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Ann N Y Acad Sci. Author manuscript; available in PMC 2012 September 1.

Published in final edited form as:
Ann N Y Acad Sci. 2011 September ; 1233: 85–90. doi:10.1111/j.1749-6632.2011.06146.x.

Cells in the supraoculomotor area in monkeys with strabismus show activity related to the strabismus angle

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Abstract

We have earlier shown that monkeys reared with daily alternating monocular occlusion for the first few months of life develop large horizontal strabismus, A/V patterns, dissociated vertical deviation (DVD), and dissociated horizontal deviation (DHD). Here, we present results from neurophysiological experiments that show that neuronal activity of cells within the supraoculomotor area (SOA) of juvenile strabismic monkeys is correlated with the angle of strabismus. There was no modulation of SOA cell activity with conjugate eye position as tested during horizontal smooth pursuit. Comparison of SOA population activity in these strabismic animals and normal monkeys (described in the literature) suggests that both vergence (misalignment in the case of the strabismic animals) thresholds and vergence position sensitivities are different in the strabismic animals compared to the normals. Our data suggest that activity within the SOA cells is important in determining the state of horizontal strabismus possibly by altering vergence tone in extraocular muscle.

Keywords

strabismus; oculomotor; neurophysiology; near response area; vergence

Introduction

Infantile forms of strabismus occur in as much as 5% of all children.[1–3] A common feature among the different factors that lead to strabismus and correspondingly the different approaches to producing animal models for strabismus is that binocular vision is disrupted in early life due to breakdown in either motor fusion (e.g., surgical strabismus models) or sensory fusion (e.g., optically induced strabismus), or both.[4–6] We have previously reported that rearing infant monkeys with daily alternating monocular occlusion for the first several months of life results in a permanent strabismus whose properties include A/V patterns, dissociated vertical deviation (DVD), and alternating fixation, making the alternating monocular occlusion (AMO) model appropriate for studying human strabismus due to sensory disruption.[7–10] We have also shown, in the AMO model, that there was a direct correlation between the responses of horizontal and vertical motoneurons and the state of horizontal or vertical misalignment.[11,12] This was true for both the steady-state angle of misalignment and the eye movements associated with A and V patterns of strabismus. These studies presented the first direct evidence that the brain was intimately involved in

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Conflicts of interest
The authors declare no conflicts of interest.

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maintaining the strabismic state in the animals. The current study further examines neural involvement in setting the state of horizontal ocular misalignment. We report that cells in the supraoculomotor area (neural structure shown to contain vergence related cells in normal animals) show responses that are related to the strabismus angle in AMO monkeys with strabismus. Some of these data have appeared before in abstract form.[13]

Methods

Subjects and rearing paradigms

Two strabismic juvenile rhesus (*Macaca mulatta*) monkeys were the subjects of the study. Strabismus was induced by disrupting development of binocular vision in infant monkeys using a daily AMO method. In the AMO rearing paradigm, soon after birth (within the first 24 h), an occluding patch (dark contact lenses) is placed in front of one eye for a period of 24 h and switched to the fellow eye for the next 24 h. Thereafter, the eye of occlusion was alternated daily for a period of four months. See our other publications for further details on rearing and properties of the strabismus due to AMO rearing.

Surgical procedures

After special rearing, the AMO animals were allowed to grow normally (unrestricted vision) until approximately four years of age, before behavioral and neurophysiological experiments were begun. Sterile surgical procedures performed under aseptic conditions using isoflurane anesthesia (1.25%–2.5%) were used to stereotaxically implant a head stabilization post and a recording chamber.[14] The chamber placement allowed full access to both oculomotor nuclei and the area immediately adjacent to the oculomotor nucleus (the supraoculomotor area, SOA).[11] During the same surgical procedure, a scleral search coil was implanted in one eye according to Judge *et al.*[15] Later, in a second surgery, a second scleral search coil was implanted in the other eye. All procedures were performed in strict compliance with National Institutes of Health and the Association for Research in Vision and Ophthalmology guidelines, and the protocols were reviewed and approved by the Institutional Animal Care and Use Committees.

Experimental paradigms, data acquisition, and analysis

Binocular eye position was measured using the magnetic search coil method (Primelec Industries, Regensdorf, Switzerland). Eye coil signals were calibrated by rewarding the monkey for looking within a ± 2° window surrounding a 0.5° target spot that was rear projected on a tangent screen 60 cm away from the animal. Calibration of each eye was performed independently during monocular viewing.

