

Influence of Stimulus Eccentricity and Direction on Characteristics of Pro- and Antisaccades in Non-Human Primates

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Bell, A. H., S. Everling, and D. P. Munoz. Influence of stimulus eccentricity and direction on characteristics of pro- and antisaccades in non-human primates. *J Neurophysiol* 84: 2595–2604, 2000. The ability to inhibit reflexes in favor of goal-oriented behaviors is critical for optimal exploration and interaction with our environment. The antisaccade task can be used to investigate the ability of subjects to suppress a reflexive saccade (prosaccade) to a suddenly appearing visual stimulus and instead generate a voluntary saccade (antisaccade) to its mirror location. To understand the neural mechanisms required to perform this task, our lab has developed a non-human primate model. Two monkeys were trained on a task with randomly interleaved pro- and antisaccade trials, with the color of the central fixation point (FP) instructing the monkey to either make a prosaccade (red FP) or an antisaccade (green FP). In half of the trials, the FP disappeared 200 ms before stimulus presentation (gap condition) and in the remaining trials, the FP remained visible (overlap condition) during stimulus presentation. The effect of stimulus eccentricity and direction was examined by presenting the stimulus at one of eight different radial directions (0–360°) and five eccentricities (2, 4, 8, 10, and 16°). Antisaccades had longer saccadic reaction times (SRTs), more dysmetria, and lower peak velocities than prosaccades. Direction errors in the antisaccade task were more prevalent in the gap condition. The difference in mean SRT between correct pro- and antisaccades, the anti-effect, was greater in the overlap condition. The difference in mean SRT between the overlap and the gap condition, the gap effect, was larger for antisaccades than for prosaccades. The manipulation of stimulus eccentricity and direction influenced SRT and the proportion of direction errors. These results are comparable to human studies, supporting the use of this animal model for investigating the neural mechanisms subserving the generation of antisaccades.

INTRODUCTION

Saccades are rapid eye movements that serve to realign the fovea onto a stimulus of interest. We often make saccades to novel visual stimuli that suddenly appear or move. However, it is sometimes necessary to suppress this visual grasp reflex (Hess et al. 1946) and instead generate a saccade to alternative location. For example, when driving we must be able to selectively ignore extraneous sights and sounds while maintaining concentration on the road. This ability to selectively suppress saccade reflexes in favor of goal-oriented behaviors is necessary for optimally interacting with our environment. A task that probes this ability is the antisaccade task, first introduced by Hallett (1978), which requires the suppression of a reflexive

saccade (prosaccade) to a suddenly appearing visual stimulus and the generation of a voluntary saccade (antisaccade) to the diametrically opposite position. The importance of the antisaccade task as a clinical tool was first demonstrated by Guitton and colleagues (1985), who showed that patients with frontal lobe damage often had difficulty suppressing reflexive prosaccades, a function essential to performing the antisaccade task. Subsequently the antisaccade task has been used to examine many neurological and psychiatric disorders (see Everling and Fischer 1998 for review).

To understand the neural mechanisms responsible for the suppression of reflexive saccades and generation of voluntary saccades in the antisaccade task requires an animal model. Our lab (Everling and Munoz 2000; Everling et al. 1998a, 1999) and others (Amador et al. 1998; Funahashi et al. 1993; Gottlieb and Goldberg 1999; Schlag-Rey et al. 1997) have developed a non-human primate model that permits recording neural activity from structures known to be involved in the initiation and suppression of saccades while awake, behaving monkeys perform the task. While previous studies have reported neural activities in the task, there has been little systematic study of the behavioral characteristics of antisaccades in non-human primates. More importantly, there has been limited comparison between monkey and human behavioral performance data, an important step to validate these models. Such a step is essential to allow for the translation of animal findings to human studies.

The purpose of the present study is to investigate the influence of stimulus direction and eccentricity on the characteristics of pro- and antisaccades in the non-human primate. The animals were therefore trained to generate both pro- and antisaccades in response to stimuli presented at one of eight different radial directions (0–360°) and five different eccentricities (2–16°). A further goal of this study is to examine the effect of fixation state on saccade characteristics and task performance. Thus the task was performed in both the gap and overlap conditions (Saslow 1967).

The data reveal that non-human primates trained on a task with randomly interleaved pro- and antisaccade trials generate saccades with similar characteristics to those seen in humans. Preliminary reports of these data have been presented elsewhere (Bell et al. 1999).

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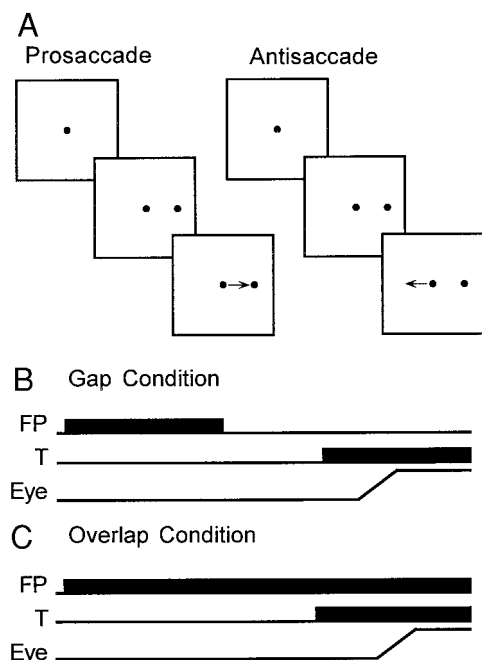


FIG. 1. Schematic representation of the pro- and antisaccade task. See METHODS for details. A: monkeys were required to generate either a saccade to a peripheral stimulus (prosaccade) or to its diametrically opposite location (antisaccade) depending on the color of the initial fixation point (red indicating a prosaccade, green indicating an antisaccade). The color of the peripheral stimulus was always red. The paradigm featured both the gap condition (fixation point is extinguished for a period of 200 ms prior to peripheral stimulus presentation (B) and the overlap condition (fixation point remains illuminated for the duration of the trial; C).

METHODS

Two adult male rhesus monkeys (*Macaca mulatta*, weighing 7 and 10 kg) were used in this study. All procedures were approved by the Queen's University Animal Care Committee and were in accordance with the Canadian Council on Animal Care policy on the use of laboratory animals. Animals were prepared for chronic behavioral experiments in a single surgical session. Eye movements were monitored using the magnetic search coil technique (Fuchs and Robinson 1966; Judge et al. 1980). Scleral search coils and a head-restraint device were implanted under ketamine/isoflurane anesthetic in aseptic conditions (see Munoz and Istvan 1998 for details).

