

Frames of reference for saccadic command, tested by saccade collision in the supplementary eye field

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Running Head: Saccade collision in SEF

Keywords: Supplementary eye field, saccade collision, frame of reference, fixed-vector saccade, goal-directed saccade

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ABSTRACT

In what frame of reference does the supplementary eye field (SEF) encode saccadic eye movements? In this study, the “saccade collision” test was used to determine whether a saccade electrically evoked in the monkey’s SEF is programmed to reach an oculocentric goal or a non-oculocentric (e.g. head or body-centered) goal. If the eyes start moving just before or when an oculocentric goal is imposed by electrical stimulation, the trajectory of the saccade to that goal should compensate for the ongoing movement. Conversely, if the goal imposed by electrical stimulation is non-oculocentric, the trajectory of the evoked saccade should not be altered. In head-fixed experiments, we mapped the trajectories of evoked saccades while the monkey fixated at each of 25 positions 10° apart in a 40° by 40° grid. For each studied SEF site, we calculated convergences indices, and found that “convergent” and “non-convergent” sites were separately clustered: non-convergent rostral to convergent. Then, the “saccade collision” test was systematically applied. We found compensation at sites where saccades were of the “non-convergent” type and practically no compensation at sites where saccades were of the “convergent” type. The results indicate that the SEF can encode saccade goals in at least two frames of reference, and suggest a rostro-caudal segregation in the representation of these two modes.

INTRODUCTION

One of the best arguments that led to the identification of an oculomotor area at the dorso-medial edge of the frontal lobe in monkeys was the ease with which saccades could be electrically evoked from this region (Fujii et al. 1995; Huerta and Kaas 1990; Russo and Bruce 1993; Schall 1991b; Tehovnik and Lee 1993; Tehovnik et al. 1998; Tian and Lynch 1995), sometimes with currents as small as 10 μ A (Schlag and Schlag-Rey 1987a). Its oculomotor function has been verified by a number of studies, notably by recording single units that start bursting prior to saccades in a particular direction (Bon and Luchetti 1992; Isoda and Tanji 2004; Kim et al. 2005; Mann et al. 1988; Mushiaki et al. 1996; Russo and Bruce 1996; Schall 1991a; Schlag and Schlag-Rey 1987a). However, still today, there is little agreement on the significance of this region now identified as the supplementary eye field (SEF). At issue is not only finding a common denominator among the roles postulated for the SEF in various cognitive tasks (Amador et al. 2000; Chen and Wise 1995; Fukushima et al. 2004; Lu et al. 2002; Olson and Gettner 1999; Schlag-Rey et al. 1997) but even, at a more elementary level, recognizing the frame of reference used in the SEF to program eye movements. How are encoded the goals of saccades evoked by electrical stimulation of SEF sites? Is it in the frame of an eye position or a head position or body position (Martinez-Trujillo et al. 2003a)? The answer is not obvious because saccades electrically evoked from the SEF appear to be of two kinds: fixed-vector and goal-directed.

In experiments on monkeys with head-fixed, the amplitude and direction of fixed-vector saccades (Fig. 1, A1) are practically constant, at least within 20-30° of the primary eye position, which is the area in which the effect of an electrical stimulation is usually tested. Saccade dimensions are solely a function of the site where stimulation is applied (thus the label "fixed-vector"). Long stimulation trains do not make saccades longer but, eventually, triggers successive stereotyped saccades at intervals of about 100 ms (a phenomenon called stair-case saccades, Robinson 1972). It is only when fixed-vector saccades extend to more than 20-30° from the primary eye position in the direction

toward which the eyes move that saccade trajectories clearly appear to converge. As Klier et al. (2001) pointed out, the appearance of convergence is illusory; it is due to the way trajectories of eye rotation are usually plotted on a flat map with a linear scale in degrees. When so represented, parallel fixed-vectors appear to converge, and the site of convergence is necessarily at 90° from the primary eye position. A point at 90° on such a map is at a finite distance from center (corresponding to the maximal coil signal) whereas, in fact, an eye turned to 90° (were it possible) would look at an infinite distance on a frontal plane. Fixed-vector saccades are the most common type of evoked saccades. They were first observed in stimulations of the superior colliculus (SC, Robinson 1972) and frontal eye field (FEF, Robinson and Fuchs 1969). For a long time, fixed-vector saccades were thought to be the only type that could be produced by stimulation anywhere in the brain.

Goal-directed saccades evoked from the SEF (Fig. 1, *B1*) are quite different. Provided that the duration of stimulation is long enough (i.e. lasting up to the end of the evoked movement) and current is above threshold, saccade amplitude and direction vary depending on the orientation of the eyes in orbit at the time of stimulation. Here convergence is real: what is invariant is no longer the vector of evoked saccades but the position of the goal. This goal cannot be oculocentric (i.e. specifiable in retinal coordinates) since its location in space remains the same whatever the initial eye orientation. Trajectories of goal-directed saccades starting far away from the goal are longer, saccade latencies tend to be shorter, and the current threshold is lower (as if the stimulated neurons were more responsive in this condition). Conversely, no movement is produced by stimulation when the eyes are already pointing within the goal or close to it. Goal areas can be small (e.g. 6° in diameter) or large (e.g. a whole quadrant) and generally their center is more than 20° eccentric.

Unfortunately, not all evoked saccades neatly correspond to one of these two prototypes. Sometimes, at least one of the attributes specified above appears missing. For instance, in one variant of the fixed-vector type, saccades cannot be evoked when the

eyes are initially oriented to a part of the field. This “unresponsive” area usually lies in the direction where normally saccade ends, and it may just occupy an entire contralateral hemi-field. As shown for instance in the case of Fig. 1, A2, no saccades could be produced that started or terminated beyond the vertical midline (however, inducing saccade collision could sometimes override this limitation). This unresponsiveness within a large visual area has been noticed in the SEF (Tehovnik and Lee 1993; Tehovnik et al. 1998), but, as far as we know, has never been reported in any other oculomotor structure.

There are also variants of goal-directed saccades, for instance, when trajectories are cut short before reaching the goal (even though the stimulation outlasts the saccade). Such saccades (Fig. 1, B2) have also been described by stimulation of the cortex around the intraparietal sulcus (Thier & Andersen 1996).

The usual explanation for the fixed-vector type of saccade is that stimulation evokes a goal whose position is specified in a retinocentric (or oculocentric) frame of reference (Schlag and Schlag-Rey 1987b). For instance, imagine that a phosphene is produced by stimulation. Then the effect of the electrical stimulation is to move the eyes toward this goal defined by a direction and a distance from the fovea [but note that Klier et al. (2001) use the term "retinal" instead of the usual term "fixed-vector" to describe this type of saccades]. In contrast, the goal for the goal-directed type is assumed to be specified in some higher level - "non-oculocentric" - frame of reference. The latter reference could be to head, body, or environment. To determine which of the latter is the correct frame of reference, the effect of stimulation should be tested with various head and/or body rotations (Klier et al. 2001; Martinez-Trujillo et al. 2003a,b). When this is not done (as in the present study), one has to use an uncommitted term like "non-oculocentric" to indicate that the frame of reference is "*something beyond oculocentric*" that we cannot further specify.

Already in early studies of the SEF, attempts were made to differentiate objectively the fixed-vector and goal-directed characteristics of electrically evoked

saccades. Two indexes of convergence were proposed: one index measures the maximum convergence angle, the other measures the maximum difference of amplitude of saccades evoked from initial points close to versus far away from the goal (Schlag and Schlag-Rey 1987a). Such measurements were much refined and extensively used by Russo and Bruce (1996), Klier et al. (2001) and Martinez-Trujillo et al. (2003b,c). Even so, little agreement came out of these studies. Thus, Russo and Bruce (1996) concluded that SEF evoked saccades (like those evoked from the FEF) were all coded in an oculocentric frame of reference. In contrast, Tehovnik et al. (1998) described SEF evoked saccades as uniformly goal-directed. Schlag and Schlag-Rey (1987a) distinguished both kinds of saccades and described them as depending on the site of stimulation in SEF. Martinez-Trujillo et al. (2003b,c, 2004) found arguments in favor of these three views but finally adopted the last one.

Theoretically, there may be a simple way to establish whether an evoked saccade is programmed to reach an oculocentric or a non-oculocentric goal. If the eyes start moving when an oculocentric goal is presented or just before it is presented, the saccade to that goal, not being immediate, compensates for the detour effected by the ongoing eye movement. In human subjects, Hallett and Lightstone (1976) have described this compensation when goals are provided by natural visual stimuli (in a double-step paradigm). Compensation also occurs in a situation called "saccade collision" in which the test saccade – instead of being directed toward a visual target - is evoked by electrical stimulation within structures such as SC and FEF (Dassonville et al. 1992; Schlag et al. 1989, 1998; Schlag and Schlag-Rey 1990; Schlag-Rey et al. 1989). The principle of saccadic collision is graphically explained in Fig. 2A. As a control, trajectories of saccades evoked from a hypothetical "fixed-vector" site are shown in black. In red is shown the deviated trajectory of a saccade evoked from one of these sites just at the time when an up-right saccade (yellow) was starting. Clearly the detour caused by the initial saccade was compensated: the deviated electrically evoked saccade terminated where it would have ended anyway (i.e. tip of black arrow) if the eyes had not been moving at the time of stimulation.

Conversely, if the goal is non-oculocentric (as provided, for instance, by an auditory target), its position in space is fixed. Therefore, whether or not the eyes are moving when the goal is presented should not matter. This situation is illustrated in Fig. 2B. The converging trajectories of saccades evoked from a "goal-directed" site are schematized in black. If a saccade (in red) is evoked when an up-right saccade (in yellow) has just started, its trajectory will be the same as if no initial (yellow) saccade had interfered. Whereas the experimental situation is the same as in Fig. 2A, the outcome is completely different: no change is produced by collision.

