Early Visual Deprivation Results in Persistent Strabismus and Nystagmus in Monkeys

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To understand to what extent visual-pattern deprivation during infancy results in strabismus and nystagmus, the authors examined the long-term consequences of this type of deprivation in monkeys during the first 50 days of life. Three cynomolgus and three rhesus monkeys had the eyelids sutured closed within 24 hr of birth. At 25 days of age, the eyelids were opened, and the eyelids of the fellow eye were sutured closed for an additional 25 days (reverse-eyelid suture). When the eyelids were opened at 50 days of age, each monkey was found to have 20–30Δ of exotropia and nystagmus, which persisted for the duration of the study (1 yr). The cynomolgus monkeys developed a monocular 8–10 Hz pendular nystagmus in the eye sutured first. The rhesus monkeys developed a conjugate nystagmus with both jerk and pendular components. The slow phases often had velocity-increasing profiles. The rhesus monkeys also had a superimposed latent component to the nystagmus during monocular viewing. One additional rhesus monkey was examined after 55 days of binocular-eyelid suturing. This monkey also developed exotropia and nystagmus resembling that of the other rhesus monkeys. These findings suggest that early pattern vision in monkeys is necessary for the development of normal ocular alignment and gaze-holding ability. Invest Ophthalmol Vis Sci 32:134-141, 1991

Infantile strabismus and congenital nystagmus (jerk nystagmus with velocity-increasing slow phases) are two of the more common pediatric oculomotor disorders. To what extent the development of these oculomotor disorders is primarily due to a sensory versus a motor defect is still controversial.1-3 The most compelling evidence that visual deprivation may cause these disorders comes from studies on children who underwent surgery for congenital cataracts.4-9 Unfortunately, the type of strabismus and nystagmus that develops in these children is not well characterized, and eye movements have not been recorded. Furthermore, many children with congenital cataracts have cerebral palsy or mental retardation,10 therefore, it is unclear to what extent the oculomotor disorders that occur in these children is due to visual deprivation versus an underlying neurologic abnormality.

To understand further to what extent early visual-pattern deprivation causes strabismus and nystagmus, we measured ocular alignment and recorded eye movements using a scleral search coil in macaque monkeys that were visually deprived for another study.11 These animals underwent monocular-eyelid suturing for the first 25 days of life, which was then reversed for the subsequent 25 days. Some of the results from this study have been published in abstract form.12,13

Materials and Methods

Subjects and Rearing Conditions

Five rhesus monkeys (Macaca mulatta) and seven cynomolgus monkeys (M. fascicularis) born in captivity in the primate colony at the National Institutes of Mental Health were used in this project. All procedures with the monkeys were approved by the Animal Care and Use Committee at NIMH (Access # LCM105) and were in compliance with the ARVO Resolution on the Use of Animals in Research.

Six of the animals had the lids of one eye sutured closed between 12–24 hr after birth. Before eyelid suture, the animals were sedated with intramuscular injections of 0.25 mg of ketamine hydrochloride. The epithelial margins of the upper and lower eyelids were trimmed, and the tarsus and skin edges were sutured closed with six 6–0 silk sutures. Postoperatively, the animals were given a 4-day course of trimethoprim and sulfadiazine and were examined daily for signs of infection and eyelid separation. If there were any question that an eyelid was beginning to open, the eyelid was resutured. After 7–10 days, the eyelids were adherent, and the sutures were removed. At 25

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days of age, the eyelids were opened with sharp dissection, and the lids of the other eye were sutured closed for another 25 days (reverse-eyelid suture). At 50 days of age, the lids of the second sutured eye were opened. One additional monkey (Rh4) had the lids of both eyes sutured closed within 24 hr of birth for 55 days.

The animals were reared individually in clear plastic incubators for the first 55 days after birth. They were housed in a primate nursery whose light was cycled (16 hr of light and 8 hr of dark). After 55 days, they were raised in cages containing one or two other monkeys of similar age. At 1–1.5 yr of age they were housed separately in cages. Five control monkeys were reared under the same conditions except they had no eyelid suturing. Ocular alignment was first measured in all animals between 50–55 days of age. Eye movements were first observed in all animals beginning on day 1 of life and were first measured by a scleral search coil between 25–80 days of age. Table 1 summarizes the deprivation schedule, the age in which the scleral search coils were implanted, and the age at which each animal was last evaluated. Two of the monkeys (Rh1 and Rh2) were studied only until the age of 56 days because they were used in a metabolic study.

**Procedure**

Ocular alignment was measured using a cover test. This was done with the monkey fixating on a toy or the tube of the feeding bottle at 33 cm. If a deviation was detected, a Krimsky corneal light reflex measurement was done. A photographic determination of ocular alignment was also performed. A 35-mm camera equipped with a 90-mm macro lens with a flash was placed 30 cm in front of the monkey. Multiple photographs on slide film were made with the animal fixating on a toy. The slides were projected, and the ocular deviations in mm were determined.