During training and experiments, the animals were seated in a primate chair (Crist Instruments) positioned 86 cm from a tangent screen spanning $\pm 35^\circ$ of the visual field in a dark, sound-attenuated room. Control of the behavioral paradigms and storage of eye position data were done using a 486 personal computer running a real-time data-acquisition software package (REX) (Hays et al. 1982). Horizontal and vertical eye positions were sampled at 500 Hz from one eye. Visual stimuli were generated by light-emitting diodes (LEDs, red and green, 0.3 cd/m²), which were back-projected onto the tangent screen and were positioned by reflecting the LEDs off two mirrors mounted on galvanometers oriented in orthogonal planes. The mirrors were located the same distance as the animal from the screen to correct for any tangent errors.

Behavioral paradigm

The monkeys were trained (see *Training protocol*) to perform a paradigm with randomly interleaved pro- and antisaccade trials (Fig. 1A). Between trials, the screen was diffusely lit (1.0 cd/m²) to prevent dark adaptation. The onset of each trial was signaled by the removal

of the background light and illumination of a central fixation point (FP). The FP was presented for a period of 700–900 ms and was either red, signifying a prosaccade trial, or green, signifying an antisaccade trial. The FP was presented in two randomly interleaved conditions: on half of the trials, the FP was extinguished 200 ms prior to the appearance of the eccentric stimulus (gap condition; Fig. 1B) and in the remaining half of the trials, the FP remained visible for the duration of the trial (overlap condition; Fig. 1C). In each block of trials, the eccentric red visual stimulus appeared pseudorandomly at one of two diametrically opposite locations for 600 ms. Stimuli were presented in blocks covering eight different radial directions (0–360°) and five different eccentricities (2, 4, 8, 10, and 16°). Monkeys were given a liquid reward if they maintained central fixation for the duration of the fixation (700–900 ms) and gap period (200 ms), if applicable, and generated a saccade in the proper direction (toward the stimulus in a prosaccade trial and in the opposite direction in an antisaccade trial) within 500 ms of eccentric stimulus appearance, and then maintained fixation there for ≥ 200 ms. In antisaccade trials only, a green stimulus was presented immediately after the saccade at the mirror location of the red stimulus. Animals performed the task until fully satiated at which point they were returned to their cages.

Training protocol

The monkeys were trained, initially, to fixate a central red LED for periods of $\leq 2,000$ ms and perform a simple saccade task consisting of a central red FP and a red stimulus of varying direction and eccentricity that was presented in both the gap and overlap conditions. The monkeys were required to generate a foveating saccade within 500 ms of stimulus presentation to obtain a liquid reward. The on-line accuracy window was kept relatively large (± 5 – 10° in any direction from the intended end point) at the start of the training and gradually decreased to $\pm 3^\circ$ eccentricity around the central FP and eccentric stimuli. Once the monkeys performed this task correctly on 90% of

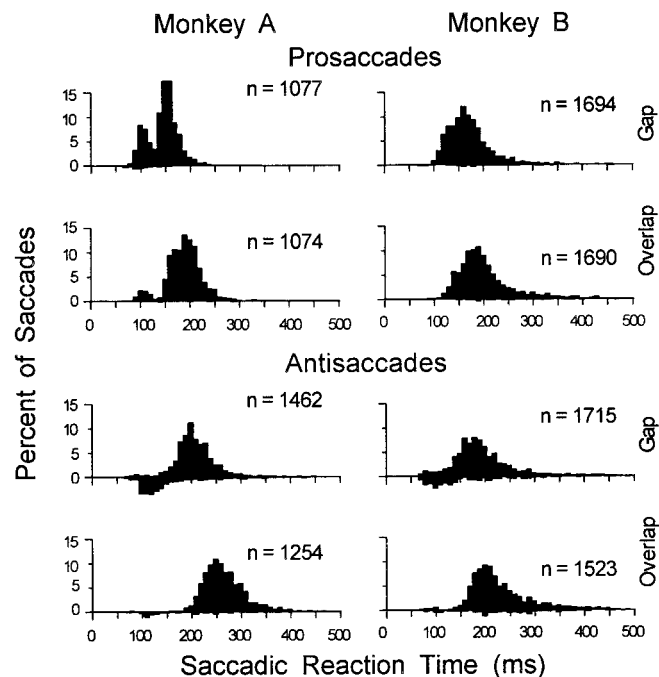


FIG. 2. The distribution of saccadic reaction times for the 2 monkeys in the different conditions of the pro- and antisaccade task. All data has been collapsed across stimulus eccentricity and direction. The bin width is 10 ms. The downward bars reveal directional errors.

TABLE 1. Mean saccadic reaction times, percentage of direction errors, and the anti/gap effects for the pro-/antisaccade paradigm

	Monkey A			Monkey B		
	Prosaccades	Antisaccades	Anti-effect	Prosaccades	Antisaccades	Anti-effect
Gap						
Correct SRT	151 ± 1	212 ± 1	61	177 ± 1	198 ± 2	22
Incorrect SRT	107 ± 4	164 ± 4		202 ± 9	175 ± 4	
Percent direction errors	2.0	27.4		3.5	22.4	
Overlap						
Correct SRT	193 ± 1	270 ± 1	77	207 ± 2	234 ± 2	27
Incorrect SRT	221 ± 28	198 ± 9		245 ± 10	244 ± 6	
Percent direction errors	0.6	7.1		3.3	13.1	
Gap effect	42	58		30	36	

Values are means ± SE. Saccadic reaction times (SRTs) are in milliseconds.

trials, they were trained on an identical task, this time using a green FP and a green eccentric stimulus. These two tasks (red FP/red stimulus; green FP/green stimulus) were eventually combined and randomly interleaved so that within a given block of trials, the FP and stimulus would be either red or green.

