Effects of Enucleation of the Nondeprived Eye on Stimulus Deprivation Amblyopia in Monkeys

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The effects of enucleation of the nondeprived eye on the visual function of the deprived eye were studied in two rhesus monkeys reared with long-term lid suture. The lids of one eye of each monkey were surgically fused at the age of approximately 1 month. The lids were opened at ages 19 months and 22 months, and the nondeprived eyes were enucleated at age 57 months. Three measures of visual function (spatial modulation sensitivity, temporal modulation sensitivity, and increment-threshold spectral sensitivity) were determined for the nondeprived and deprived eyes before enucleation and for the deprived eyes during a 9-month period following enucleation. Neither the spatial nor temporal modulation sensitivity measurements showed significant recovery of visual function. The postenucleation spectral sensitivity data showed some recovery of sensitivity over the long wavelength region of the spectrum during the first 3 months, but there was no further recovery over the next 6 months. Therefore, the results of the experiments show that enucleation of the nondeprived eye of monkeys with severe stimulus deprivation amblyopia does not result in a significant functional improvement of the deprived eye. Invest Ophthalmol Vis Sci 25:10-18, 1984

Many of the alterations of the visual system produced by unilateral form deprivation in infant subjects can be attributed to competitive interactions between the afferent inputs from the two eyes during early development. According to the competition model, inadequate pattern stimulation of the deprived eye gives the nondeprived eye a competitive advantage, and as a result, its neurons gain control of central connections at the expense of those from the deprived eye.1-3 In the cat, there is evidence that indicates that these antagonistic binocular interactions may actually persist beyond the classical critical period of development, possibly throughout the lifetime of the animal.4-9 For example, it has been shown that the proportion of neurons in the striate cortex, which respond to stimulation of the deprived eye of an adult cat that had been reared with monocular lid suture, increases dramatically following enucleation of the nondeprived eye.5-8 The increase in control of cortical neurons by the deprived eye occurs immediately after enucleation and does not require subsequent visual experience.5-8 Therefore, these results suggest that, prior to enucleation, activity from the deprived eye was being suppressed or inhibited by activity from the nondeprived eye. In addition to these physiological studies, it has also been reported that a considerable amount of behavioral recovery of visual acuity can be obtained in cats using the enucleation paradigm,9,10 although there is some controversy regarding the degree of recovery.11,12

For the primate there is also evidence of antagonistic binocular interactions in adult amblyopes. For example, active inhibitory interactions in humans have been suggested by studies that have demonstrated that visual function of an amblyopic eye is adversely affected by stimulation of the nonamblyopic eye.13-15 In the monkey, Hendrickson et al16 have shown that photocoagulation of the retina of the deprived eye at 9 months of age resulted in a better visual acuity of the deprived eye than in the deprived eyes of monkeys with earlier lesions or without lesions. These data suggest that the initially deprived eye was suppressed by the sensory input from the normal eye. However, investigations of interocular inhibition in monocularly lid-sutured cats17 have shown that it is necessary to eliminate both the sensory input and the proprioceptive input from the extraocular muscles of the normal eye to significantly disinhibit the deprived eye. Therefore, the most unequivocal test of the existence of functional, but suppressed neural connections from a deprived eye of a primate would be an enucleation experiment similar to those performed with cats.5-8 In this respect, it has been reported,18 although not well documented, that human adult strabismic amblyopes may recover visual acuity of the amblyopic eye if the nonamblyopic
eye is lost as a result of trauma or disease. The present study was undertaken to investigate the effects of enucleation of the nondeprived eyes on the functional capability of the deprived eyes of two rhesus monkeys who had been reared with long-term monocular lid suture. The two monkeys had been subjects in several previous behavioral investigations of the sensitivity of their deprived and nondeprived eyes so that their pre-enucleation visual functions were well documented. Three measures of visual function (spatial modulation sensitivity, temporal modulation sensitivity, and increment-threshold spectral sensitivity) were assessed by behavioral methods over a 9-month interval following enucleation of the nondeprived eyes.

Materials and Methods

The subjects for the experiments were two well-trained rhesus monkeys (Macaca mulatta). The eyelids of the monkeys were sutured closed when they were about 1 month of age using procedures previously described. The eyelids were opened at age 19 months for one monkey (7722) and at age 23 months for the other monkey (7721). The refractive status of each eye was determined under cycloplegia by retinoscopy and with an infrared optometer (Bausch and Lomb Ophthalmetron) after the eyelids were opened. One subject (7722) was found to be a compound myopic anisometrope and the other subject (7721) was a compound hyperopic anisometrope. The refractive errors for both subjects were determined at 1-year intervals, and the amount of anisometropia was found to be nearly stable over a period of 3 years. The nondeprived, left eyes of both monkeys were enucleated under aseptic operating room conditions using standard surgical procedures when they were 4 years and 9 months old. Therefore, they had approximately 3 years (3 yrs 2 mos for subject 7722, and 2 yrs 10 mos for subject 7721) of passive binocular visual exposure before their left eyes were enucleated. Both monkeys recovered from the surgery without any apparent adverse physiological or behavioral effects.