Data were collected as the monkeys performed fixation and horizontal or vertical sinusoidal smooth pursuit (0.2 Hz, ± 10°) tasks under monocular viewing conditions at the 60 cm target viewing distance. Eye and target position feedback signals were processed with anti-aliasing filters at 400 Hz before digitization at 1Khz with 12-bit precision (Alpha-Lab System, Alpha-Omega Engineering, Nazareth, Israel). Extracellular neuronal responses were acquired using epoxy-coated tungsten microelectrodes with impedance of approximately 1 mega-ohm. Following head-stage amplification and post-amplification, raw spike data were acquired in our data acquisition system at a sampling rate of 32 KHz. Spike sorting was performed offline using a template matching algorithm (Spike 2 software, CED Cambridge Electronic Design, England). Data analysis was performed with custom software routines written in Matlab (Mathworks). Unit response was represented as a spike density function that was generated by convolving time stamps with a 20-ms Gaussian.[16] The goal of the analysis was to correlate state of misalignment with neuronal responses within the SOA.

Results

Properties of strabismus

Static eye misalignment in animals raised with the AMO method has been described before.[8,11] Briefly, AMO animals develop large horizontal strabismus, “A” or “V” patterns, and DVD. In addition, animals also show a dissociated horizontal deviation (DHD) wherein the angle of horizontal misalignment varies depending on eye of fixation. In this study, when tested while viewing a straight-ahead target, animal S1 had an exotropia of approximately 20° during right eye viewing and 30° during left eye viewing. Under the same testing conditions, animal S2 had an exotropia of 10° during right eye viewing and 20° during left eye viewing. We were able to take advantage of the variation in the strabismus angle with fixating eye to identify and test the SOA cells.

SOA cell responses during changes in eye misalignment

Data were acquired from 28 cells from within the SOA of the two animals (19 from S1 and 9 from S2). These cells were localized to a region that was 1–2 mm dorsal and dorsolateral to the oculomotor nucleus. Since our chamber was inclined in the coronal plane at an angle of 20° to the mid-saggital plane, we often encountered the SOA cells about 1–2 mm before encountering burst-tonic cells in the oculomotor nucleus within the same electrode track penetration.

All the SOA cells described in this report showed an increase in firing rate when the angle of exotropia was reduced by changing the eye of fixation. Since both the animals showed less exotropia during right eye viewing, the SOA cells increased activity when switching fixation from left eye to right eye. These cells were therefore called the near-response cells, as they increased firing rate for a convergence eye movement. Note that the eyes are not converged *per se*; rather, there is a reduction in the degree of divergent misalignment (exotropia). Note also that all the testing was performed at a single viewing distance of 60 cm. The change in neuronal response and the corresponding change in misalignment are brought about by changing the eye of fixation (DHD) and not due to changing target distance.

Figure 1 shows an example of a typical near-response cell in the SOA of the strabismic animal. The top panel shows the eye positions while the bottom panel shows the neuronal responses. When the animal is viewing with his right eye, the angle of misalignment is less than the angle of misalignment when viewing with his left eye. Correspondingly, the neuronal response is less when viewing with the left eye than when viewing with the right eye. The change in neuronal response leads the change in eye misalignment, suggesting that the neuronal drive from within the SOA is helping to set the state of eye misalignment. In addition to the near-response cells, we also observed fewer cells that showed the opposite kind of response, i.e., an increased firing rate for an increase in angle of exotropia (far-response cells), but they are not the subject of this report.

SOA cell responses during conjugate eye movements

To verify that the SOA cells indeed encoded eye misalignment and not an eye position signal, we also tested these cells during conjugate eye movements. Figure 2 shows an example of neural response of SOA cells during horizontal smooth pursuit, right eye viewing. Clearly, there is no modulation due to the tracking of the sinusoidally moving target. None of the cells in our sample showed any evidence of response modulation during smooth-pursuit eye movements. The combined response characteristics shown in Figures 1 and 2 are evidence that the neuron indeed encodes eye misalignment (vergence change) and not eye position.

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Quantification of eye misalignment (vergence) sensitivities of SOA cells

In order to quantify the relationship between the firing rate of the SOA cells and the strabismus angle, we performed a regression between the average firing rate and the corresponding strabismus angle. Data obtained during fixation with each eye viewing were used to develop the fit. In order to improve the power of the fit, we also included additional data points that corresponded to some spontaneous variations in the animal's state of eye misalignment. From the regression line for each cell, we calculated the slope, indicating the sensitivity (spikes/s/degree of misalignment) of the cell, and the threshold, indicating the angle of exotropia at which the cell commenced firing.