In this Introduction, we have argued that distinguishing fixed-vector from goal-directed saccades by merely observing their trajectories is not an easy task. This is particularly true when the goal of evoked goal-directed saccades is very eccentric because then the convergence is not greater than it would be if the goal were oculocentric. The objective of the present study was to use the saccade collision test to distinguish the frame of reference used to code goal location. Applied to ambiguous cases, the test should reveal whether an initial saccade will change the trajectory of the evoked saccade (i.e. compensating as in Fig. 2A) or will not change it (as in Fig. 2B). A clear, systematic, change with collision will indicate oculocentric coding; a lack of change will indicate a non-oculocentric coding.

METHODS

Under Fluothane anesthesia, a monocular search coil was implanted in 4 female Rhesus monkeys according to the method designed by Judge et al. (1980). After a few weeks of training, a second surgery was performed to place a well on the midline above the area of the supplementary eye field. The monkeys sat in a primate chair with their head fixed, facing a tangent screen 61° horizontal by 50° vertical at a distance of 132 cm. Dim visual stimuli (dots of 0.25° diameter) produced by a Tektronix 608 oscilloscope were back-projected on the screen through a wide-angle lens. Each training and

experimental trial started by presenting a point of fixation. The animals were rewarded with apple juice for making a saccade toward a target that could appear at varying locations when the point of fixation was extinguished after a random time (up to 800 ms). Point of fixation and targets were positioned with joysticks. All experiments were run in a dim uniform red light, the frame of the screen being barely perceptible. The surgical procedures, training, and care of the monkeys followed the guidelines of the National Institute of Health's Guide for the Care and Use of Laboratory Animals, and the experimental protocol was approved by the UCLA Animal Research Committee.

In experimental sessions, tungsten microelectrodes were lowered in the frontal lobe through the dura until the first signs of neuronal activity were recorded. Electrical stimulation was then applied and tested every 250-300 μm (or more often if a single-unit was captured) to a depth of 2-2.5 mm. Trains of electrical stimuli consisting of 0.2 ms duration, 330 Hz, bipolar pulses, cathodal first, with a 100 μA maximum current (usually 40-65 μA) were delivered by a constant-current Haer stimulator. However, as accurate current readings with such stimulators are impossible when electrode impedance is high, the latter was continuously monitored on line by a Bak amplifier and this served to calculate the current of stimulation actually delivered. Usually, the electrode impedance started around 1-1.5 $\text{M}\Omega$ but usually would drop to 300-400 $\text{K}\Omega$. The current of stimulation was just supra-threshold to avoid stimulating too large a population of neurons and thus obtaining ambiguous results. In all cases, the train duration (150-240 ms) was adjusted to terminate after the end of the evoked saccade (by changing the number of pulses), to make sure that this saccade was not cut short because the stimulation did not last long enough.

We were particularly interested in exploring the SEF along its rostro-caudal axis. The main criterion to choose a stimulation site was to obtain reproducible results when stimulation was repeated with minimum supra-threshold current. A point of fixation was turned on for 200-400 ms randomly at one of 25 sites, 10° apart horizontally and vertically, forming a 40° -wide by 40° -high grid on the projection screen (see examples in

Fig. 1). Electrical stimulation was applied 200 ms after the point of fixation was turned off. Recorded saccade trajectories (1 KHz resolution) provided a saccade map characteristic of a given site in SEF. When, at some sites, no saccades could be evoked, the trials were repeated and, if still unsuccessful, the absence of response was marked by a dot on the map (e.g. see Fig. 1).

To estimate convergence, three measurements were made on the map of saccades collected at each site. The reason to have several measurements was that none of them is objectively perfect. Originally, it was our intention to use the method designed by Russo and Bruce (1993) and perfected by Klier et al. (2001) and Martinez-Trujillo et al. (2003a, b, and c, and 2004). This method ideally takes into account the angle of convergence and the amplitude of evoked saccades to calculate “convergence indices”. The amplitude is represented by the so-called “characteristic vector” for a given site, which is: “the theoretical gaze trajectory that would be evoked by stimulating the site when the animal is looking straight ahead” (Martinez-Trujillo et al. 2003b). So defined, the characteristic vector of a stimulated site is calculated from experimental data by multiple linear regression. Unfortunately, in some cases in our SEF study, no saccades could be evoked from the primary position (i.e.: straight ahead), as we noted in the Introduction. In other cases, we found the calculated characteristic vector to be quite different from the trajectory actually evoked from the primary position. Therefore, we had to rely on other measurements explained in Fig. 3 taking a particular SEF site as an example.. First, we measured the *maximal angle of convergence* using the two most converging trajectories equally distant from the goal (see Fig. 3A) This is not very different from the CI_d (convergence index based on saccade direction: Martinez-Trujillo 2003b). In practice, this angle, as we measured it, varied between 0° and 88°. Second, we determined the *goal eccentricity*. This is an estimate somewhat different from the CI_a (convergence index based on saccade amplitude: Martinez-Trujillo 2003b). As an estimate, it may be less rigorous but probably more significant than the CI_a. Assuming that trajectories converge to a point (i.e. "goal"), the distance of this point was measured in degrees from the center of the screen. As the start and end points of saccades, in coil recordings, are in fact sine

values, the scale in Fig. 3B is drawn to represent trajectories in sine values. We used straight lines to extrapolate the trajectories of saccades up to their hypothetical convergence point. However, as saccades are curved, using straight lines tends, in fact, to overestimate the eccentricities of goals reported in Figs. 7 and 12. Third, we calculated an *index of start-end convergence*. This was a new index, obtained by, first, measuring two surfaces: the area (in blue) circumscribing all the initial sites of evoked saccades and the area (in red) circumscribing all the termination sites of the same saccades and, second, by calculating the ratio between these two surfaces. This ratio = 1 if all saccades are parallel and of equal amplitude (i.e. fixed-vector), and it is >1 if saccades converge. It would be up to ∞ if all saccades terminated to a single point (which never happened). The values of the index of start-end convergence ranged from 0.92 (i.e. close to 1) to 11.25. The quantitative data presented in this report were obtained by systematic explorations in the last two monkeys.

In these 2 monkeys, we also systematically ran the *collision test*. This test, tried and perfected on the first two monkeys, consisted of stimulating during an initial saccade to a visual target. Examples of the application of this test are given in Figs. 8 and 9. The vector of the initial (natural) saccade had first to be selected. Ideally, for the greatest sensitivity of the test, this vector should be orthogonal to the general direction of electrically evoked saccades. However, to facilitate the measurements, we made it either vertical (as in Figs. 8 and 9) or horizontal whichever was closest to the orthogonal direction. The initial saccade went from a point of fixation to a target which was turned on for 50 ms, 200 ms after the point of fixation was turned off. Electrical stimulation was applied at varying intervals (0 up to 400 ms) after the eyes left the electronic window around the point of fixation.

Figure 4 explains the procedure used to quantify the results of the collision test. In this case, the initial saccade (yellow) was vertical; its amplitude was **a**. The vector of the saccade (red) evoked from the site where the target was to be located is shown as "Control". The vertical component of that evoked saccade was **c**. A train of stimuli

(bottom trace in left panel) was applied after a "Delay of stimulation" (from saccade onset to stimulation onset). In this condition of "Collision", the vector **b** of the evoked saccade (red) became longer than the control **c**, as if partly compensating for the displacement **a** of the eyes. The value of this compensatory effect for each stimulus delay is calculated as the difference between **b** and **c** divided by **a**, and multiplied by 100 to represent a percentage of deviation. In previous experiments on SC, the relation between the amount of compensation and the delay of stimulation has been shown to be exponential (Schlag et al. 1998). For the purpose of comparison, we took a mean percentage of deviation from the evoked saccades with less than 50 ms of delay of stimulation and used it as a single compensation index to characterize each SEF site.

RESULTS

Distribution of fixed-vector and goal-directed saccades in SEF

In all monkeys, stimulation of the SEF evoked saccades. Figure 5 shows our most complete topographical survey in one monkey (monkey H). At all sites, saccade latencies ranged from 43 to 100 ms except for 3 of the most caudal sites in this monkey (see Fig. 6). Figure 5 presents 23 trajectory maps of saccades and each of these maps is referred to its corresponding site of stimulation marked on the central grid by a circle. Small squares indicate sites where stimulation was ineffective. The insert at right is a view from the top, looking down on the surface of the monkey's frontal lobe, to locate the area explored. In these raw data, it is apparent that the trajectories of saccades evoked from the most caudal SEF sites were more convergent than the trajectories of saccades evoked from rostral sites. A similar rostro-caudal differentiation was noted in all monkeys, even though the surveys were much less extensive.

The results of the 3 tests of convergence described in the Methods were used in an attempt to verify objectively the apparent rostro-caudal distribution, seen in Fig. 5, in terms of evoked saccade types. These results (ordinates) for monkey H are plotted in Fig.

7 against the rostro-caudal level of sites of stimulation (abscissa). Clearly, for the most caudal sites, the angle of convergence was greater, the goal was less eccentric, and the ratio of initial-versus-terminal areas was larger. Correlations between the convergence indices were highly significant ($P < 0.001$ for all three pairs of comparisons). The lower two plots of Fig. 7 suggest the existence of 2 populations, which we have arbitrarily separated by drawing a vertical line between levels 5 and 6. For reference to the map, we have redrawn this line horizontally in Fig. 5.