We followed the method described by Judge et al. to implant the eye coils used for the eye-movement recording. Eye movements from each eye were recorded while the monkey sat in a small primate chair with its head fixed. During the first 6 months of life, the monkey's head was held stationary by velcro straps attached to the chair. At 6 months of age, a removable aluminum halo, of the type used in human beings, was attached to the animal's skull to immobilize the head during eye-movement recordings. The recording system was calibrated by rotating the magnetic field coils around the animal by 20° in each direction and measuring the average eye position while the animal was in the dark. After the age of 6 months, the monkeys were trained to fixate and follow a target light by reinforcing good performance with a sweetened liquid. The horizontal and vertical eye position of each eye was then calibrated based on fixation of a small visual target. The animals were examined for spontaneous nystagmus and for latent nystagmus by covering one eye.

Visual spatial acuity was measured using Teller Acuity Cards (VisTech Consultants, Dayton, OH). These cards have a square-wave grating pattern displayed on one end of the card and a homogeneous gray field matched in mean luminance on the other end. Visual spatial acuity was determined using the preferential-looking technique described by Preston et al. During the test the monkey either sat in a monkey chair or was held by one of the investigators with its head held fixed. The monkey's face was positioned 38 cm in front of the gratings. Visual spatial acuity was tested monocularly. The eye not being tested was occluded with an opaque eye patch. The cards were shown in order of increasing spatial frequency. The investigator was masked to which eye was sutured first, the spatial frequency of the card, and the side of the card on which the grating appeared.

Nonparametric statistics were used because of our small sample size. Differences between independent samples (first lid-sutured eye against second lid-sutured eye) were analyzed using a paired t-test.

**Results**

**Ocular Alignment**

All visually deprived animals had exotropia ranging from 20–30° when both eyes were opened between 50–55 days of age. Figure 1 shows a photo-
Fig. 1. Cynomolgus monkey (Cn1) at 63 days of age following reverse-eyelid suture for the first 51 days of age (top), and rhesus monkey (Rh4) at 68 days of age following 55 days of binocular-eyelid suture (bottom). These animals had 20Δ and 30Δ of exotropia, respectively.

Graph of the corneal light reflex of monkey Cn1 at 63 days of age and monkey Rh4 at 68 days of age, who had 20 and 30Δ of exotropia, respectively. Ocular alignment was repeatedly measured in five experimental animals until at least 1 yr of age (Fig. 2). Figure 2A shows the ocular alignment based on the Krimsky corneal light reflex test. Figure 2B shows the photographic determination of ocular alignment. On the left vertical axis of Figure 2B the ocular displacement is plotted in mm. By comparing the measurements in Figures 2A and 2B, we calculated the average Hirschberg ratio for the five animals to be 9.5° ± 1.4° (or 19Δ) per mm of corneal light-reflex displacement. The exotropia persisted for the duration of the study. All of the monkeys with reverse-eyelid suturing had marked fixation preferences for the eye that was sutured last. The monkey with binocular lid suturing (Rh4) did not have a fixation preference. Ocular alignment was also measured in the control animals beginning at 2 weeks of age, and all were found to have normal ocular alignment.

Eye Movements

In the light, none of the monkeys in the control group had spontaneous nystagmus at birth or at any time during the study. In the dark, however, two of the monkeys in the control group had persistent down-beat nystagmus with linear slow phases (average slow phase eye velocity, 4°/sec) detected during eye-movement recordings.

All of the animals that were visually deprived developed spontaneous nystagmus in the light and dark that persisted for the duration of the study. We will describe the nystagmus separately for the cynomolgus and rhesus monkeys because the degree and type of nystagmus was species dependent.

The cynomolgus monkeys developed a monocular pendular nystagmus in the eye that was deprived first directed in both the vertical and horizontal planes, which was present both in the dark and light. The average frequency of the pendular nystagmus was 8–10 Hz. These animals also generated large dynamic overshoots during saccadic eye movements in the eye deprived first. The time of onset of the pendular nystagmus and dynamic overshoots is unknown as both were present by 10 weeks of age when the binocular eye coils were first inserted. Two of these monkeys also had down-beat nystagmus in the dark similar to the control monkeys. None of the animals developed latent nystagmus. Spontaneous eye movements from cynomolgus monkey Cn3 at age 62 weeks are shown in Figure 3.

The rhesus monkeys developed pendular nystagmus and dynamic overshoots with the same features as the cynomolgus monkeys, except that they occurred in both eyes. In addition, the rhesus monkeys developed a binocular jerk nystagmus directed in both the horizontal and vertical planes. The slow phases of the jerk nystagmus were often velocity increasing, especially in eccentric gaze. These monkeys also had a latent component to their jerk nystagmus when one eye was occluded with a patch, ie, during left-eye viewing, there was a conjugate left-beating nystagmus, and during right-eye viewing, there was a conjugate right-beating nystagmus. All three types of
nystagmus and the dynamic overshoots were found in both the reverse-sutured monkeys and the binocularly sutured monkey. Spontaneous eye movements in one of the reverse-eyelid sutured monkeys (Rh3) at age 150 weeks are shown in Figure 4. Those in the binocularly sutured rhesus monkey (Rh4) at age 105 weeks during binocular and monocular viewing are shown in Figures 5 and 6, respectively.