Table 1 shows the average sensitivities and the thresholds of the population of SOA cells in each of the two monkeys. Included in the table is also the average sensitivity and threshold for near-response cells recorded from normal monkeys as reported by Mays.[17] Data in the table show that there is a significant reduction of sensitivity in SOA cells when compared to the normal. In addition, there is also a significant shift in the threshold toward a divergent state.

Discussion

We have for the first time identified cells that appear to carry a signal related to the strabismus angle. These cells were identified within the supraoculomotor area in the strabismic monkeys. It is highly likely that these cells are the same as those that have been reported to encode the vergence angle in normal animals.[17–19] First, the anatomical locations of these cells correspond very well to the midbrain near-response region identified before. We were also able to verify the location of the recording via histological reconstruction of electrode track penetrations. Second, the neuronal response characteristics correspond very well to the near response cells of the normal animal. Near-response cells in the normal animal show modulation related to vergence (difference in position of the two eyes) but not to conjugate eye movements. Similarly, cells in our sample show responses related to the strabismus angle (difference in position of the two eyes—Fig. 1) but not conjugate eye movements (Fig. 2). Finally, we encountered many more near-response cells than far-response cells, similar to the distribution reported earlier.

A significant finding in our study is that the response characteristics of the near-response cells in the strabismic monkey are altered from the normal animal. The threshold (vergence angle at which the neuron commences firing) for the normal animals is close to 0.0°, while the threshold for the strabismic monkeys was approximately −40° and −27° (40° and 27° of exotropia or divergence) respectively. We suggest that the fact that SOA cells show significant levels of activity even in the divergent state is evidence that these cells are indeed involved in maintaining the state of strabismus. The reduced thresholds can perhaps be explained from within a recently developed framework for binocular control.[20,21] Thus, King and colleagues proposed that the neural integrators encoded monocular eye position and that they provided inputs to the SOA such that SOA activity encoded the difference in position of each eye. In the normal monkey, during a conjugate eye movement, SOA activity would simply provide a DC signal to the medial rectus motoneurons that may be referred to as the "vergence tone." During a vergence movement (again in the normal monkey), the SOA cells provide a required positional command to the medial rectus motoneurons that eventually helps to adduct each eye. Our data are compatible with the idea that the SOA cells do not encode a "vergence" command *per se*; rather, they encode the difference between the positions of the two eyes (strabismus angle). If they were simply encoding vergence, we might not have expected to observe the reduced thresholds and we might have predicted that these cells would be silent in the exotropic state. We, of course, cannot comment on whether the difference signal is arriving from monocular neural integrators, but it stands to reason that there is some representation of each eye's position upstream of the SOA. Note that we cannot rule out the classical Hering model for binocular control, wherein the SOA supplies a vergence command to medial rectus motoneurons. If the thresholds of the SOA cells were adaptively altered (a vergence offset) in the strabismic animals toward the divergent (exotropic) direction, then any modulation of SOA cell activity could be the source of the vergence command that leads to observed change in eye misalignment. There is in fact some evidence that SOA cells can adapt to different levels of tonic vergence. Morley and colleagues showed that relationship between SOA cell activity and vergence was altered in approximately 70% of cells following phoria adaptation.[22] Perhaps a similar adaptive mechanism can cause reduced thresholds in the strabismic monkeys.

The second difference between the normal and strabismic animals' SOA activity was the reduced sensitivities (Table 1). We suggest that the reduced sensitivity for vergence could manifest as a reduced vergence tone in extraocular muscle and therefore result in the monkeys maintaining an exotropic state. In support of our hypothesis, the strabismic animal with the lower sensitivity had a larger exotropia and a more reduced threshold. Note that we are not claiming that the SOA activity is the reason that the animals developed an exotropia in the first place. Rather, we suggest that the SOA cells are the substrate that helps *maintain* the divergent state.

Several studies have shown that many of the SOA cells encode not only the vergence angle but also ocular accommodation.[23,24] Unfortunately, we did not have the technical capability to monitor or control levels of accommodation in our animals. Potentially some of the misalignment sensitivity measures developed here for the SOA cells could be contaminated by sensitivity to accommodation. However, it is highly unlikely that the observed differences in threshold and sensitivity of the SOA population of the strabismic monkeys compared to the normal animals is driven by changes in the accommodative component alone.