Results of collision

The collision test was applied at all stimulation sites in monkeys H and I. It was also applied but not systematically in the other monkeys. Examples of collision are provided in Figs. 8 and 9. Figure 8 shows, from left to right, the saccades evoked from the same SEF site at various delays of stimulation (indicated in ms underneath) after the start of a visually guided downward saccade (see red arrow in top-right inset). Tick marks on each trace indicate stimulation onsets. The rightmost trace is the control vector. Clearly evoked saccades were elongated vertically "as if" to compensate for the downward trajectory accomplished by the initial saccade. When the initial saccade was horizontal (not shown) instead of vertical, it was the horizontal component of the electrically evoked saccade that was modified. This compensation was largest for short delays of stimulation and it diminished progressively with longer delays. The decrease in amplitude was exponential; its time course is graphically described in Fig. 10.

Completely different results were obtained when the same collision test was applied to apparently goal-directed saccades (see bottom-right inset in Fig. 9). As shown in this figure, , the vector of the evoked saccade did not change systematically in the same manner as in the case of fixed-vector saccades when stimulation was applied at varying intervals (indicated in ms) after the onset of a downward saccade. To make this point clear, 12 traces with varying delays of stimulation were superposed in the middle

panel of the lower row. Were these fixed-vector saccades, their directions would vary considerably. No compensation was seen in the cases of goal-directed saccades.

In Fig. 10, the time course of compensation (measured as a percentage of deviation, see Methods) is compared with the type of saccade evoked from different SEF sites. The first 2 examples (A and B) appear to be fixed-vector types. In both cases, the collision test caused compensation and the amount of this compensation (see Methods) declined exponentially as a function of the delay of stimulation (D and E). This exponential relationship is similar to that previously described for the FEF (Dassonville et al. 1989) and the SC (Schlag et al. 1998). The progressive decline has been explained by the fact that, the later the electrical stimulation is applied, the smaller is the remaining portion of the initial saccade that needs to be compensated. The second of these examples (B and E) is included in this figure especially because it was sometimes claimed that a significant difference between fixed-vector and goal-directed saccades is that the first are short while the second usually are long. But the evoked saccades in B were large (30°) and, obviously, this was not a relevant factor because compensation occurred anyway. By contrast, there was no compensation in the case of apparently goal-directed saccades (Fig. 10, C): the percentage of deviation was random (F) and, in this case, most of the points were negative (probably due to noise). This is the opposite of what would have happened if there had been any compensation.

As pointed out in the Introduction, there were SEF sites where evoked saccades did not fit perfectly well with the definitions of “fixed-vector” and “goal-directed”. It is obviously interesting to discover what effect, if any, collision would have on such atypical saccades. This is shown in Fig. 11 for the 2 cases of Fig.1 (A2 and B2). Compensation was clear in these 2 cases.

We found a good agreement between the measurements of mean percentage of compensation and the values of the indices of convergence. The relation between mean % of compensation and each of these three indices is illustrated in Fig. 12. Again, these

plots, like those of Fig. 6, suggest a segregation into 2 populations, although this is less evident for the measurements in terms of angle of convergence. All 3 statistical correlations (Spearman's rho) were significant at the level $P < 0.001$. In Fig. 13, we have reproduced the map of Fig. 5 but here the classification of SEF sites in two groups is based on a completely different criterion: the presence or absence of compensation in the collision test. Individual values of the index of compensation are indicated in the upper right corner for each site on the map. Comparison of these two maps (Figs. 5 and 13) suggests a remarkable agreement on the existence of two populations: fixed-vector sites rostrally, goal-directed caudally, with few discrepancies which, interestingly, happened to be concentrated in the intermediate zone.

DISCUSSION

One of the assumptions on which the present research was based is that so-called goal-directed saccades appear to converge toward a common goal because, indeed, they have a goal and that goal is not oculocentric (Dominey et al. 1997; Schlag and Schlag-Rey 1990). However, other interpretations for the observed convergence have to be considered.

One of the earliest interpretations invokes the possibility that electrically elicited saccades are artifactual, namely by not engaging a cerebellar mechanism that normally would contribute to the adjustment of saccade parameters, including saccade trajectory (Bruce 1990; Russo and Bruce 1993). Although this hypothesis appears reasonable, there is no proof that the cerebellum is not involved when saccades are electrically triggered. Furthermore, there is no evidence on the kind of distortion that would be expected if the cerebellum were not involved under these circumstances. The kinematics of electrically evoked saccades is undistinguishable from that of natural saccades (Martinez-Trujillo et al. 2003b).

Another hypothesis is that the eye movements evoked by SEF electrical stimulation in head-restrained monkeys do not represent the whole gaze shift but only its eye component, the head component being absent (e.g. Crawford and Guitton 1997). However, because it implies a separate control for the eye and head contributions to gaze shifts, this hypothesis is not quite compatible with the view “that the SEF encodes gaze and that the details of eye-head coordination during natural gaze shifts are specified downstream from this area” (Martinez-Trujillo et al. 2003b). Martinez-Trujillo et al. (2003c) found that, even with head free, a head-centered code appeared to provide the best explanation of their SEF stimulation data.

Finally, Thier and Andersen (1996) have proposed to explain the phenomenon of convergence of saccadic vectors as the result of a combined stimulation of adjacent fixed-vector and “centering” sites. Although such an explanation may apply to the parietal cortex, it is not likely for the SEF because we never found any site there characterized by evoked “centering” saccades (saccades directed toward the center of orbit). Neither is it likely that the appearance of saccade convergence could be due to the spread of stimulation current from a part of the SEF to another part. There is no obvious basis for such a hypothesis.

Rejecting these hypotheses, we think that the difference between our two categories of saccade vectors (converging and non-converging) is due to the inclusion of an eye position signal in the programming of the first type of saccades but not in the programming of the second type. Under the condition of collision however, the eye position signal is introduced in the computation of fixed-vector trajectories which, thus, become goal-directed. In contrast, the trajectory of goal-directed saccades is not altered because the programming of their trajectories already takes into account the orbital position of the eyes.

We wish to draw three essential conclusions from this research. The first is that saccade collision is a helpful addition to the battery of tests used to determine the frame

of reference for commanding eye movements. The second is that saccade commands are represented in different frames of reference in the SEF. The third is that SEF sites with different frames of reference appear clustered.

On the first point, one should note that the saccadic collision test utilizes a different principle from the tests of convergence (i.e. measuring the angle of convergence, the eccentricity of the goal, and the ratio between saccade departure area and termination area). Each one of the latter has its advantages as well as its limitations, but all three tests of convergence are attempts to quantify patterns of trajectories starting from different orbital positions. In contrast, the saccade collision test can theoretically provide a diagnosis requiring only performing two stimulations at each site (i.e. one control and one collision). The aim is to determine whether the trajectories will be the same or different in these situations? The saccade collision test may be particularly useful to recognize fixed-vector saccades when the vectors of these saccades are long and, therefore, give the false impression of converging.

Our second main conclusion (the multiplicity of frames of reference represented in the SEF) is in agreement with the last study of Martinez-Trujillo et al. (2004) with head-free monkeys. These authors have distinguished sites at which saccades are coded in one of three possible frames of reference: in order of relative frequency, head-centered, eye-centered, and body-or-space-centered sites. As the head of our monkeys was fixed, we could not differentiate the last two categories, head or body/space. However, in one incidental observation in which we fixed the head 30° toward one side, we could ascertain that the space coordinates of the goal were the same as with the head at 0°. This supports the conclusion of Martinez-Trujillo et al. (2004) that even a body/space coding exists in the SEF. It may be the reason why vestibular signals reach the SEF (Fukushima et al. 2004).

How could be explained the long, puzzling, disagreement in the literature, on the type of saccade coding in the SEF? Russo and Bruce (1993) did not find much

convergence in saccades evoked from the SEF. It seems that their stimulating sites were concentrated in the most anterior portion (located above the anterior half of the upper limb of the arcuate sulcus) of the area that we have studied here. We agree that it is within this anterior portion that fixed-vector sites are found. (For a comparison of SEF maps explored in different laboratories, see Sommer and Tehovnik 1999). Russo and Bruce (1993) also used rather short trains of stimuli (<70 ms), a procedure that could cut short goal-directed saccades before they reach their goal and, thus, make their vectors more similar to true fixed-vectors. That may explain why they did not find clearly goal-directed saccades evoked from the SEF. At the other extreme, Tehovnik and Lee (1993) reported that they had been unable to evoke fixed-vector saccades from the SEF. These authors produced the most complete map of the SEF in terms of saccade direction. From the zone corresponding to the most anterior portion of the area we have studied here, they produced the largest contraversive saccades. Even as fixed-vector, such saccades are the most curved (in a planar projection) and thus their true type may be hard to recognize. As we noted, the collision test is particularly useful in this case.

Our third conclusion is that saccade commands coded in different frames of reference could be separately represented in the SEF. We found the fixed-vector sites to be concentrated rostrally with respect to the goal-directed sites. Whenever tested with long stimulation, stair-case saccades were evoked only from this anterior region. This organization was consistent in 4 monkeys. As explained above, our observations also fit with the SEF maps of Tehovnik (1995), Tehovnik and Lee (1993), which locate the representation of the most eccentric saccades rostrally, and with the results of Russo and Bruce (1993) who observed saccades very similar to those that we could elicit from the most rostral part of the SEF. However, more explorations are needed to ascertain that the topographic organization we propose can be generalized. There may be more than two areas and their relative disposition may vary among monkey of the same species. Mann et al. (1988) have defended the view that the effects of stimulation in SEF are modifiable by experience. Nevertheless, our findings clearly establish that types of saccades elicited from the SEF can be represented in separate adjacent areas. It is known that the SEF

receives projections from the central superior lateral nucleus, a thalamic intralaminar nucleus, which contains eye position neurons (Schlag and Schlag-Rey 1984, see also Wyder et al. 2003). Such an input, signaling eye position, is required for a head-coding mechanism, but we found no evidence in the literature that it reaches only a particular sector of the SEF. The only reported difference that we found between anatomical connections of rostral and caudal SEF parts concerns projections to FEF (Schall et al. 1993), but it relates only to the size of evoked saccades.