The next stage of the training procedure consisted of having eccentric stimuli of both colors appearing in opposite hemifields for each trial regardless of the color of the FP. Therefore within a block of trials, there were both red and green stimuli present, and the FP was varied randomly between either red or green. The monkeys were required to direct a saccade to the eccentric stimulus whose color matched the FP. As the monkeys improved on this task to the point where they were >75% accurate, the green eccentric stimulus was removed from trials with the red FP (i.e., prosaccade trials), and a temporal delay was added between the appearance of the red and green stimuli for trials with a green FP (i.e., antisaccade trials). The delay was gradually increased from 0 to 600 ms at which point the green stimulus appeared 100 ms after the monkey's time allowance to generate an antisaccade had expired. The monkeys received a small reward if they delayed their saccade until the green stimulus appeared, whereas they received a large reward if, immediately after the appearance of the red stimulus, they generated a saccade to the location where the green stimulus would appear. Therefore to optimize reward on trials with a green FP, monkeys had to generate an anticipatory saccade to where the green stimulus would appear corresponding to the mirror position of the red stimulus which was visible. This represents the final form of the pro- and antisaccade task used in this study. Both monkeys were trained extensively on this task and reached a consistent level of performance (95% prosaccades; 80% antisaccades) before the data collection.

Data analysis

Data analysis was performed on a SunSparc2 workstation. Data were first run through a saccade marking program, which was used to identify the beginning and end of each saccade based on velocity and acceleration template matching (Waitzman et al. 1991). All marks were later verified by the experimenter and adjusted if necessary. Saccades with latencies <80 ms had a 50% probability of being correct and were therefore rejected on the basis that they were anticipations. Likewise, those with latencies above 500 ms were assumed to be due to lack of attention and excluded. An off-line accuracy criteria of ±45° in direction from the intended saccade direction at any amplitude was used. This large acceptance window was necessary to fully examine the characteristics of antisaccades, whose kinematics and accuracy were considerably more variable than prosaccades. A direction error in the antisaccade task was defined as a saccade that fell within the acceptance window but on the stimulus side. Saccade gain was defined as the ratio of saccade amplitude to stimulus eccentricity. A gain above 1.0 indicated the saccade overshoot the desired end point (i.e., hypermetric), and a gain <1.0 indicated the saccade fell short (i.e., hypometric). As both monkeys showed similar trends, data were collapsed for the majority of the analyses. Statistical significance was tested with Student's *t*-tests ($P < 0.05$). All results are reported as means ± SE.

RESULTS

Distribution of saccadic reaction times

The distribution of saccadic reaction times (SRTs) for both monkeys, collapsed across stimulus eccentricity and direction,

TABLE 2. Breakdown of express and regular latency saccades for the pro-/antisaccade paradigm

	Monkey A		Monkey B	
	Pro	Anti	Pro	Anti
Express saccades				
Gap				
Correct	106 ± 1 (93.0)	101 ± 2 (83.6)	111 ± 1 (93.1)	99 ± 1 (66.5)
Incorrect	102 ± 2 (7.0)	107 ± 1 (16.4)	89 ± 2 (6.9)	100 ± 1 (33.5)
Overlap				
Correct	106 ± 1 (100)	91 ± 6 (83.3)	110 ± 4 (100)	96 ± 2 (47.5)
Incorrect	0 (0)	108 ± 2 (16.7)	0 (0)	101 ± 2 (52.5)
Regular saccades				
Gap				
Correct	162 ± 1 (99.4)	215 ± 1 (78.4)	180 ± 1 (96.7)	204 ± 2 (83.1)
Incorrect	131 ± 5 (0.6)	187 ± 4 (21.6)	213 ± 9 (3.3)	213 ± 5 (16.9)
Overlap				
Correct	198 ± 1 (99.4)	271 ± 1 (94.4)	208 ± 1 (96.8)	236 ± 2 (87.9)
Incorrect	221 ± 28 (0.6)	224 ± 10 (5.6)	248 ± 9 (3.2)	261 ± 6 (12.1)

Values are means ± SE with percentages in parentheses.

are shown in Fig. 2 and are summarized in Table 1. Both monkeys generated prosaccades with significantly shorter SRTs compared with antisaccades for both the gap and overlap conditions (see Table 1; $P < 0.001$). Furthermore both monkeys had a larger anti-effect, defined as the difference in mean SRT between anti- and prosaccades, in the overlap condition compared with the gap condition (see Table 1). SRTs were also significantly shorter for both pro- and antisaccades generated in the gap condition compared with the overlap condition (gap effect; see Table 1; $P < 0.001$). Both monkeys showed a larger gap effect, defined as the difference in mean SRT between the gap and overlap conditions, for antisaccades compared with prosaccades (see Table 1).

Monkey A showed a bimodal distribution of SRTs in the prosaccade task, particularly in the gap condition (Fig. 2). The two modes correspond to saccades of express (SRT: 80–120 ms) and regular latency (SRT: >120 ms) (Fischer and Boch 1983; Pare and Munoz 1996). A summary for the two monkeys is shown in Table 2. *Monkey B* did not produce a bimodal distribution of prosaccadic SRTs and generated very few correct prosaccades at express saccade latency. Both monkeys, however, generated reflexive prosaccades in the antisaccade task (i.e., directional errors), which occurred at express saccade latency. These were observed most often in the gap condition.

Influence of stimulus eccentricity and direction

SACCADIC REACTION TIME. Prosaccades were generated with shorter SRTs than antisaccades at all stimulus eccentricities and directions tested (Fig. 3; $P < 0.001$). Likewise, the gap condition consistently featured shorter SRTs than the overlap condition for all stimulus locations in both the pro- and anti-tasks ($P < 0.001$). Figure 3A shows the influence of stimulus eccentricity on SRT with data collapsed across stimulus direction. Mean SRTs were relatively large (mean SRT for all conditions: 225 ± 6 ms) for stimuli at 2° eccentricity. As eccentricity increased, mean SRTs decreased reaching a minimum at $8\text{--}10^\circ$ eccentricity (mean SRT across both 8 and 10° for all conditions: 202 ± 5 ms). When stimulus eccentricity was increased further, mean SRTs began increasing reaching a mean SRT for all conditions of 227 ± 9 ms for stimuli at 16° . Therefore there appeared to be an optimum eccentricity of $\sim 8\text{--}10^\circ$ where SRTs were at a minimum. This trend was present in both pro- and antisaccade tasks in both the gap and overlap conditions but was more prominent in the overlap condition.