Acknowledgments

We wish to thank Dr. Michael Mustari for help with surgical implantation, Dr. Anand Joshi for helpful discussions on the manuscript, and Michelle Swann for technical assistance. This work was supported by NIH grant RO1-EY015312 (VED); UHCO core grant P30 EY 07551; and Yerkes base grant RR00165.

References

1. Lorenz B. Genetics of isolated and syndromic strabismus: facts and perspectives. Strabismus. 2002; 10:147–156. [PubMed: 12221494]
2. Govindan M, et al. Incidence and types of childhood exotropia: a population-based study. Ophthalmology. 2005; 112:104–108. [PubMed: 15629828]
3. von Noorden, GK.; Campos, EC. Binocular Vision and Ocular Motility: Theory and Management of Strabismus. Mosby; St. Louis: 2002.
4. Economides JR, et al. Ocular motor behavior in macaques with surgical exotropia. J Neurophysiol. 2007; 98:3411–3422. [PubMed: 17928552]
5. Smith EL 3rd, et al. Binocularity in kittens reared with optically induced squint. Science. 1979; 204:875–877. [PubMed: 441743]
6. Crawford ML, von Noorden GK. Optically induced concomitant strabismus in monkeys. Invest Ophthalmol Vis Sci. 1980; 19:1105–1109. [PubMed: 7410001]
7. Das VE. Alternating fixation and saccade behavior in nonhuman primates with alternating occlusion-induced exotropia. Invest Ophthalmol Vis Sci. 2009; 50:3703–3710. [PubMed: 19279316]
8. Das VE, et al. Incomitance in monkeys with strabismus. Strabismus. 2005; 13:33–41. [PubMed: 15824015]

9. Das VE, et al. Conjugate adaptation of saccadic gain in non-human primates with strabismus. J Neurophysiol. 2004; 91:1078–1084. [PubMed: 14586028]
10. Fu L, et al. Horizontal saccade disconjugacy in strabismic monkeys. Invest Ophthalmol Vis Sci. 2007; 48:3107–3114. [PubMed: 17591880]
11. Das VE, Mustari MJ. Correlation of cross-axis eye movements and motoneuron activity in non-human primates with “A” pattern strabismus. Invest Ophthalmol Vis Sci. 2007; 48:665–674. [PubMed: 17251464]
12. Joshi, AC.; Das, VE. ARVO e-abstr. 2011. Medial Rectus Motoneuron Activity in Non-Human Primates with Exotropia.
13. Das, VE. ARVO e-abstr. 2010. Response Properties of Cells in the Supraoculomotor Area in Non-Human Primates with Strabismus.
14. Das VE, et al. Information processing by parafoveal cells in the primate nucleus of the optic tract. Exp Brain Research. 2001; 140:301–310.
15. Judge SJ, Richmond BJ, Chu FC. Implantation of magnetic search coils for measurement of eye position: an improved method. Vision Res. 1980; 20:535–538. [PubMed: 6776685]
16. Richmond BJ, Optican LM. Temporal encoding of two-dimensional patterns by single units in primate inferior temporal cortex. II. Quantification of response waveform. J Neurophysiol. 1987; 57:147–161. [PubMed: 3559669]
17. Mays LE. Neural control of vergence eye movements: convergence and divergence neurons in midbrain. J Neurophysiol. 1984; 51:1091–1108. [PubMed: 6726313]
18. Mays LE, et al. Neural control of vergence eye movements: neurons encoding vergence velocity. J Neurophysiol. 1986; 56:1007–1021. [PubMed: 3783225]
19. Zhang Y, Gamlin PD, Mays LE. Antidromic identification of midbrain near response cells projecting to the oculomotor nucleus. Exp Brain Res. 1991; 84:525–528. [PubMed: 1864324]
20. King WM, Zhou W. New ideas about binocular coordination of eye movements: is there a chameleon in the primate family tree? Anat Rec. 2000; 261:153–161. [PubMed: 10944576]
21. King WM, Zhou W. Neural basis of disjunctive eye movements. Ann N Y Acad Sci. 2002; 956:273–283. [PubMed: 11960811]
22. Morley JW, Judge SJ, Lindsey JW. Role of monkey midbrain near-response neurons in phoria adaptation. J Neurophysiol. 1992; 67:1475–1492. [PubMed: 1629759]
23. Judge SJ, Cumming BG. Neurons in the monkey midbrain with activity related to vergence eye movement and accommodation. J Neurophysiol. 1986; 55:915–930. [PubMed: 3711972]
24. Zhang Y, Mays LE, Gamlin PD. Characteristics of near response cells projecting to the oculomotor nucleus. J Neurophysiol. 1992; 67:944–960. [PubMed: 1588393]