In this study, we have considered only two frames of reference that we called oculocentric and non-oculocentric. However, there is good evidence that the SEF contains cells active with saccades oriented – not in an environmental direction (say: rightward) – but toward objects and sides of objects (say: to the right side of object; Olson and Gettner 1995). Does it mean that there is another frame of reference (object-centered) beside those that we considered? And, if so, what would electrical stimulation evoke in that case? We think that object-centered coding is in a different category. Object-centered coding concerns the stage of processing where a goal is designated already in one of two different languages: oculocentric or non-oculocentric. Imagine an object-centered mechanism that would be specialized for the right side of "things". If you are walking on campus while reading a book, such a mechanism can as well bring your gaze to the right of the page you are reading or to the right of a figure on that page (oculocentrically), or to the right side of the building you are facing (in non-oculocentric coordinates). You make saccades from words to words, sometimes jumping to a figure, checking the legend, and then returning to the text. Meanwhile, you try to follow a path, avoiding collision with obstacles. It seems that, for this complex behavior, it would be impractical to have all targets, the words, the figure, and the environment in a single frame of reference. In fact, you do not care where the page you read is located in your view of the environment or where the building to your right is located with respect to the page. It would be more useful to have two maps simultaneously available like separate windows on your computer screen, the page you read probably in oculocentric

coordinates, and the campus probably in supra-oculocentric coordinates (probably body/space coordinates since you are walking).

ACKNOWLEDGEMENT

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This work was supported by National Eye Institute Grant EY-05879.

REFERENCES

- Amador N, Schlag-Rey M, and Schlag J.** Primate antisaccade. II. Supplementary eye field neuronal activity predicts correct performance. *J Neurophysiol* 91: 1672-1689, 2004.
- Bon L and Luchetti C.** The dorsomedial frontal cortex of the macaca monkey: fixation and saccade-related activity. *Exp Brain Res* 89: 571-580, 1992.
- Bruce CJ.** Integration of sensory and motor signals in primate frontal eye fields. In: *From signal and sense: Local and global order in perceptual maps*, edited by Edelman GM, Gall WE, and Cowan CM. New York: Wiley-Liss, 1990.
- Chen LL and Wise SP.** Neuronal activity in the supplementary eye field during acquisition of conditional oculomotor associations. *J Neurophysiol* 73: 1101-1121, 1995.
- Craford JD and Guitton D.** Visual-motor transformations required for accurate and kinematically correct saccades. *J Neurophysiol* 78: 1447-1467, 1997.
- Dassonville P, Schlag J, and Schlag-Rey M.** Microstimulation of primate frontal eye field specifies retinotopic goal of evoked saccade. *Soc Neurosci Abstr* 19: 239, 1989.
- Dassonville P, Schlag J, and Schlag-Rey M.** The frontal eye field provides the goal of saccadic eye movements. *Exp Brain Res* 89: 300-310, 1992.
- Dominey PF, Schlag J, Schlag-Rey M, and Arbib M.** Colliding saccades evoked by frontal eye field stimulation: artifact or evidence for an oculomotor compensatory mechanism underlying double-step saccades? *Biol Cybern* 76: 41-52, 1997.
- Fujii N, Mushiake H, Tamai M, and Tanji J.** Microstimulation of the supplementary eye field during saccade preparation. *NeuroReport* 6: 2565-2568, 1995.
- Fukushima J, Akao T, Takeichi N, Kurkin S, Kaneko CR, and Fukushima K.** Pursuit-related neurons in the supplementary eye fields: Discharge during pursuit and passive whole body rotation. *J Neurophysiol* 91: 2809-2825, 2004.
- Hallett PE and Lightstone AD.** Saccadic eye movements towards stimuli triggered by prior saccades. *Vision Res* 16: 99-106, 1976.
- Huerta MF and Kaas J.** Supplementary eye field as defined by intracortical microstimulation: connections in macaques. *J Comp Neurol* 293: 299-330, 1990.

Isoda M and Tanji J. Cellular activity in the supplementary eye field during sequential performance of multiple saccades. *J Neurophysiol* 88: 3541-3545, 2002.

Judge SJ, Richmond BJ, and Chu FC. Implantation of magnetic search coils for measurement of eye position: an improved method. *Vision Res* 20: 535-538, 1980.

Kim YJ, Badler JB, and Heinen SJ. Trajectory interpretation by supplementary eye field neurons during ocular baseball. *J Neurophysiol* 94: 1385-1391, 2005.

Klier EM, Wang H, and Crawford JD. The superior colliculus encodes gaze commands in retinal coordinates. *Nature Neurosci* 4: 627-632, 2001.

Lu X, Matsuzawa M, and Hikosaka O. A neural correlate of oculomotor sequences in supplementary eye field. *Neuron* 34: 317-325, 2002.

Mann SA, Tau R, and Schiller PH. Conditional task-related responses in monkey dorsomedial frontal cortex. *Exp Brain Res* 69: 460-468, 1988.

Martinez-Trujillo JC, Klier EM, Wang H, and Crawford JD. Contribution of head movement to gaze command coding in monkey frontal cortex and superior colliculus. *J Neurophysiol* 90: 2770-2776, 2003b.

Martinez-Trujillo JC, Medendorp WR, Wang H, and Crawford JD. Frames of reference for eye-head gaze commands in primate supplementary eye fields. *Neuron* 44: 1057-1066, 2004.

Martinez-Trujillo JC, Wang H, and Crawford JD. Electrical stimulation of the supplementary eye field in the head-free macaque evokes kinematically normal gaze shifts. *J Neurophysiol* 89: 2961-2974, 2003a.

Martinez-Trujillo JC, Wang H, and Crawford JD. Macaque supplementary eye field encodes gaze in head centered coordinates. *Soc Neurosci Abstr* 555.2, 2003c.

Mushiaki H, Fujii N, and Tanji J. Visually guided saccade versus eye-hand reach: Contrasting neuronal activity in the cortical supplementary and frontal eye fields. *J Neurophysiol* 75: 2187-2191, 1996.

Olson CR and Gettner SN. Object-centered direction selectivity in the macaque supplementary eye field. *Science* 269: 985-989, 1995.

Robinson DA. Eye movements evoked by collicular stimulation in the alert monkey. *Vision Res* 12: 1795-1808, 1972.

Robinson DA and Fuchs AF Eye movements evoked by stimulation of the frontal eye fields. *J Neurophysiol* 32: 637-648, 1969.

Russo GS and Bruce CJ. Effect of eye position within the orbit on electrically elicited eye movements: A comparison of the macaque monkey frontal and supplementary eye fields. *J Neurophysiol* 69: 800-818, 1993.

Russo GS and Bruce CJ. Neurons in the supplementary eye field of Rhesus monkeys code visual targets and saccadic eye movements in an oculocentric coordinate system. *J Neurophysiol* 76: 825-848, 1996.

Schall JD. Neuronal activity related to visually guided saccadic eye movements in the supplementary motor area of Rhesus monkeys. *J Neurophysiol* 66: 530-558, 1991a.

Schall JD. Neuronal activity related to visually guided saccades in the frontal eye fields of Rhesus monkeys. Comparison with supplementary eye fields. *J Neurophysiol* 66: 559-579, 1991b.

Schall JD, Morel A and Kaas JH. Topography of supplementary eye field afferents to frontal eye field in macaque: Implications for mapping between saccade coordinate systems. *Vis. Neurosci.* 10: 385-393, 1993.

Schlag J, Pouget A, Sadeghpour S and Schlag-Rey M. Interactions between natural and electrically evoked saccades. III. Is the nonstationarity the result of an integrator not instantaneously reset? *J Neurophysiol* 79: 903-910, 1998.

Schlag J and Schlag-Rey M. Visuomotor functions of central thalamus in monkey. I. Unit activity related to spontaneous eye movements. *J Neurophysiol* 51: 1149-1174, 1984.

Schlag J and Schlag-Rey M. Evidence for a supplementary eye field. *J Neurophysiol* 57: 179-200, 1987a.

Schlag J and Schlag-Rey M. Does microstimulation evoke fixed-vector saccades by generating their vector or by specifying their goal? *Exp Brain Res* 68: 442-444, 1987b.

Schlag J and Schlag-Rey M. Colliding saccades may reveal the secret of their marching orders. *Trends Neurosci.* 13: 410-415, 1990.

Schlag J, Schlag-Rey M, and Dassonville P. Interactions between natural and electrically evoked saccades. II. At what time is eye position sampled as a reference for the localization of a target. *Exp Brain Res.* 76: 548-558, 1989.