During the daily experimental sessions, the animals were placed in a standard primate chair inside a sound-attenuating room. The primate chair was fitted with a response lever on the waist plate and a drinking spout on the neck plate through which the unconditioned reinforcer was delivered. A lens holder device was positioned in cosine phase with the center of the screen. The lens holder consisted of two lens wells cut in aluminum plates to hold 1 inch diameter lenses to correct the refractive errors of the subjects. The plates were positioned so that the monkey’s eyes were centered in the lens wells when his mouth was on the juice spout. The head restraints prevented lateral and rotational head movements to insure the proper alignment of the monkey’s eyes with the lens wells.

The behavioral procedures and psychophysical methods used in the present study have been previously described. Briefly, the behavioral procedure was a temporal interval detection task for which the monkeys were trained to press and hold a response lever at the beginning of each trial, which was signaled by the onset of an auditory cue. The lever press initiated a variable interval foreperiod preceding the presentation of the detection stimulus. If the monkey released the lever within a brief (limited-hold) period following the onset of the detection stimulus, it was assumed that he had detected the stimulus and he was rewarded with a conditioned reinforcer (tone) after each trial and orange drink on a random probability basis. On the other hand, if the monkey failed to release the lever within the criterion period it was considered that he had failed to detect the stimulus. Data were collected using a descending method of limits, where the threshold was defined as the stimulus intensity for which the monkey failed to release the lever within the criterion response period in two consecutive trials. However, to maintain stimulus control of the animals’ behavior, the stimulus intensity was not predictably increased after two consecutive misses, but rather, the second and each subsequent miss had a 0.5 probability of resetting it. The final threshold estimates were derived from the geometric mean of 12 threshold measurements with resulting standard errors of approximately 0.05 log units, ie, approximately the size of the symbols used in the figures.

The stimuli for the contrast sensitivity experiments were electronically generated on the CRT of an oscilloscope using the method described by Campbell and Green. The stimulus field subtended a 4-degree visual angle at the 114 cm viewing distance. The space average luminance of the screen was 40 cd/m². The subjects viewed the screen monocularly with natural pupils. In the spatial modulation sensitivity experiments, the detection stimulus was a stationary vertical grating presented for 500 msec with square-wave onset and offset properties. The determination of the spatial modulation sensitivity of the subjects’ deprived eyes required the use of very low spatial frequency gratings (0.12 to 0.25 c/deg). In order to prevent the occurrence of increments or decrements in the mean luminance of the test field, the low-frequency gratings were positioned in cosine phase with the center of the screen. In the temporal modulation sensitivity experiments, the luminance of the entire screen was sinusoidally modulated about its mean luminance to produce a uniform field flicker. To prevent the occurrence of any extraneous luminance transients, the modulation input to the oscilloscope was gated to initiate the stimulus...
flicker at a zero crossing point of the modulation voltage. The flickering stimulus was presented for a 1-second viewing duration and the limited-hold period was 1,200 msec.

The optical system used in the spectral sensitivity experiments was a two-channel Maxwellian view system with a 2.5 mm exit pupil. The source for both channels was a 1,000 watt, heat-filtered xenon arc lamp. The background and test fields subtended visual angles of 10 degrees and 2 degrees, respectively. The test flash duration was 50 msec, and the limited-hold period was 500 msec. The monochromatic stimuli, obtained by a Jarrel-Ash Mark X monochromater with a 10-nm half-band width, were superimposed on a 3,000 troland achromatic background.

**Results**

**Spatial Modulation Sensitivity**

The results of the spatial modulation sensitivity experiments are shown in Figure 1. The data in Figures 1A and B show the contrast sensitivity functions for the nondeprived eyes (open circles) and deprived eyes (filled circles) prior to enucleation. These data are similar to the previously published data for these animals and show a profound reduction in contrast sensitivity of the deprived eye relative to the nondeprived eye. The extrapolated cut-off spatial frequency for the deprived eyes of both animals is approximately 0.3 c/deg, which is about 6 octaves lower than that for the nondeprived eyes. The data for the deprived eyes shown in Figures 1A and B were collected one month prior to enucleation and are nearly identical to the data collected only one week after the lids were opened (see Fig. 1, ref 21). Therefore, three years of passive binocular experience had not resulted in any measurable recovery.