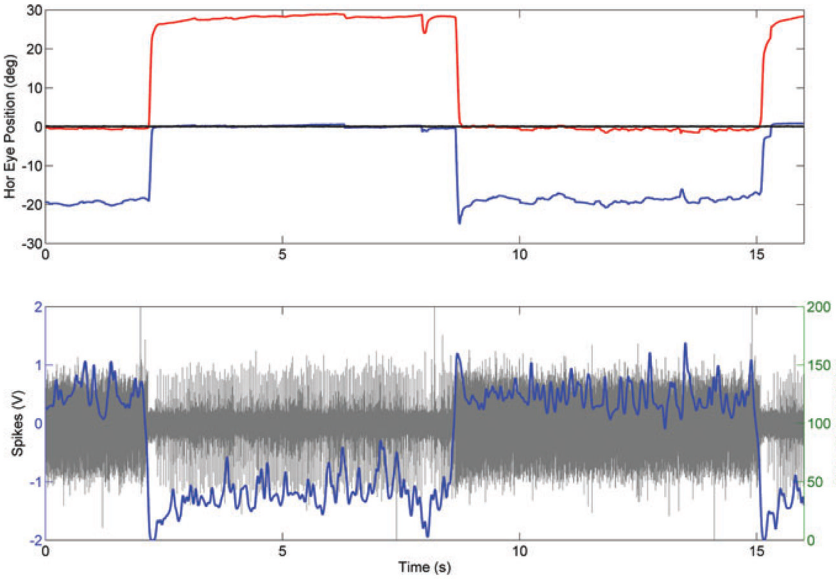

Figure 1.

Activity of a typical SOA near-response cell in animal S1 during monocular fixation with either the right eye or the left eye. Top panel shows eye positions (right eye, red; left eye, blue) and the bottom panel shows neural activity (gray, raw action potentials; blue, unit spike density function). Upward and rightward eye positions are positive. The eye positions in the top panel show that the non-fixating eye is abducted (exotropia). The panels show that when the animal views with his right eye, the angle of exotropia is less, and the neural firing rate of the SOA cell is high. When the animal switches fixation to the left eye, the angle of exotropia increases (divergent movement) and there is a reduction in firing rate.

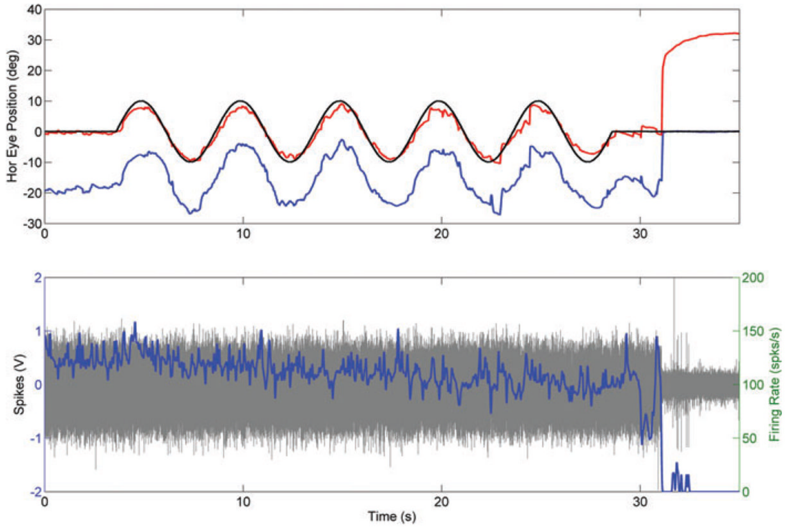

Figure 2.

Activity of SOA cell during monocular right eye viewing horizontal smooth pursuit. Top panel shows eye positions (right eye, red; left eye, blue; target, black) and bottom panel shows neural activity (gray, raw action potentials; blue, unit spike density function). Since the animal is viewing with the right eye, the left eye is deviated to the left. The unit response shows no modulation associated with the tracking response, indicating that the cell is not encoding eye position.

Table 1

Population characteristics of near-response SOA cells

Population properties	Strabismic monkey M1	Strabismic monkey M2	Normal monkey (from Ref. 17)
Sensitivity to eye misalignment	4.5 spks/s/degree	5.85 spks/s/degree	10.6 spks/s/degree
Threshold	−39.3° (exotropia)	−27.3° (exotropia)	−0.5˚