- Schlag-Rey M, Amador N, Sanchez H, and Schlag J.** Antisaccade performance predicted by neuronal activity in the supplementary eye field. *Nature* 390: 398-401, 1997.
- Schlag-Rey M, Schlag J, and Dassonville P.** How the frontal eye field can impose a saccade goal on superior colliculus neurons. *J Neurophysiol* 67: 1003-1005, 1992.
- Schlag-Rey M, Schlag J, and Shook B.** Interactions between natural and electrically evoked saccades. I. Differences between sites carrying retinal error and motor error signals in monkey superior colliculus. *Exp Brain Res*. 76: 537-547, 1989.
- Sommer M and Tehovnik EJ.** Reversible inactivation of macaque dorsomedial frontal cortex: effects on saccades and fixation. *Exp Brain Res* 124: 429-446, 1999.
- Tehovnik EJ.** The dorsomedial frontal cortex: eye and forelimb fields. *Behav Brain Res* 67: 147-163, 1995.
- Tehovnik EJ and Lee K.** The dorsomedial frontal cortex of the rhesus monkey: topographic representation of saccades evoked by electrical stimulation. *Exp Brain Res* 96:430-442, 1993.
- Tehovnik EJ, Slocum WM, Tolias AS, and Schiller PH.** Saccades induced electrically from the dorsomedial frontal cortex: evidence for a head-centered representation. *Brain Res* 795: 287-291, 1998.
- Thier P and Andersen RA.** Electrical microstimulation suggest two different forms of representation of head-centered space in the intraparietal sulcus of rhesus monkeys. *Proc Natl Acad Sci* 93: 4962-4967, 1996.
- Tian JR and Lynch JC.** Slow and saccadic eye movements evoked by microstimulation in the supplementary eye field of the Cebus monkey. *J Neurophysiol* 74: 2204-2210, 1995.
- Wyder MT, Massoglia DP and Stanford TR.** Quantitative assessment of the timing and tuning of visually-related, saccade-related, and delay period activity in primate central thalamus. *J Neurophysiol* 90: 2029-2052, 2003.

FIGURE LEGENDS

Figure 1. Types of saccades electrically evoked from SEF sites. Saccade trajectories as plotted on a flat screen (scale in degrees). Dots mark eye positions where stimulation did not evoke movements. A1: Typical fixed-vector type. A2: Atypical fixed-vector type. B1: Typical goal-directed type. B2: Atypical goal-directed type. Note that, in all figures presenting actual records, evoked saccades are all directed rightwards.

Figure 2. Schematic illustration of the effect of saccade collision on fixed-vector saccades (A) and on goal-directed saccades (B). Control saccade vectors starting from different eye positions are shown by black arrows. Collision is obtained by stimulation (onset marked by red tick) during a visually-guided saccade (yellow). In this example, the visually-guided saccade was straight up. The stimulation produces the saccade vector shown by a red arrow. In the case of fixed-vector saccades (A), our hypothesis is that the vector of the electrically evoked saccade (red) will be different from the vector of the control saccade (black). If the stimulation is given early enough, the electrically evoked saccade will terminate close to the point of termination of the control saccade starting at the origin of the visually-guided saccade (i.e. this would be perfect compensation). As stimulation is given later and later, the deviation of the electrically evoked saccade wanes off progressively and disappears. In the case of goal-directed saccades (B), our hypothesis predicts no deviation at all (i.e. the red vector is the same as the black vector).

Figure 3. Three measurements of convergence using the set of goal-directed saccades evoked from the same site. A: Angle of convergence. This is the angle made by 2 straight lines (red) extrapolating the 2 most separated evoked saccades down to their meeting point. B: Goal eccentricity. Using the same straight lines as in A (red), the distance is measured between the point of convergence and the primary position of the eye. As this distance is often greater than 30° , the eccentricity must be measured on a scale in sines of angles. C: Index of start-end convergence. All the starting points of saccades are circumscribed (blue area); all the ending points of the same saccades are similarly

circumscribed (red area). The index is the calculated blue/red ratio of the encompassed areas.

Figure 4. Graphic definition of the parameters considered in the saccade collision test. Visually-guided saccade in yellow, stimulation-evoked saccade in red. At left, schematic time representation of saccade collision. The delay of stimulation is measured from the start of the visually-guided saccade to the start of the stimulation train. At right, uni-dimensional (vertical, in this case) measurements of saccade vectors. a = visually-guided saccade; b = stimulation-evoked saccade; c = control saccade from same initial position. Onset of stimulation indicated by tick mark in collision record. The % of deviation, calculated as shown, varies with the delay of stimulation.

Figure 5. Topographic distribution of stimulation results in SEF of monkey H. At the center, half-mm grid with all the sites of stimulation. The location of this grid is indicated in red on the top view of the frontal lobe (inset). Numbers at left of grid correspond to AP-levels in Fig. 6, numbers at top of grid are distances in mm from midline. Sites of stimulation are coded as follows: blue circles = fixed-vector; red circles = goal-directed; orange circles = doubtful or undetermined (see above and Fig. 1); black squares = no eye movement.

Figure 6. Latencies of evoked saccades illustrated in Fig. 5, as a function of anterior-posterior levels. All latencies ranged from 43 to 100ms, except for the most posterior sites: H0630 (199 ms), H0620 (156 ms), and H0711 (144 ms).

Figure 7. Evaluation of saccade convergence as a function of anterior-posterior levels of the sites of stimulation. Results of the 3 tests of convergence plotted against AP levels. Some points at 90°, having the same coordinates, are superposed in the middle diagram. The vertical dash line (between levels 5 and 6) corresponds to the dash line on the grid in Fig. 5. Horizontal dash lines are best estimates to separate data into 2 populations.

Figure 8. Actual records of saccade collision at a presumed fixed-vector site, with different delays of stimulation (indicated below each trace). Stimulation onset at tick mark. Two ms between successive dots. Note the progressive decrease of compensation as the delay of stimulation increased. Upright inset: map of all the controls for this SEF site, indicating the initial eye position (in screen coordinates) at which the collision test was performed in this case.

Figure 9. Actual records of saccade collision at a presumed goal-directed site, with different delays of stimulation (indicated for each diagram). Same details of presentation as in Fig. 8. In the middle diagram of the lower row, traces are superposed to visualize their similarity.

Figure 10. Comparison of the time course of the % of deviation for 3 sites of stimulation where the collision test was performed. A-C: maps of control vectors. A = short fixed-vector; B = long fixed-vector; C = goal-directed. D-F: corresponding plots of % of deviation as a function of delay of stimulation. Vertical bars: ranges of dispersion of component amplitude of controls (i.e. % of deviation), derived from the equation explained in Fig. 4, using the mean amplitude of initial saccade in collision trials as the value of a . D time constant = 55 ms. E time constant = 119 ms. Note that in F there was no evidence of compensation.

Figure 11. Time course of the % of deviation for the 2 sites of stimulation A2 and B2 in Fig. 1, which produced saccades with atypical properties. Same presentation as lower panels in Fig. 10.

Figure 12. Relationship between indexes of convergence and percentage of compensation in saccade collision for all sites tested in monkey H.

Figure 13. Revisiting the data in Fig. 5 on the basis of collision results. Same presentation as in Fig. 5. The coding of SEF stimulation sites (i.e. color of circles) is now based on the results of the saccade collision test.

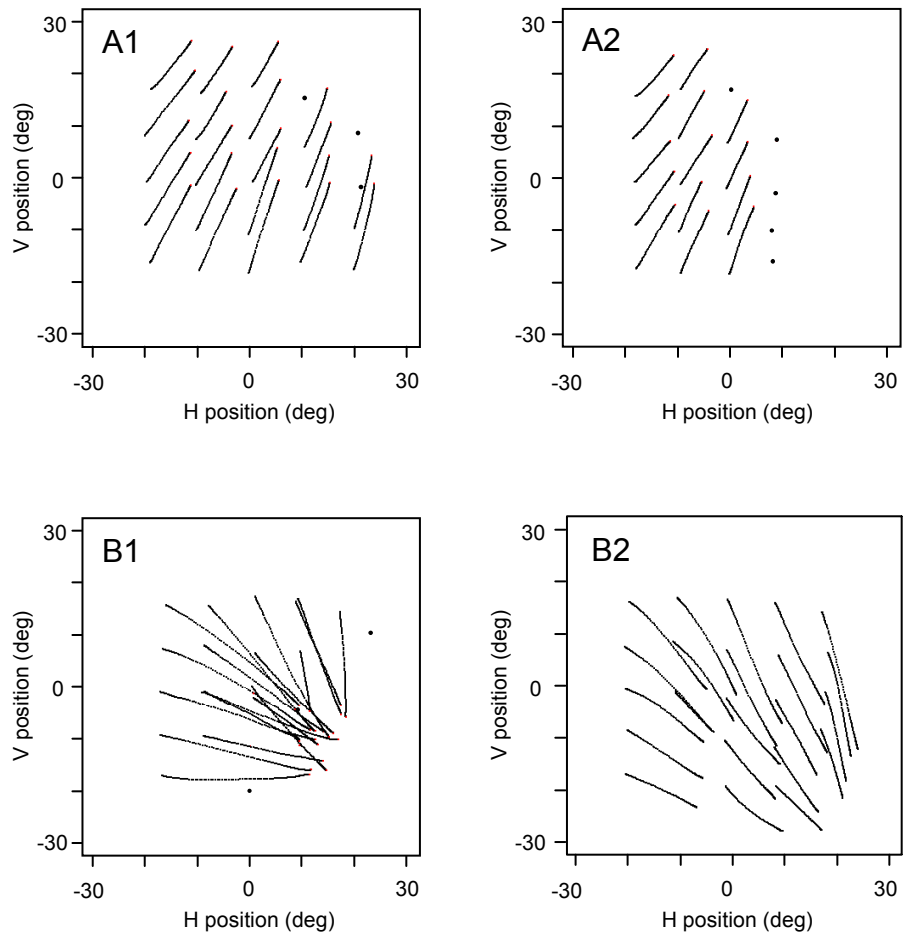


Figure 1

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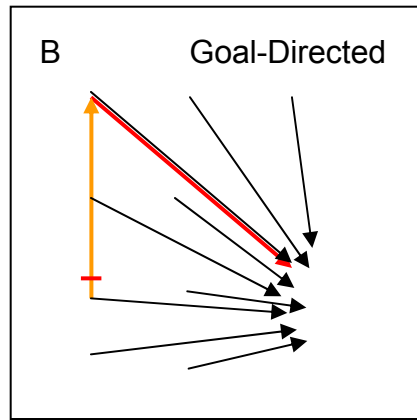
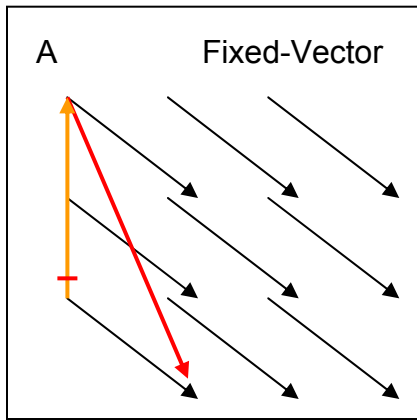


