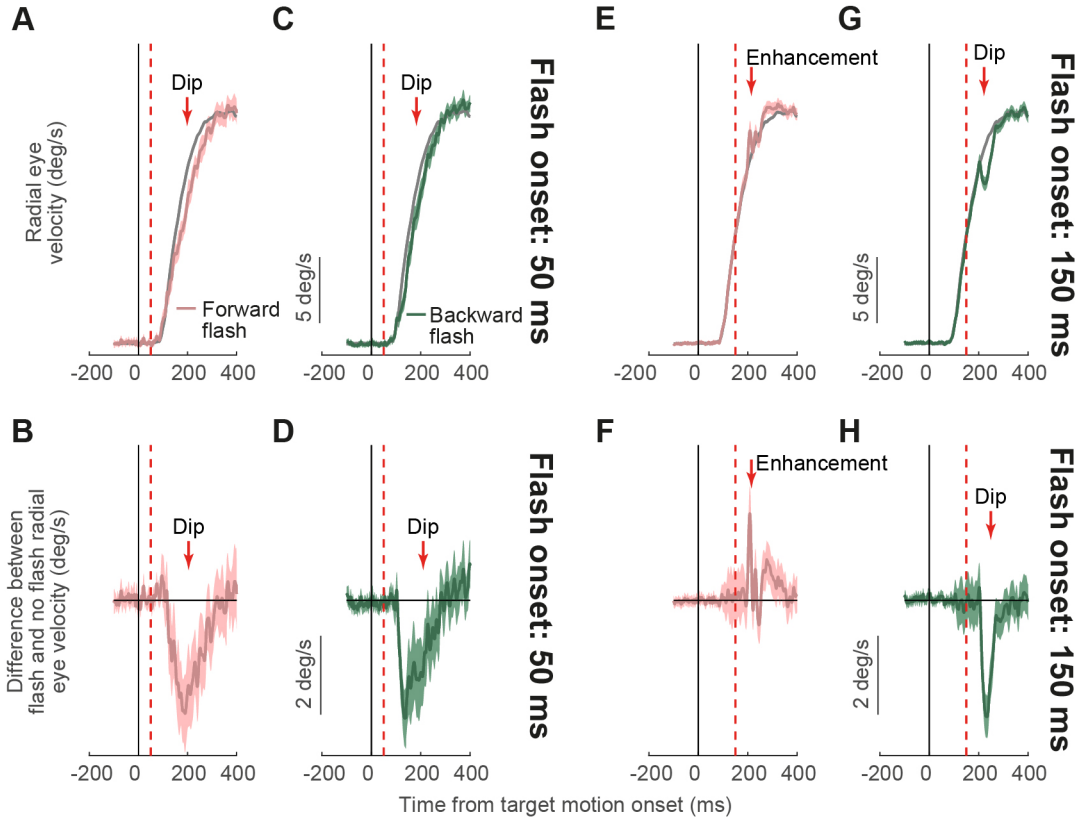

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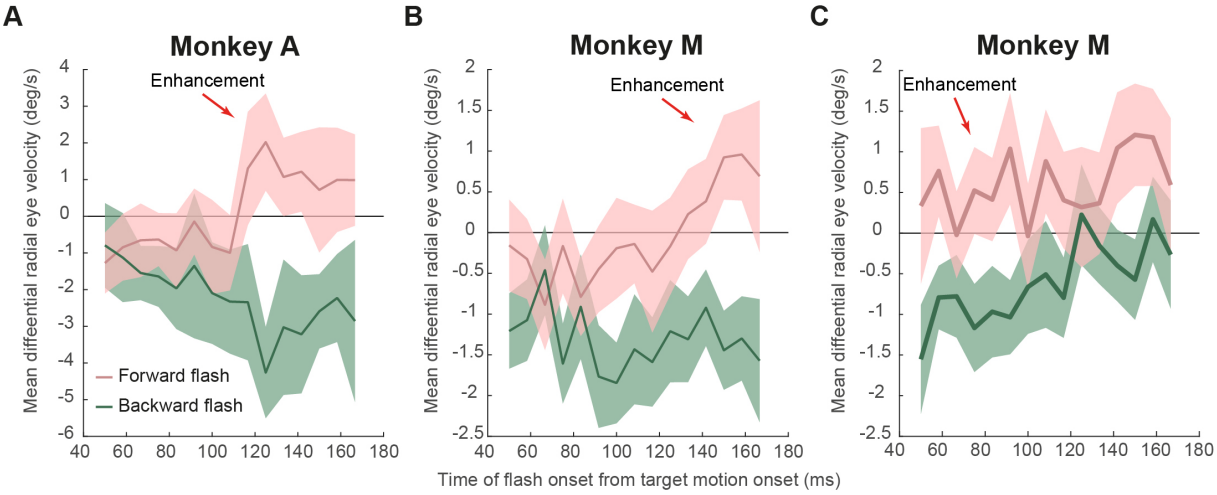
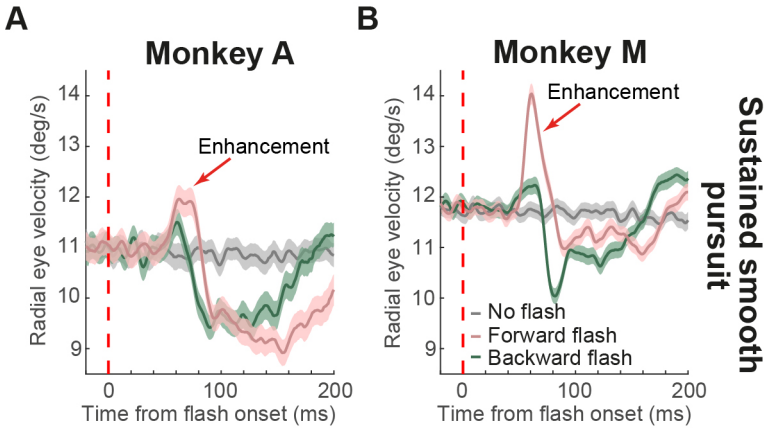
Figure 4


A
Monkey A
Enhancement
Mean differential radial eye velocity (deg/s)
Forward flash
Backward flash
B
Monkey M
Enhancement
C
Monkey M
Enhancement
Time of flash onset from target motion onset (ms)