For prosaccades, SRTs tended to be shorter when stimuli were located in the upper hemifield (Fig. 3B; $45\text{--}135^\circ$; mean SRT for gap condition and overlap conditions: 149 ± 1 and 183 ± 1 ms, respectively) compared with the lower hemifield ($225\text{--}315^\circ$; mean SRT: 194 ± 2 and 227 ± 2 ms, respectively; $P < 0.001$) but showed no apparent preference between stimuli located in either the right ($45, 315$, and 360° ; mean SRT for gap and overlap conditions: 156 ± 2 and 197 ± 3 ms, respectively) or left hemifields ($135\text{--}225^\circ$; mean SRT: 158 ± 2 and 196 ± 3 ms, respectively; $P > 0.05$). For antisaccades, SRTs were shortest when stimuli were located in the lower hemifield (mean SRT for gap and overlap conditions: 196 ± 2 and 243 ± 2 ms, respectively) compared with the upper hemifield (mean SRT: 232 ± 2 and 274 ± 2 ms, respectively; $P < 0.001$) with again, no preference between the right (mean SRT for gap and overlap conditions: 194 ± 2 and 241 ± 2 ms, respectively) and

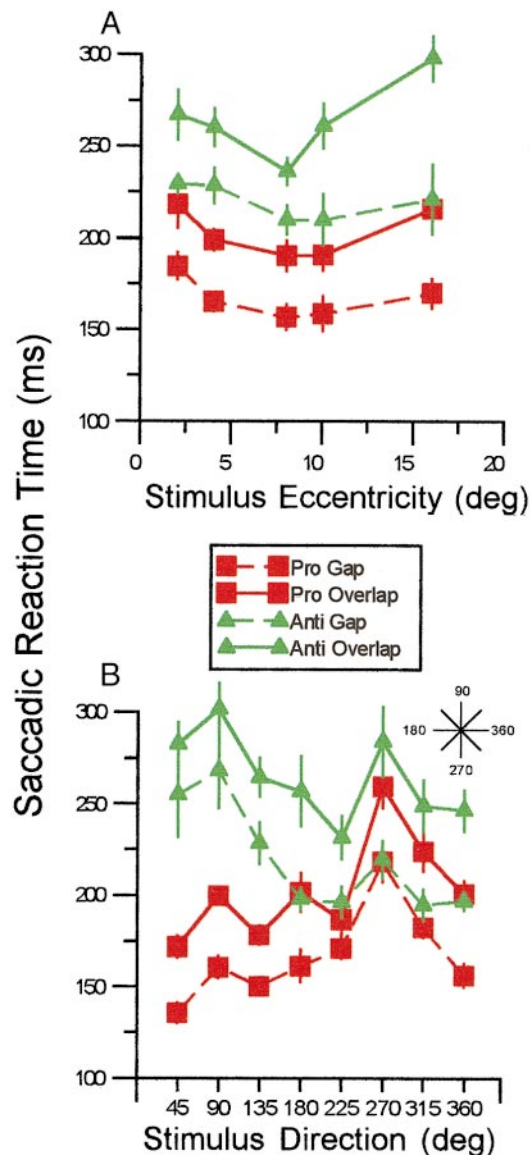


FIG. 3. The effect of stimulus eccentricity and direction on saccadic reaction time of all correct pro- (red squares) and antisaccades (green triangles) in both the gap (dashed lines) and overlap (solid lines) conditions. Note that in the case of antisaccade trials, the saccadic reaction times are for saccades directed away from the indicated stimulus location. A: data are collapsed across stimulus direction. B: data collapsed across stimulus eccentricity.

left hemifields (mean SRT: 196 ± 2 and 239 ± 3 ms, respectively; $P > 0.05$). Thus in both pro- and antisaccade tasks, SRTs were shorter when saccades were directed toward the upper hemifield compared with the lower hemifield.

DIRECTION ERRORS. Both monkeys made very few errors in the prosaccade tasks with the vast majority of direction errors occurring in the antisaccade trials. Furthermore the overlap condition featured significantly fewer direction errors compared with the gap condition (see Table 1).

Both monkeys had difficulty performing correct pro- and antisaccades to stimuli at 2° eccentricity (Fig. 4A; error rates for pro- and antisaccades: 7.7 and 17.9%, respectively). Performance improved when the stimulus eccentricity was increased to 4° (error rates: 0.8 and 4.5%, respectively). As eccentricity was increased beyond 4° , however, their performance steadily declined, reaching a maximum error rate of

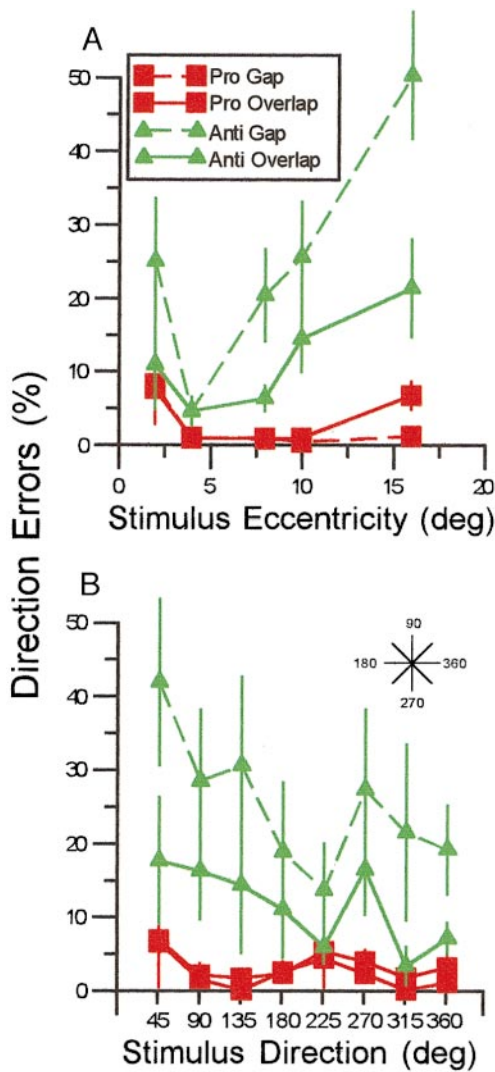


FIG. 4. The effect of stimulus eccentricity and direction on the frequency of directional errors of all pro- (red squares) and antisaccades (green triangles) in both the gap (dashed lines) and overlap (solid lines) conditions. *A*: data collapsed across stimulus direction. *B*: data collapsed across stimulus eccentricity.

3.8% for prosaccades and 36.3% for antisaccades for stimuli at 16° eccentricity. This trend was much more pronounced for antisaccades than for prosaccades ($P < 0.05$).