Figure 2

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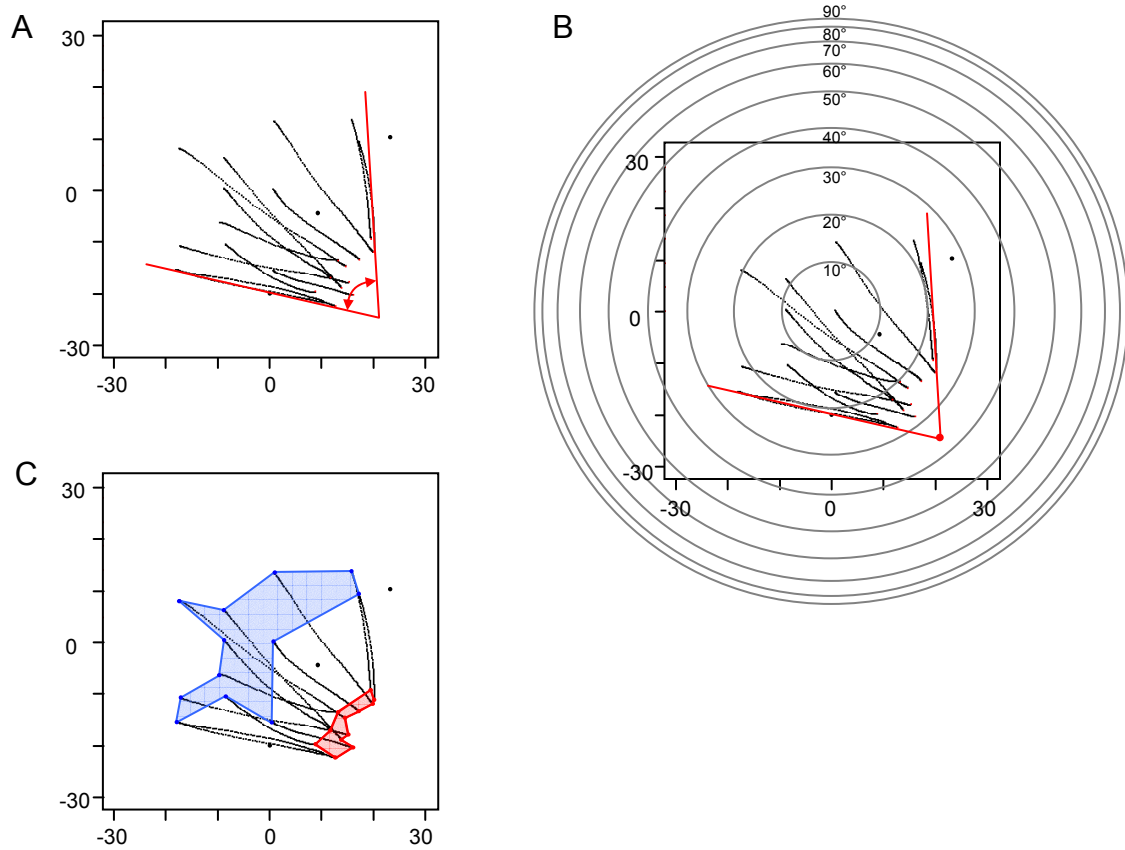