1128 Figure 5

1129



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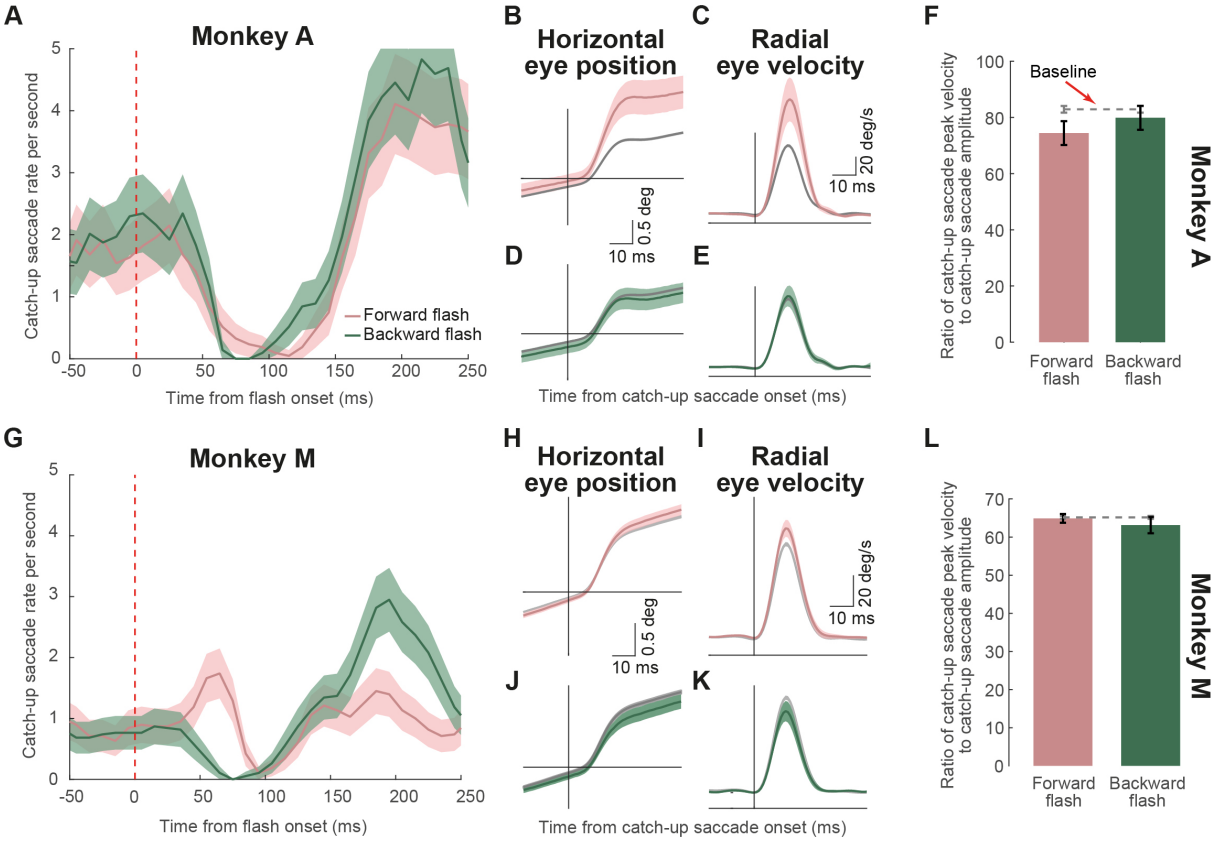
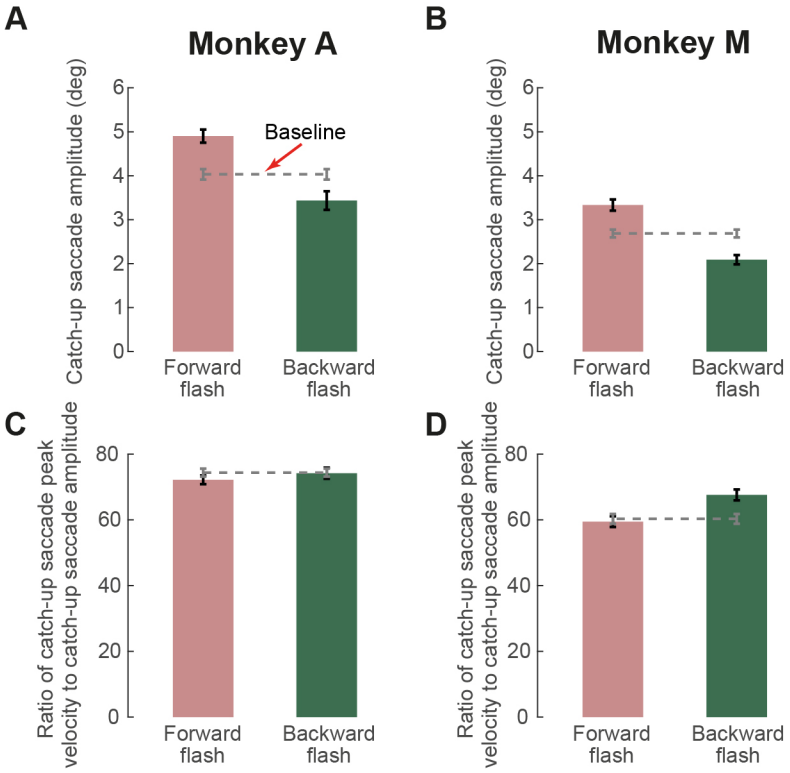
1132

Figure 6


A
Monkey A
Catch-up saccade rate per second
Forward flash
Backward flash
Time from flash onset (ms)
B
Horizontal eye position
0.5 deg
10 ms
C
Radial eye velocity
20 deg/s
10 ms
D
E
Time from catch-up saccade onset (ms)
F
Baseline
Ratio of catch-up saccade peak velocity to catch-up saccade amplitude
Forward flash
Backward flash
Monkey A
G
Monkey M
Catch-up saccade rate per second
Time from flash onset (ms)
H
Horizontal eye position
0.5 deg
10 ms
I
Radial eye velocity
20 deg/s
10 ms
J
K
Time from catch-up saccade onset (ms)
L
Ratio of catch-up saccade peak velocity to catch-up saccade amplitude
Forward flash
Backward flash
Monkey M