There was a small difference between the error rates for antisaccades when stimuli were presented in the upper hemifield (Fig. 4*B*; 45–135°; error rate for antisaccades: 21.6%) compared with the lower hemifield (225–315°; error rate: 17.1%; $P > 0.05$). Antisaccades to the oblique stimuli (45 and 225°, 135 and 315°), however, showed that the monkeys produced a greater number of errors in response to oblique stimuli presented in the upper hemifield (45–135°) compared with the lower hemifield (225–315°), particularly the upper right quadrant (45°). There was little difference between the error rates for stimuli located in the right (error rates for pro- and antisaccades: 1.6 and 16.5%, respectively) and left (error rates: 1.6 and 12.3%, respectively; $P > 0.05$) hemifields.

SACCADIC GAIN. Stimulus location affected the gain of antisaccades significantly (Fig. 5; $P < 0.01$) but had little effect on the gain of prosaccades. Moreover, the state of fixation (i.e., gap vs. overlap condition) had no effect on the gain of both pro- and antisaccades.

At small eccentricities (2, 4°), antisaccades were significantly hypermetric (i.e., overshooting the stimulus; mean gain for 2° stimuli: 2.8 ± 0.2). At 8° eccentricity, the gain of pro-

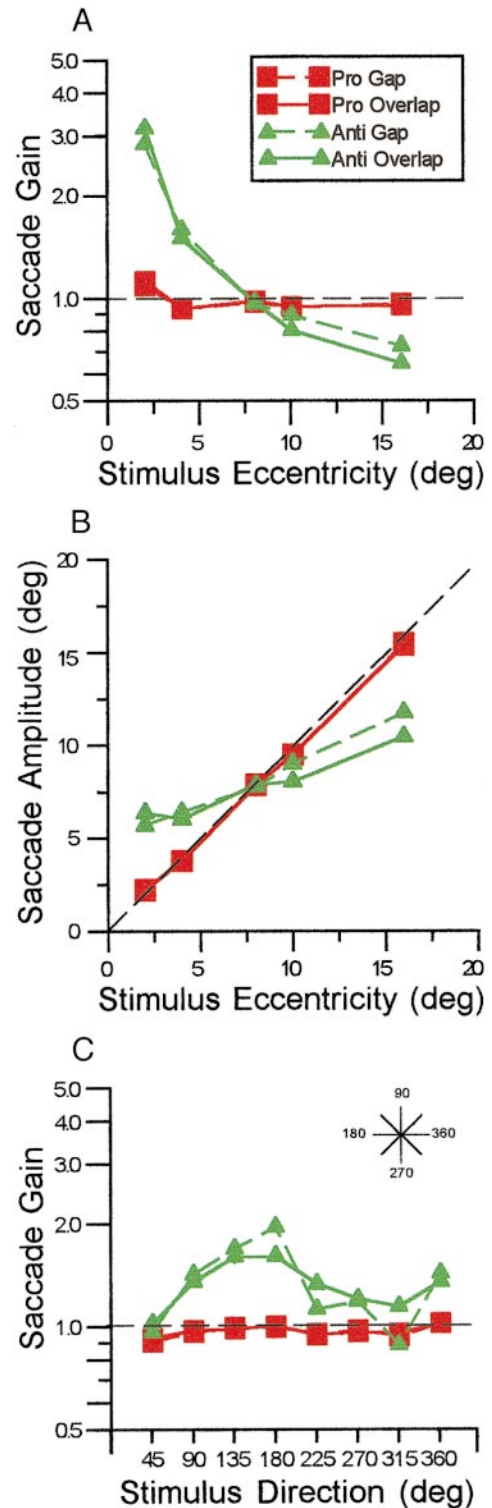


FIG. 5. The effect of stimulus eccentricity (*A*) and direction (*C*) on saccade gain and the effect of stimulus eccentricity on saccade amplitude (*B*) for all correct pro- (red squares) and antisaccades (green triangles) in both the gap (dashed lines) and overlap (solid lines) conditions. The black dashed lines represent a gain of 1.0 where every change in stimulus eccentricity is matched by a change in saccade amplitude. *A* and *B*: data collapsed across stimulus directions. *C*: data collapsed across stimulus eccentricity.

and antisaccades were the same (mean gain: 1.0 ± 0.02 and 1.0 ± 0.06 , respectively). At larger stimulus eccentricities ($\geq 10^\circ$), antisaccades were grossly hypometric (mean gain for stimuli at 16° : 0.6 ± 0.04). Although, the amplitude of antisaccades did change in response to increasing stimulus eccentricity (Fig. 5B), they were much less influenced compared with prosaccades, and were generated with an average amplitude between 8 and 10° .

When collapsed across stimulus eccentricity, antisaccades were hypermetric across all stimulus directions (Fig. 5C). Antisaccades directed toward the upper hemifield (i.e., stimuli in lower hemifield; mean gain: 1.4 ± 0.03) had larger gains than those directed to the lower hemifield (mean gain: 1.1 ± 0.02 ; $P < 0.001$). Similarly stimuli presented in the left hemifield (mean gain: 1.8 ± 0.05) elicited antisaccades with larger gains than those presented in the right hemifield (mean gain: 1.4 ± 0.03 ; $P < 0.001$).

PEAK SACCADIC VELOCITY. Both monkeys showed a significant interaction between stimulus location and peak saccadic velocity for pro- and antisaccades whereas the state of fixation (i.e., gap vs. overlap condition) had no effect (Fig. 6). As stimulus eccentricity increased, so too did peak saccadic velocity (Fig. 6A). Note the rate of increase for prosaccades was much greater than for antisaccades. Furthermore antisaccades, with the exception of stimuli located at 2° eccentricity, had slower peak velocities compared with

prosaccades for all stimulus eccentricities tested. However, because both animals made hypermetric antisaccades in response to small stimulus eccentricities (see Fig. 5), the peak velocities for these antisaccades were in fact slower than prosaccades of equal amplitude. Thus these results show that for all amplitudes of saccades, antisaccades were generated with lower peak saccadic velocities than prosaccades (see Fig. 6, C and D), consistent with previous human (Smit et al. 1987) and monkey (Amador et al. 1998) studies. Stimulus direction showed no discernable pattern of influence on peak saccadic velocity with the exception that prosaccades were faster than antisaccades across all stimulus directions tested (Fig. 6B).