Figure 3

Park, Schlag-Rey, Schlag

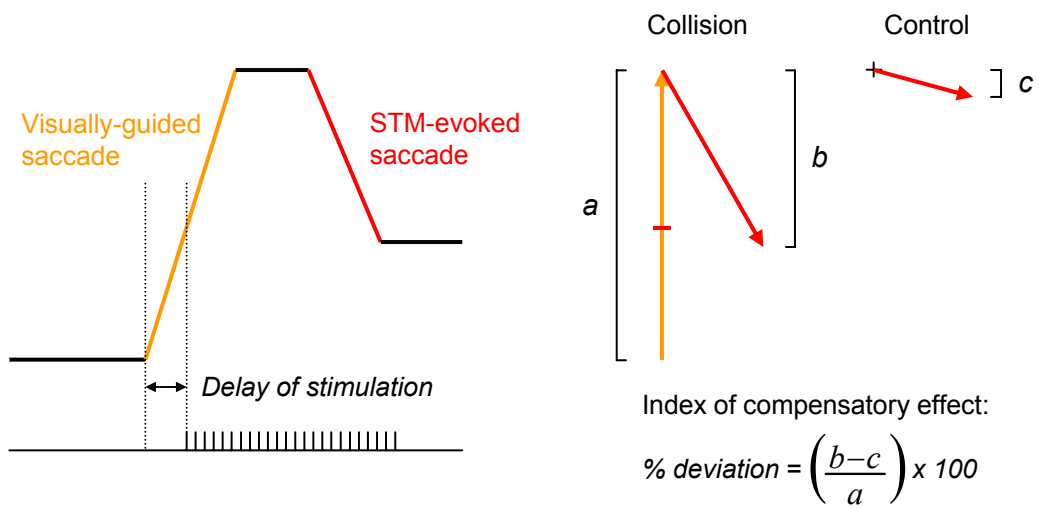


Figure 4

Park, Schlag-Rey, Schlag

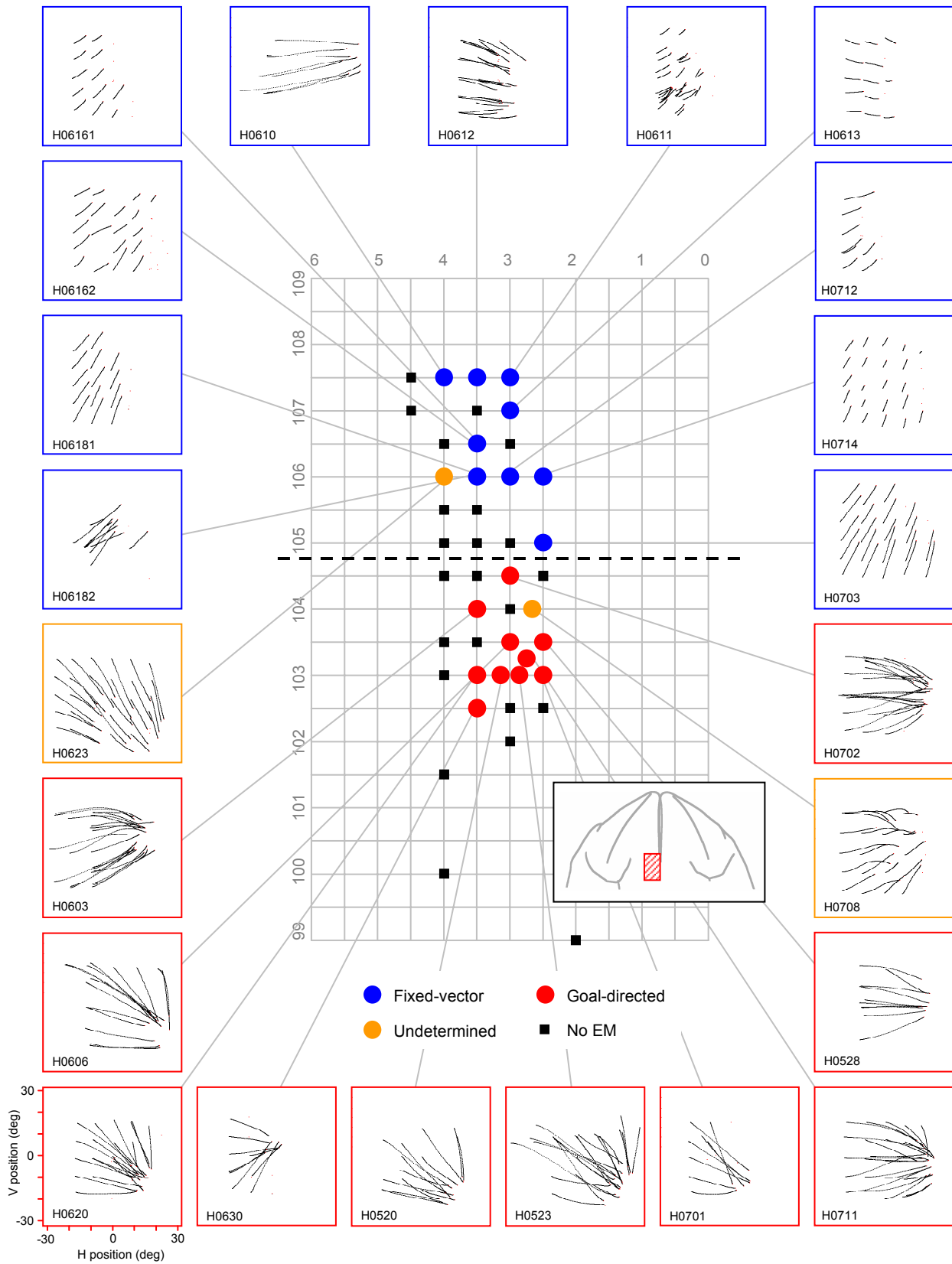


Figure 5

Park, Schlag-Rey, Schlag

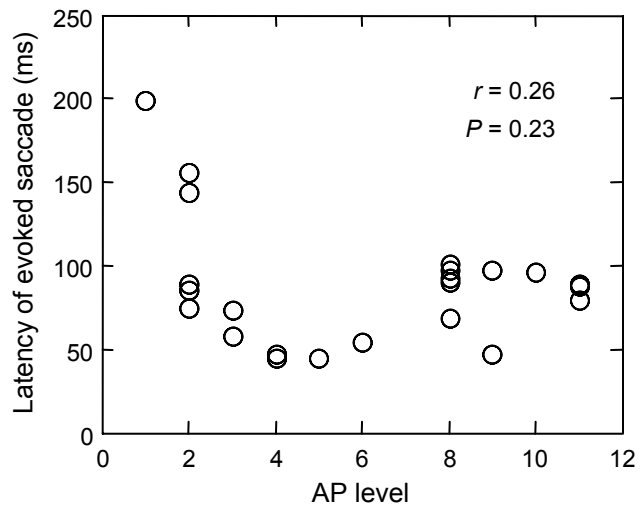


Figure 6

Park, Schlag-Rey, Schlag

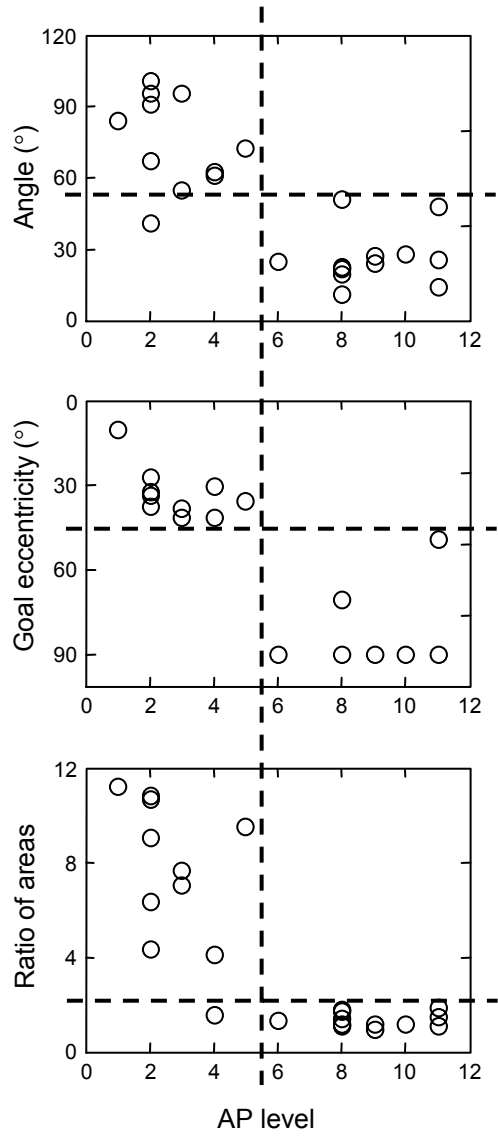


Figure 7

Park, Schlag-Rey, Schlag

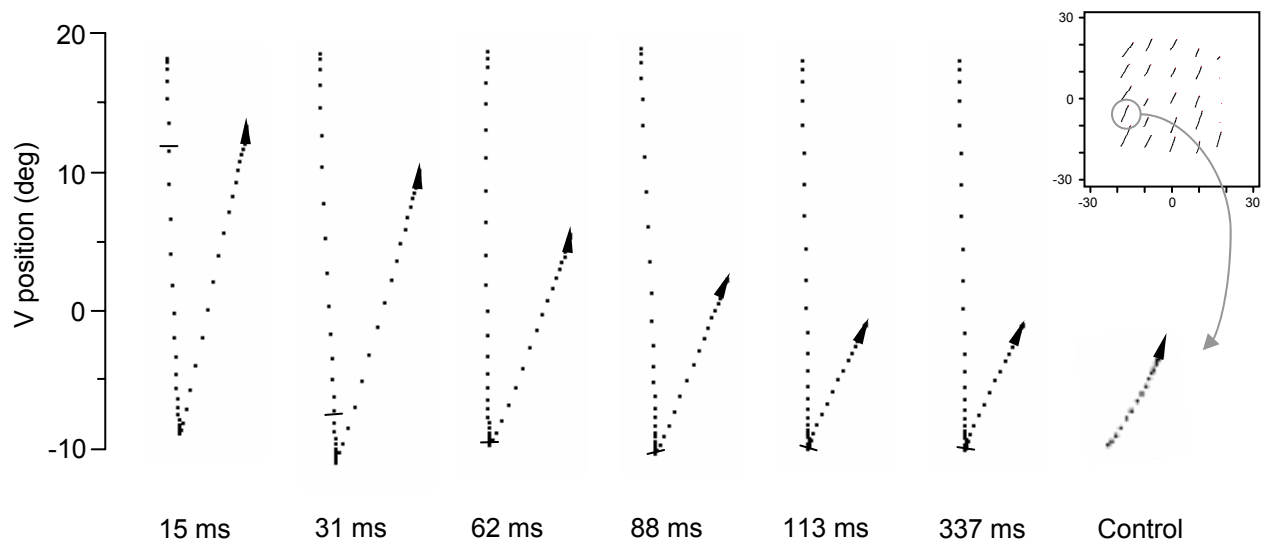


Figure 8