1138 Figure 7

1139



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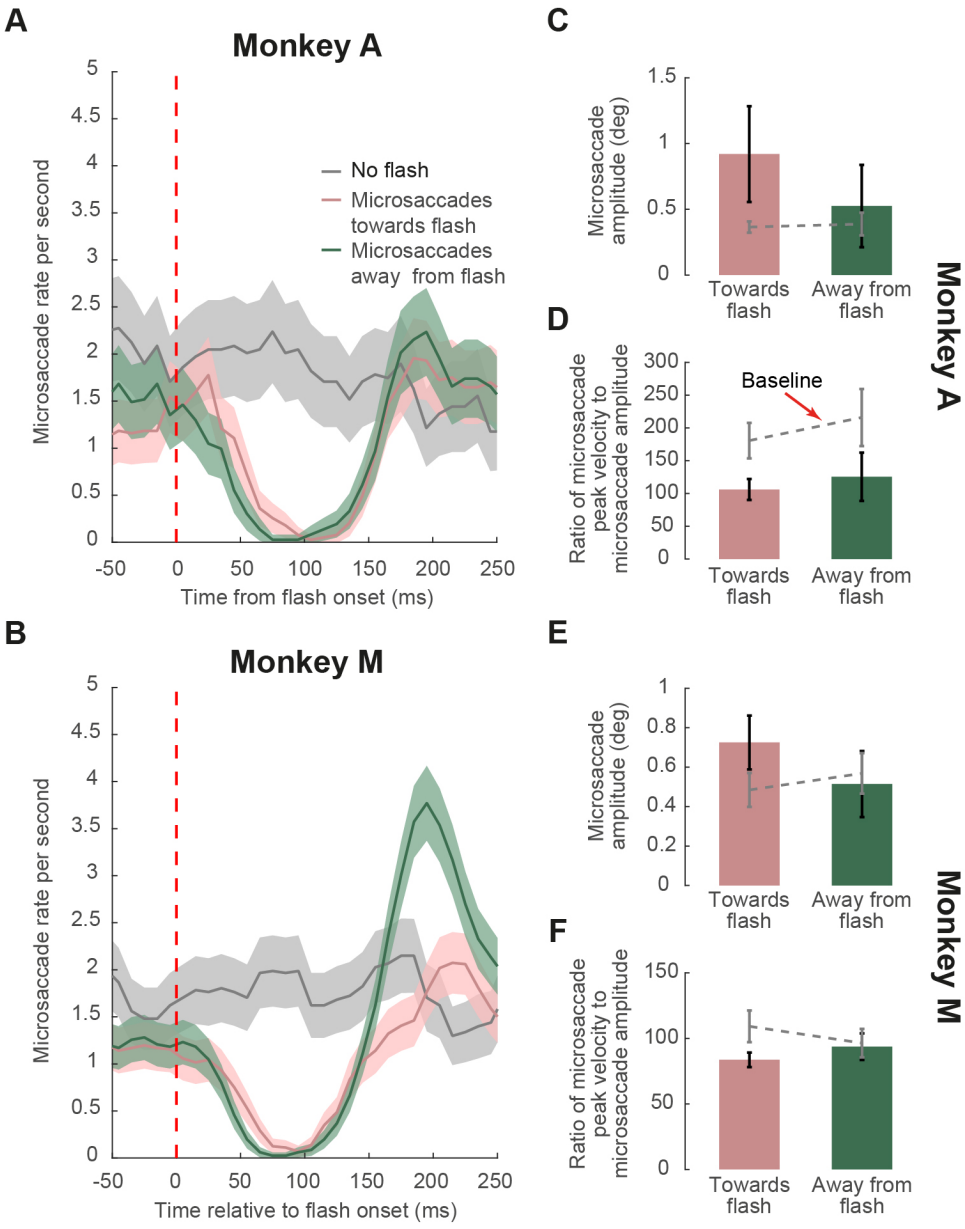
1142