The spatial modulation sensitivity data for the deprived eyes, following enucleation of the nondeprived eyes, are shown in Figures 1C and D (the coordinate system in Figs. 1C and D has been expanded compared to Figs. 1A and B). The filled circles represent the preenucleation data replotted from Figures 1A and B and the lines connect the ±1 SD values of these sensitivity estimates. The diamonds represent data collected about 4.5 months after enucleation of the nondeprived eye, and the squares represent data collected about 8.5 months following enucleation. All of the postenucleation data fall within the ±1 SD boundaries of the preenucleation data for both animals. Therefore, these experiments also fail to show significant recovery of visual function as a result of enucleation of the nondeprived eye.

**Temporal Modulation Sensitivity**

The data from the temporal modulation sensitivity experiments are shown in Figure 2 following the same presentation format as the spatial modulation sensitivity data shown in Figure 1. The pre-enucleation data (Figs. 2A and B), as previously reported, show a relative reduction in sensitivity of the deprived eyes (filled circles) compared to the nondeprived eyes (open circles) at all temporal frequencies. However, the relative individual differences between the two eyes are not as large in the temporal domain as in the spatial domain, with the extrapolated critical fusion frequencies for the deprived eyes being only about 1 octave lower than that of the nondeprived eyes. The preenucleation data (filled circles) for the deprived eyes have been replotted for comparison to the postenucleation data in Figures 2C and D. The postenucleation data from these experiments are in agreement with the spatial modulation sensitivity experiments in showing that the data collected during the fourth (diamonds) and eighth (squares) months after enucleation fall within the ±1 SD boundaries of the preenucleation data for both animals. Therefore, these experiments also fail to show significant recovery of visual function as a result of enucleation of the nondeprived eye.

**Increment-Threshold Spectral Sensitivity**

The pre-enucleation data from the increment-threshold spectral sensitivity experiments are shown in Figure 3. In these experiments, the monochromatic test field was superimposed upon a white adaptation field of 3000 trolands. The data for the nondeprived eyes (Figs. 3A and B; open circles) show the three distinct peaks that are typical for increment-threshold spectral sensitivity data obtained with a moderately intense white background for either humans or monkeys. In contrast, pre-enucleation data for the deprived eye, which replicate the results of the previous experiments on these animals, show a unimodal function with a peak at approximately 500 nm rather than the three peaks seen for the other eye. The data for the deprived eyes of both animals are adequately described by the CIE scotopic luminosity curve. Therefore, the pre-enucleation data show that the sensitivity of the deprived eye is determined by the scotopic mechanism even at photopic luminance levels. In addition, the sensitivities of the deprived eyes are over 2 log units lower than the nondeprived eyes at the peak of the spectral sensitivity function.

The increment-threshold spectral sensitivity functions following enucleation of the nondeprived eyes are shown in Figure 4 for subject 7722 and in Figure 4.
Fig. 1. Spatial modulation sensitivity functions for the two subjects. Panels A and B show the pre-enucleation data (open circles for the nondeprived eyes and filled circles for the deprived eyes). Panels C and D show a comparison of the pre-enucleation and postenucleation data for the deprived eyes on an expanded coordinate system. The filled circles are the pre-enucleation data replotted from above and the lines connect the ±1 SD values of these data. The diamonds and squares represent data collected 4.5 months and 8.5 months after enucleation, respectively.

5 for subject 7721. The data collected 1 week after the enucleation (Figs. 4A and 5A) are nearly identical to the pre-enucleation data and are still well described by the CIE scotopic luminosity function over the entire spectral range. However, the spectral sensitivity function obtained 3 months after enucleation (Figs. 4B and 5B) show an elevation in sensitivity over the long wavelength region of the spectrum compared to the pre-enucleation functions. At 3 months postenucleation, although the sensitivity data for wavelengths less than 600 nm were still well described by the scotopic luminosity curve, there was an obvious photopic intrusion for wavelengths longer than 600 nm. The shapes of these spectral sensitivity functions are very similar...
Fig. 2. Temporal modulation sensitivity functions for the two subjects. Panels A and B show the pre-enucleation data (open circles for the nondeprived eyes and filled circles for the deprived eyes). Panels C and D show a comparison of the pre-enucleation and postenucleation data for the deprived eyes. The filled circles are the pre-enucleation data replotted from above and the lines connect the ±1 SD values of these data. The diamonds and squares represent data collected 4.5 months and 8.5 months after enucleation, respectively.

To human scotopic spectral sensitivity functions derived from visually evoked cortical potentials. The nature of the photopic intrusion is interesting because that portion of the curve is too narrow to be described by either the CIE photopic luminosity curve for the standard observer or by the absorption spectrum of the long wavelength sensitive cones of the primate eye. Although the nature of the photopic mechanism, which determines the spectral sensitivity of the eye at the long wavelengths, cannot be defined with certainty, the curves drawn through the data points for each monkey are the same functions drawn through the long wavelength portion of the spectral sensitivity data for their nondeprived eyes shown in Figure 3. These curves were derived from the linear, subtractive-interaction equations proposed by Sperling and Harwerth and would indicate that both the long wavelength sensitive cones and the middle wavelength sensitive cones are involved in the function. However, regardless of the mechanism involved, these data show an elevation in the spectral sensitivity of the deprived eye at 3 months postenucleation that had not occurred with approximately 3 years of passive binocular experience.