Figure 6, C and D, shows the effect of saccadic amplitude on peak saccadic velocity (i.e., main sequence relationship) (Bahill et al. 1975) for each monkey with data collapsed across stimulus eccentricity and direction. Functions were fit using a linear regression with corresponding correlation values provided in the legend. Data from both monkeys revealed that as the amplitude of saccade increased, so did peak velocity for both pro- and antisaccades. However, for any given saccadic amplitude, antisaccades were slower than prosaccades and the main sequence revealed a shallower slope (prosaccade slope: 45.0 s^{-1} , antisaccade slope: 31.8 s^{-1}). The state of fixation (i.e., gap vs. overlap) had no effect on the main sequence for either pro- or antisaccades.

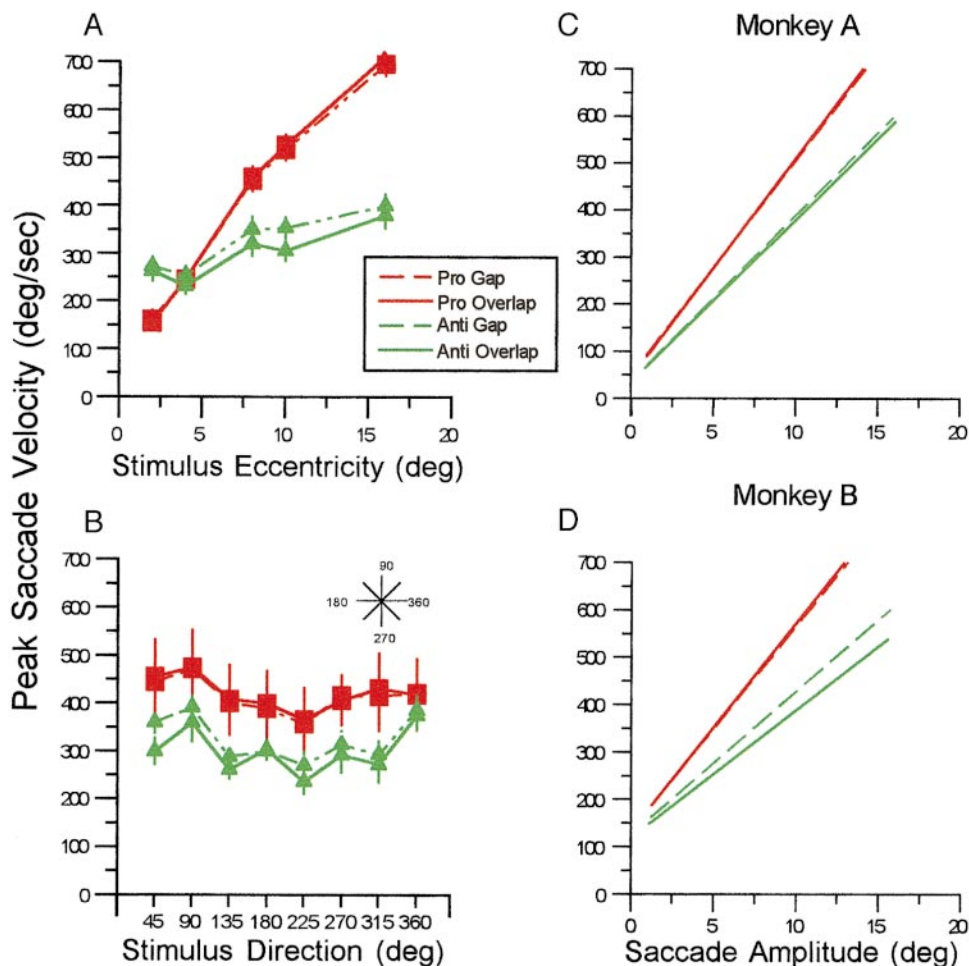


FIG. 6. The effect of stimulus eccentricity, direction (A and B) and the effect of saccade amplitude (C and D) on peak saccadic velocity of all correct pro- (red squares) and antisaccades (green triangle) in both the gap (dashed lines) and overlap (solid lines) conditions. A, C, and D: data collapsed across stimulus direction. B: data collapsed across stimulus eccentricity. Main sequence relationships for both monkeys (C and D) were fit using a linear regression with the following correlation values: prosaccades/gap $r^2 = 0.92$; prosaccades/overlap $r^2 = 0.93$; antisaccades/gap $r^2 = 0.75$; antisaccades/overlap $r^2 = 0.77$ for monkey A and prosaccades/gap $r^2 = 0.92$; prosaccades/overlap $r^2 = 0.92$; antisaccades/gap $r^2 = 0.83$; antisaccades/overlap $r^2 = 0.81$ for monkey B.

DISCUSSION

The present study has shown that non-human primates can be trained to generate both pro- and antisaccades to stimuli throughout the visual field. We have also described the systematic effects of stimulus eccentricity and direction on their behavioral characteristics. These results from non-human primates follow very similar trends in terms of metrics, kinematics, and reaction times to those observed in human studies (Fischer and Weber 1992, 1997; Goldring and Fischer 1997; Hallett 1978; Smit et al. 1987; J. M. Dafoe, J. R. Broughton, I. T. Armstrong, and D. P. Munoz, unpublished data) and provide important supporting evidence for the use of this animal model in the study of antisaccades.