1143 Figure 8

1144



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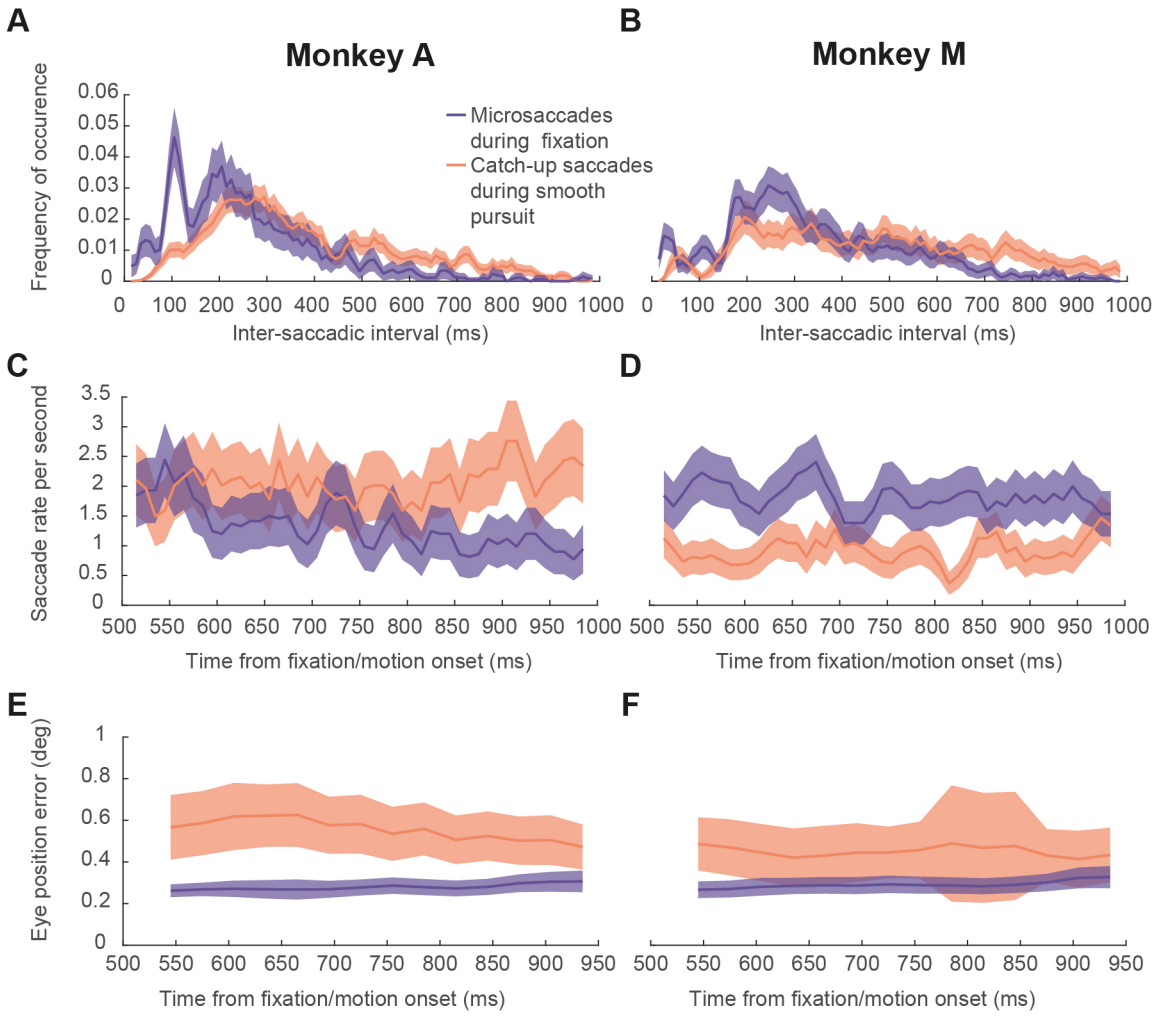
1150 Figure 9

1151



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