The spectral sensitivity functions determined 6 months after enucleation (Figs. 4C and 5C) and
months after enucleation (Figs. 4D and 5D) show that there was no further change in the functions. The data at both of the longer postenucleation periods closely match those collected three months after enucleation. Therefore, the elevation in spectral sensitivity that occurred between the second week and third month after enucleation was apparently a permanent change, but was not the beginning of a more substantial improvement in sensitivity.

Discussion

The results of these experiments show that enucleation of the nondeprived eyes of monkeys with stimulus deprivation amblyopia does not result in a significant functional improvement of their deprived eyes. Even 9 months after enucleation, there was no significant recovery of visual function as measured by the spatial modulation sensitivity or temporal modulation sensitivity functions. The increment-threshold spectral sensitivity data at 3 months postenucleation showed some recovery of photopic sensitivity in the long wavelength region of the spectrum, but there was no further recovery beyond that seen at 3 months. Even though there was some improvement in the spectral sensitivity of the deprived eye, it was not found immediately after enucleation and may have simply been associated with the forced usage of the eye rather than with a release from suppression. Therefore, the results indicate that the reduced visual capacities of monkeys following long-term monocular lid suture cannot be attributed to an active suppression of the amblyopic eye by the normal eye. However, complete recovery should not have been expected since previous experiments on monkeys have shown that some of the visual system alterations resulting from lid suture were caused by disuse rather than abnormal binocular interactions, since parts of the afferent visual system were affected.
Fig. 4. Postenucleation increment-threshold spectral sensitivity functions for subject 7722 using a 3,000 td white background. Data are shown for four postenucleation periods indicated on the graphs. The broad curve with a peak at 505 nm is the CIE scotopic luminosity curve. The curve drawn through the data for the long wavelengths in panels B, C and D is the same curve (shown in Fig. 3) that was drawn through the data points at these wavelengths for the nondeprived eye.

where binocular interaction cannot occur.\textsuperscript{35,36} Moreover, histological studies\textsuperscript{37} of lid-sutured monkeys have shown retinal ganglion cell changes, which would logically not be reversed by the elimination of any type of interocular suppression.

The results of these experiments with monkeys are somewhat different than what might have been anticipated from the previous studies on cats.\textsuperscript{5-9} In cats, the enucleation of the nondeprived eye resulted in a rapid and substantial increase in the percentage of cortical cells that could be driven by the deprived eye.\textsuperscript{5-8} and in some, but not all, studies, a recovery of behaviorally determined visual acuity.\textsuperscript{9-12} However, the physiological experiments in cats showed that most of the cortical neurons, which were responsive to stimulation by the deprived eye, did not have normal receptive fields.\textsuperscript{5-8} Therefore, there may be considerable intersubject variability in these studies or there may be an imperfect translation of physiological recovery to behavioral recovery. It should also be noted that if the same amount of intersubject variability exists for monkeys, our sample of two monkeys may not com-
completely define the recovery of visual function following enucleation in this species.

The lack of recovery in visual function following enucleation may have been related to the type and depth of the amblyopia of the subjects used in the present study. It is well known that stimulus deprivation amblyopia in humans is usually more severe and more intractable than other forms of amblyopia, e.g., strabismic or anisometropic amblyopia. While it is known to clinicians that amblyopic patients may recover some function of the amblyopic eye after loss of vision of the nonamblyopic eye, such observations have been limited to strabismic amblyopia. We are not aware of similar observations in patients with stimulus deprivation amblyopia. Therefore, the lack of recovery in the monkey subjects of the present study may be related to the fact that they had a very severe amblyopia (0.3 c/deg extrapolated cut-off spatial frequency, which converts to a 20/2000 Snellen fraction), which was caused by stimulus deprivation. It is possible that more substantial recovery of visual function could result from enucleation of the nonamblyopic eyes of monkeys with less severe amblyopia or with other forms of amblyopia where abnormal binocular interactions could play a greater role. However, from the present studies it seems appropriate to conclude that humans with long-standing stimulus deprivation amblyopia of a severe degree should not expect very much

Fig. 5. Postenucleation increment-threshold spectral sensitivity function for subject 7721. See Figure 4 for details.
improvement in the function of that eye if the non-amblyopic eye is lost as a result of disease or trauma.

Key words: amblyopia, stimulus deprivation, enucleation, monkeys, animal psychophysics, contrast sensitivity, spectral sensitivity

Acknowledgment
The authors thank Mark Ruttum, MD for his assistance with the surgical procedures.

References