Park, Schlag-Rey, Schlag

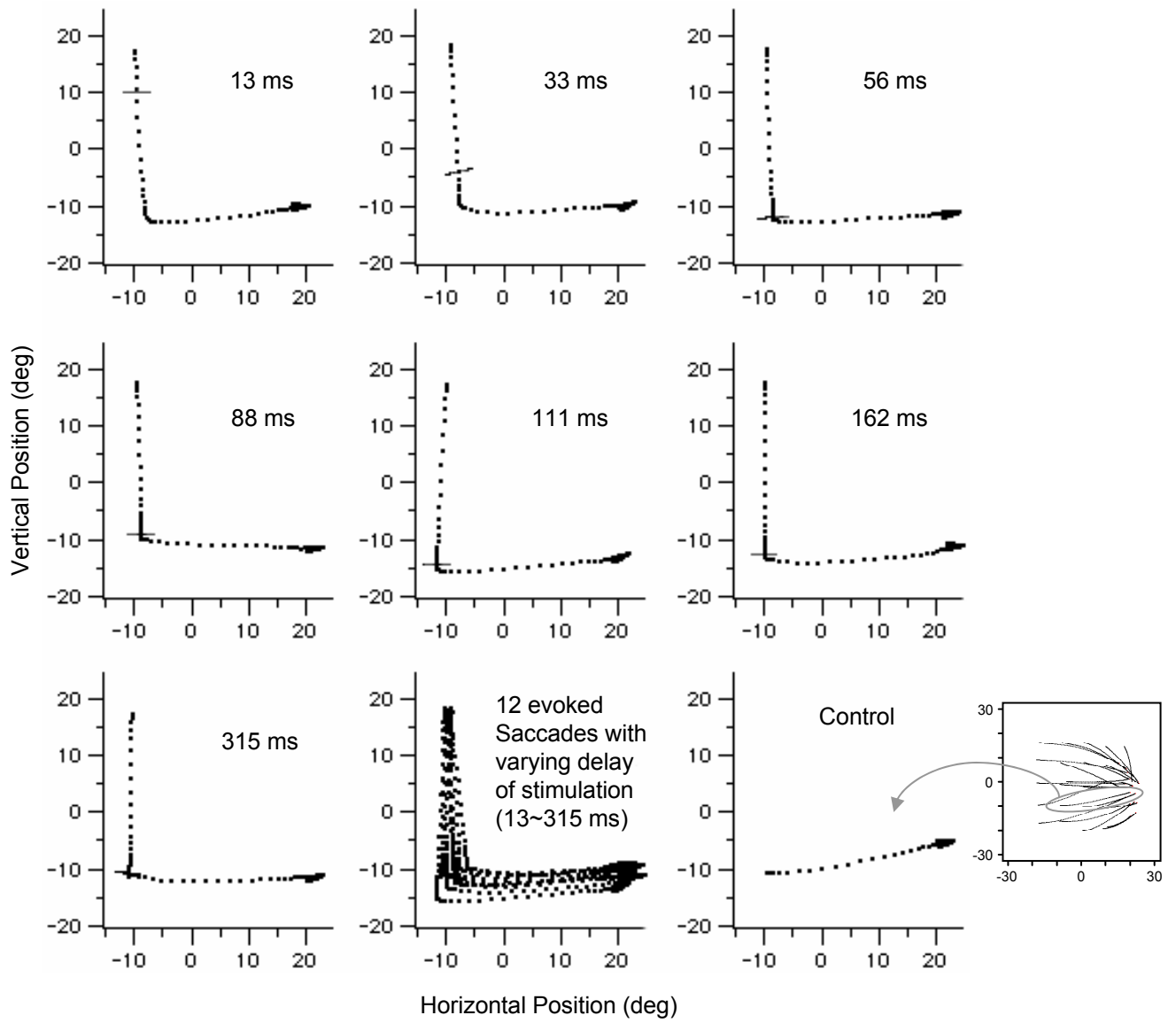


Figure 9

Park, Schlag-Rey, Schlag

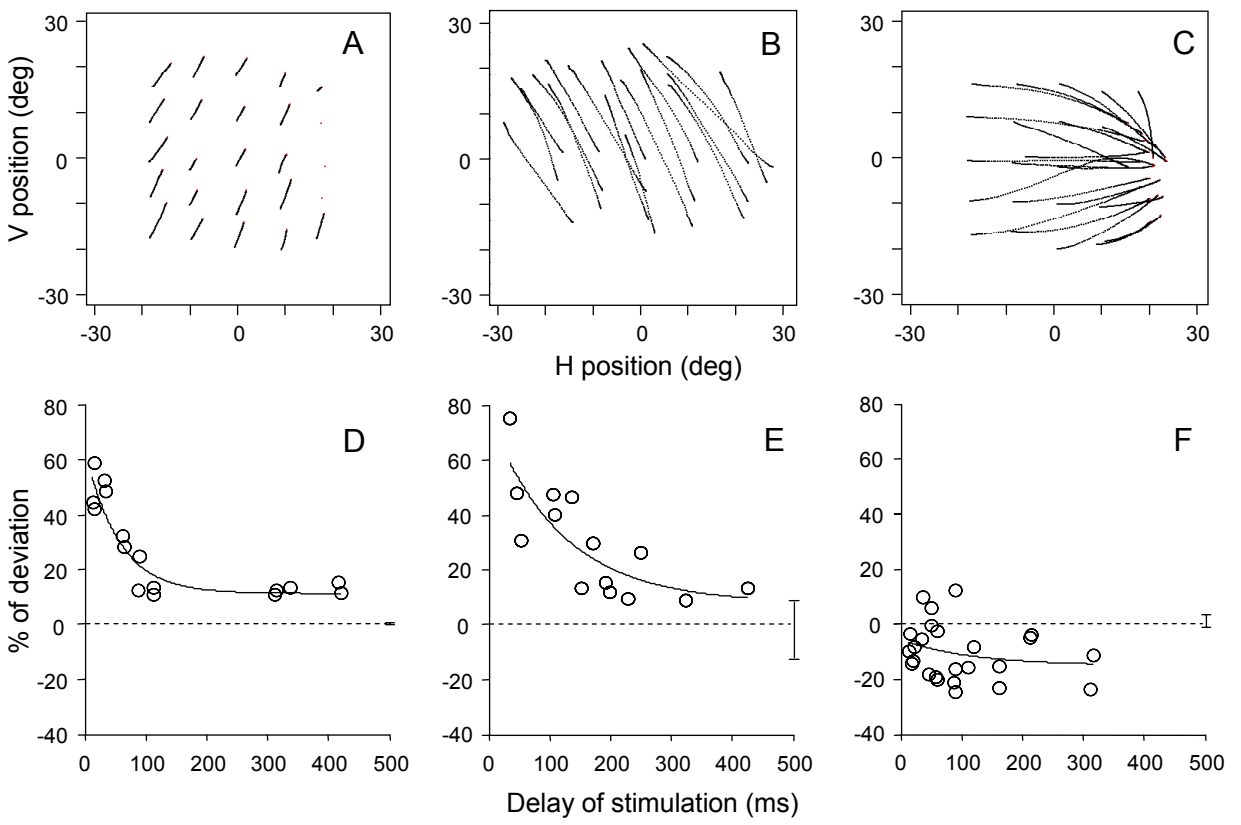


Figure 10

Park, Schlag-Rey, Schlag

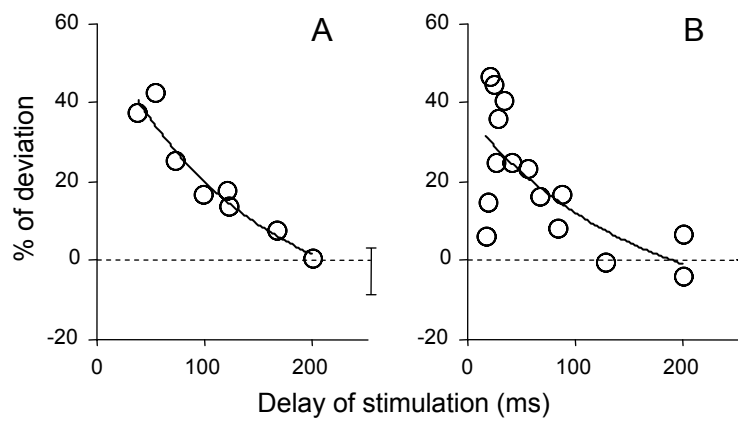


Figure 11

Park, Schlag-Rey, Schlag

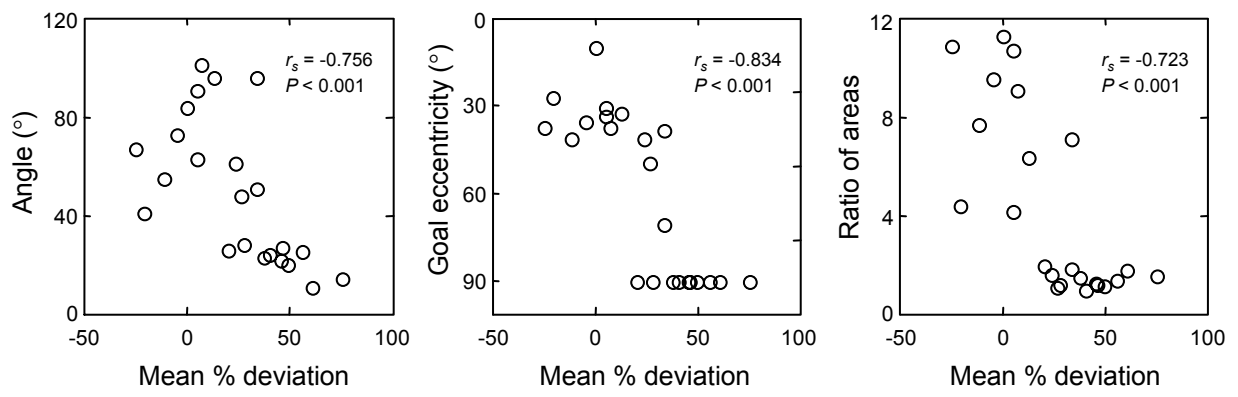


Figure 12

Park, Schlag-Rey, Schlag

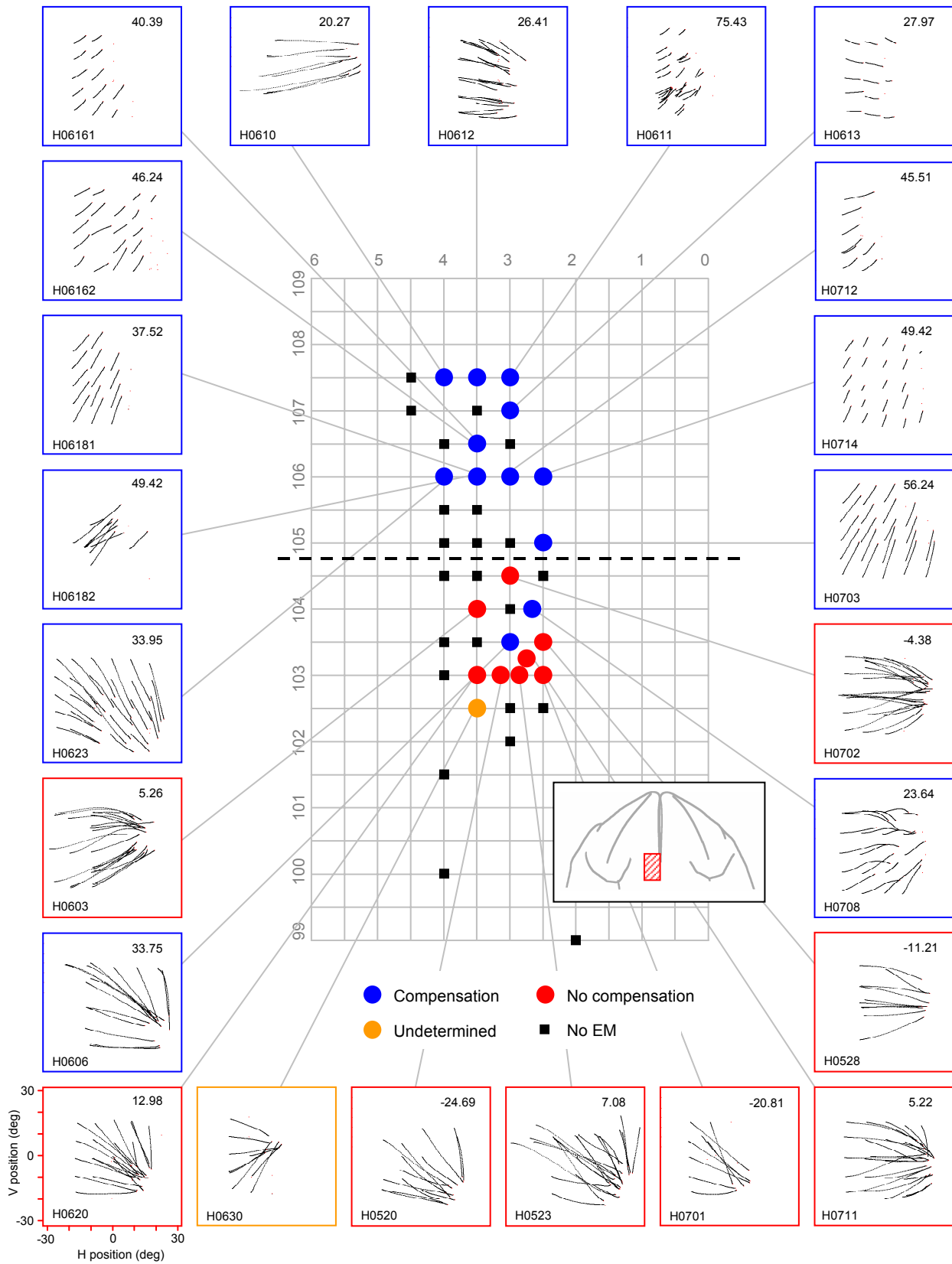


Figure 13

Park, Schlag-Rey, Schlag