Influence of stimulus location on saccade behavior

The location of the stimulus (i.e., stimulus eccentricity and direction) had systematic and often significant effects on the behavioral characteristics of both pro- and antisaccades. Much of the data suggested the presence of “preferred” stimulus locations that optimized performance. For example, the relationship between SRT and stimulus eccentricity revealed an optimal eccentricity of $\sim 8-10^\circ$ where SRTs were at a minimum (Fig. 3A). A possible contributing factor for the optimal amplitude of $8-10^\circ$ may be related to population coding of saccade metrics in the midbrain superior colliculus (SC). Saccade-related neurons in the intermediate layers of the SC are organized into a motor map coding the direction and amplitude of contraversive saccades (Robinson 1972). Small amplitude saccades are represented more rostrally. Prior to the execution of a saccade, a population of saccade-related neurons (Lee et al. 1988) spanning 1.5 mm of the SC are active (Munoz and Wurtz 1995b). The rostral SC also contains a subset of neurons that discharge during visual fixation and small amplitude, contraversive saccades (“fixation neurons”) (Krauzlis et al. 1997; Munoz and Wurtz 1993). It has been argued that there exists a continuum between the saccade-related neurons and the fixation neurons across the motor map of the SC (Munoz and Wurtz 1995b). In addition, anatomical studies have revealed stronger connections from the rostral SC onto brain stem omnipause neurons (Buttner-Ennever et al. 1999). Thus for small amplitude saccades, the rostral superior colliculus may provide a conflicting signal composed of both fixation and saccade commands that could take additional time to disambiguate, resulting in longer latencies for smaller amplitude saccades. In fact, it has already been argued that such a mechanism may account for the dead zone of express saccades, in which monkeys and humans are unable to generate small amplitude (i.e., $<2^\circ$) saccades at express saccade latency (Weber et al. 1992). Saccades of larger amplitudes (i.e., $>8-10^\circ$) should not experience this form of interference. However, their latencies are still increased. This could be due, in part, to a sensory perception factor. As the target increases in eccentricity, its location on the retina moves away from the fovea and progressively into an area of reduced visual acuity. Consequently the resulting sensory response, which will ultimately contribute to the activity necessary for driving a saccade, will be diminished, leading to the pattern of increased latencies for larger amplitude saccades shown in this and other studies (Kalesnykas and Hallett 1994).

An examination of the relationship between stimulus direc-

tion and SRTs reveals that in both the pro- and antisaccade tasks, SRTs were reduced when saccades were directed toward the upper hemifield compared with the lower hemifield (Fig. 3B). It is tempting to assume that this may be due to potential overtraining with these locations. However, this cannot account for the fact that similar trends have been reported in other studies employing both non-human primates (Boch et al. 1984) and humans (Fischer and Weber 1997; Kalesnykas and Hallett 1994; J. M. Dafoe, J. R. Broughton, I. T. Armstrong, and D. P. Munoz, unpublished data). The apparent bias toward certain stimulus locations could, therefore represent a neurological bias in the actual oculomotor system. For example, Thier and Andersen (1996) showed that the lateral intraparietal area (area LIP) features a greater representation for the upper visual field than the lower visual field. Consequently when Li and colleagues (1999) reversibly inactivated area LIP with muscimol, they found that saccades to the upper hemifield were significantly more impaired (i.e., hypometric with longer SRTs over controls) than those to the lower hemifield. This adaptation could represent a type of evolutionary and/or ecological inclination favoring quick orienting movements toward certain locations over others in an effort to maximize efficiency for specific behaviors. In support of this hypothesis, Previc (1990) argued that while the upper hemifield is primarily associated with extra-personal space, the lower hemifield deals more with peri-personal space. Optimizing movements to the upper hemifield could confer an advantage with respect to scanning for food or predators. Hemifield preferences in humans may also be influenced by societal/cultural effects. Previous human studies have reported biases in SRT and direction errors favoring the initiation of rightward saccades compared with leftward saccades (Fischer and Weber 1997; Fischer et al. 1997; Munoz et al. 1998; J. M. Dafoe, J. R. Broughton, I. T. Armstrong, and D. P. Munoz, unpublished results) This bias may be due, in part, to differences associated with reading patterns (i.e., horizontal-left-to-right) (Abed 1991) and would explain the absence of this trend in animals.

If the oculomotor system is indeed optimized for generating saccades to certain locations, this would explain, in part, why the monkeys had particular difficulties in performing antisaccades to certain locations. Both monkeys had increased difficulties in generating antisaccades in response to stimuli located in the upper hemifield, thus requiring *downward* saccades. The production of an antisaccade requires the suppression of a reflexive prosaccade to the stimuli and the generation of a motor program to a “virtual” target. The increase in direction errors in the antisaccade tasks to stimuli in the upper hemifield (Fig. 4B) may be a result of greater difficulty in suppressing an “optimized” movement (i.e., saccade to the upper hemifield) in favor of generating a “nonoptimized” movement (i.e., saccade to the lower hemifield). Furthermore the correct downward antisaccades they did generate were more hypermetric relative to those generated to the upper hemifield (Fig. 5B). Therefore if the monkey is successful in suppressing the reflexive prosaccade, an antisaccade that he makes to the lower hemifield (“nonoptimized”) would likely be less accurate than one generated to the upper hemifield (“optimized”).

A final point to consider, with respect to a potential neurological preference for certain saccade directions, relates to the finding that memory-guided saccades to the upper hemifield tend to be hypermetric whereas those to the lower hemifield are

often hypometric (Gnadt et al. 1991; White et al. 1994), similar to the pattern of dysmetria seen with antisaccades in this study. Why this "upward drift" was present in memory-guided saccades and antisaccades but not prosaccades may be due to the increased cognitive requirements of these two tasks and consequently, involvement of higher structures. Dias and Segraves (1999), for instance, showed that when the frontal eye fields (FEF) were inactivated with muscimol, this upward drift was attenuated. This provides further evidence that some of the directional biases/effects seen in this and other studies may be due to influences from cortical structures related to goals/target selection such as area LIP and the FEF and could consequently represent an adaptation in behavioral strategies.

It is important to note that in addition to the factors outlined in the preceding text, another possible contributing factor to the dysmetria of antisaccade amplitudes (Fig. 5A) is that, during training, it is possible that the monkeys may have received additional training on stimulus locations between 5 and 10°, which may have biased performance.

State of fixation

Consistent with results from human studies (Boch and Fischer 1986; Boch et al. 1984; Fischer and Boch 1983; Fischer and Ramsperger 1984; Fischer et al. 1993; Kalesnykas and Hallett 1987), the monkeys in our study generated saccades with shorter SRTs and a greater frequency of direction errors when stimuli were presented in the gap condition compared with the overlap condition. In contrast, the state of fixation appeared to have no effect on saccadic gain or saccadic velocity. Introducing a gap between the disappearance of the FP and the appearance of a saccadic target has been shown to increase preparatory activity in the paramedian pontine reticular formation (D. P. Munoz, M. C. Dorris, M. Pare, and S. Everling, unpublished observations), the SC (Dorris et al. 1997; Munoz and Wurtz 1995a) and the FEF (Dias and Bruce 1994; Everling and Munoz 2000). In addition, fixation-related signals in the superior colliculus are attenuated during gap conditions (Dorris and Munoz 1995). These changes in neuronal activity facilitate the initiation of saccadic eye movements and are believed to be responsible for the reduced SRTs seen in the gap condition (Dorris and Munoz 1995, 1998; Dorris et al. 1997; D. P. Munoz, M. C. Dorris, M. Pare, and S. Everling, unpublished data). Furthermore the presence of a gap period also facilitates the occurrence of express saccades (see Fischer and Weber 1997; Pare and Munoz 1996 for review). Express antisaccades, however, were not generated by either monkey. In fact, no study has ever presented evidence of correct antisaccades initiated at express saccade latency. Although the gap condition reduces fixation-related inhibition and increases preparatory activity, the appearance of a stimulus produces a transient visual burst of activity among saccade-related neurons in the superior colliculus contralateral to the stimulus but *ipsilateral* to the movement direction on an antisaccade trial (Everling et al. 1998a). This phasic visual burst coincides with reduced excitability of saccade-related neurons in the SC contralateral to the saccade direction therefore requiring additional time to generate a movement following this stimulus-related inhibition.

The increased error rates seen in the gap compared with the overlap condition can also be accounted for by changes in

excitability of neurons in the SC and FEF. In the case of antisaccade trials, both humans (Fischer and Weber 1997) and monkeys (current study see Fig. 2) (Amador et al. 1998) generate reflexive prosaccades during antisaccade trials (i.e., direction errors) at very short latencies, often in the range of express saccades (80–120 ms), particularly when using the gap condition (Fischer and Weber 1992, 1997). The reduction in fixation-related inhibition and the reciprocal increase in preparatory activity that occurs in the gap condition facilitates the generation of short-latency saccades. Because the animals presumably know that within a block of trials, the stimulus will appear at one of only two locations, there will be an increase in preparatory activity among saccade-related neurons coding for movements to either location (Everling and Munoz 2000; Everling et al. 1998a, 1999). Thus when the eccentric stimulus appears after the gap period, if the preparatory activity is high enough, the added stimulus-related phasic visual burst can be enough to surpass a saccadic threshold and drive a rapid, reflexive saccade to the stimulus.

The state of fixation had no effect on the gain or velocity of the saccades. Based on the evidence discussed in the preceding text, it would appear that SRT and the generation of a direction error are highly correlated to preparatory activity as well as other neuronal activity related to processing occurring prior to saccade initiation. Saccadic gain and velocity, on the other hand, deal with the actual execution of the saccade and are more dependent on the saccadic generating circuit in the brain stem reticular formation which may not be influenced by the state of fixation (Everling et al. 1998b; D. P. Munoz, M. C. Dorris, M. Pare, and S. Everling, unpublished data). It is, therefore possible that the neural correlates of saccadic gain and velocity are not associated with motor preparatory or fixation-related activity but other characteristics of activity within saccade-related structures. However, in contrast, Pratt (1998) reported a difference in peak velocity of prosaccades in humans in the gap and overlap conditions. A more detailed analysis of the relationship between neural activity and saccadic velocity may be required before any final conclusions can be made.

Non-human primates and antisaccades

One of the difficulties in using non-human primates as a model for the study of antisaccades is the uncertainty as to whether or not the animals understand the fundamentals of the task. Here we have employed a training regimen yielding consistent performance across three monkeys (2 providing data for this study and a 3rd animal that was subsequently trained for use in single-cell recording studies). These results are, for the most part, very consistent with the behavior observed in humans. After the completion of the training period, the monkeys were able to generate antisaccades to stimuli at all locations, indicating that they understood the task and were not simply generating saccades to locations of previous rewards. Moreover on occasions when the monkeys made a direction error to the stimulus during an antisaccade trial, they would often follow this up with a short-latency corrective saccade to the appropriate location despite not being rewarded for this behavior.

Another study that employed non-human primates to examine the behavioral characteristics of antisaccades was per-

formed by Amador and colleagues (1998). The paradigm used by these authors had several important differences which resulted in predictable differences in behavioral measures. First, they used an immediate condition and a delayed condition for stimulus presentation in contrast to our gap and overlap conditions. Successful completion of memory-guided delayed tasks requires the suppression of reflexive saccades and consequently significantly alters SRTs (Gnadt and Andersen 1988; Hikosaka and Wurtz 1983). Both monkeys in the Amador et al. (1998) study had, on average, longer SRTs than those in our study.

Second, one of the monkeys in the Amador et al. (1998) study (*MkA*) featured shorter SRTs in the antisaccade tasks compared with the prosaccade tasks, and SRTs for antisaccade direction errors were longer than correct antisaccades. These results are different from those obtained from a second monkey (*MkD*) as well as what is reported in the human literature (Fischer and Weber 1992, 1997; J. M. Dafoe, J. R. Broughton, I. T. Armstrong, and D. P. Munoz, unpublished results). Despite the fact that antisaccade SRTs were shorter than prosaccade SRTs in *monkey MkA*, they were still in the range of not only the antisaccade SRTs of the other monkey (*MkD*) but also the monkeys used in our study, allowing for differences in the experimental conditions. Thus it was performance on prosaccade trials that resulted in the differences in the SRT trends. Therefore it is likely the emphasis this particular monkey received during training on suppressing reflexive movements and delaying saccades is responsible for the increased SRTs for prosaccades (both correct prosaccades and incorrect antisaccades).

Despite the differences, these studies have both shown that it is possible to train primates to perform antisaccades revealing their suitability for neurophysiological studies into antisaccade generation. However, unlike humans who seem to automatically generate similar strategies to perform antisaccades, the method of training primates to perform antisaccades can have profound effects on how they complete the task. Consequently this can affect not only their behavior but may also have an effect on the neural correlates to this behavior. Therefore caution may be required in interpreting results from neurophysiological studies of primates and antisaccades to account for training effects.

Conclusions

The production of an antisaccade requires two functions: suppression of a reflexive prosaccade and the generation of a voluntary saccade to the opposite location in the absence of a guiding visual stimulus. The results from this study have shown that a monkey's ability to perform these two functions are affected by the location to where the saccade will be directed. We have also provided further evidence for a behavioral and neurological bias for certain saccade locations. Both pro- and antisaccades, directed toward the upper hemifield had short SRTs and were more accurate compared with those made to other locations. In addition, monkeys seem more likely to generate direction errors to those locations as opposed to other locations. The animals used in this study have shown similar trends to humans; this supports the use of monkeys to study the neural circuitry controlling the generation of antisaccades and translation of these findings to humans.

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