The time of onset of the nystagmus in the reverse-sutured rhesus monkeys was 2–3 days after the reversal. At that time the monkeys had a conjugate jerk nystagmus with slow phases directed up and nasally with respect to the opened eye. The slow phases appeared linear, and the peak velocity reached a maximum of 10°/sec. During week 2 after the reversal, the nystagmus became more variable. At that time the jerk nystagmus occurred in all directions, and the slow phases were often of increasing velocity. We are not certain when the nystagmus developed in the binocularly sutured rhesus monkey. Binocular eye coils were inserted a few days before the eyes were opened (age 52 days). At that time the monkey already had a conjugate horizontal and vertical jerk nystagmus with velocity-increasing slow phases. The pendular nystagmus did not become evident until 9–12 days after the eyelids were opened.

Visual Spatial Acuity

Five of the experimental animals had serial visual spatial acuity measurements (Fig. 7). Reliable visual spatial acuity measurements could not be obtained after approximately 1 yr of age because the monkeys were easily distracted at this age and habituated to the grating cards. For the four animals with reverse-eyelid suturing, the mean visual spatial acuity of the eye sutured first at 1 yr of age was 0.83 ± 0.73 cycles per degree (cpd) compared with 7.4 ± 2.0 cpd in the fellow eye; this was statistically significant based on a paired student t-test (t = 7.04; P = 0.006). The visual spatial acuity in the monkey with binocular-eyelid suture (Rh4) at 1 yr of age was 7.4 cpd, but this had to be measured binocularly because of the degree of latent nystagmus this animal had during monocular viewing. The visual spatial acuity of normally reared rhesus monkeys at 1 yr of age ranges from 20–30 cpd.17

Discussion

Strabismus due to Visual Deprivation

All monkeys in the visually deprived group developed exotropia, which persisted for the duration of the study. We do not believe the strabismus was caused by insertion of the eye coils at an early age for the following reasons. All of the cynomolgus monkeys in the visually deprived group had exotropia at age 55 days when both eyes were opened, and the eye coils were not inserted into these monkeys until 69–75 days of age (Table 1). No strabismus developed in the monkeys in the control group even though they had binocular eye coils inserted at approximately the same age as the monkeys in the visually deprived group.

What determines whether exotropia or esotropia develops after visual deprivation during infancy? In human infants with visual impairment from a number of different causes including congenital cataracts,
both esotropia and exotropia occur, although esotropia has been reported more frequently. In these studies, however, the precise onset of visual deprivation was unknown. Therefore, whether the type of ocular misalignment that occurs depends on the timing of deprivation during infancy is unknown.

This question may be answered by primate studies. Two previous studies describe strabismus in monkeys after visual deprivation, although in these studies esotropia was reported. In these studies, the animals were reared in the dark for 3–6 months beginning at 2 weeks of age or received monocular lensectomy at 3–7 days of age followed by aphakic correction and partial-to-complete occlusion of the fellow eye for 3–7 months. Thus, visual deprivation in these studies was delayed several days to weeks after birth. Visual deprivation in our study was initiated within 24 hr of birth, and in each case, exotropia developed. Further study is needed to confirm whether visual deprivation initiated during the first few days of life primarily results in exotropia, and deprivation initiated later during infancy primarily results in esotropia.

What is the possible mechanism for deprivation-induced strabismus? We speculate that strabismus occurs due to a loss of fusion combined with a change in vergence tone. Monkeys reared with 4–8 weeks of monocular, binocular, or reverse-eyelid suturing during infancy have a persistent loss of binocularity driven cells in striate cortex. Similarly, human subjects given short-term patching during infancy do not develop normal binocularity. As a result of this loss of binocularity, both monkeys and humans would not be expected to develop normal fusion. They would, therefore, lack one of the major sensory systems used to detect ocular misalignment. Whether exotropia or esotropia then develops may depend on the vergence tone at the time of visual deprivation.
Fig. 5. Spontaneous nystagmus in the binocular-eyelid sutured rhesus monkey (Rh4) at 105 weeks of age. Hr, right eye horizontal eye position; Vr, right eye vertical eye position; Hl, left eye horizontal eye position; Ll, left eye vertical eye position. One-second tick marks are shown above the trace. In the dark this animal has a low-velocity, left-beating nystagmus whose slow phases are primarily linear. There is also a small amplitude conjugate pendular nystagmus. When the monkey views the inside of a full-field patterned OKN drum, the spontaneous jerk nystagmus with linear slow phases is nearly extinguished, but the monkey has pendular nystagmus. In the light there is also a jerk nystagmus with velocity-increasing slow phases in eccentric gaze.

Vergence tone has not been examined in monkey infants, but it has been examined in human infants. Over 50% of newborn infants have significant exotropia, whereas <1% had significant esotropia.24 Over the course of the next 6 months, the exotropia is corrected, and 0.1% of the infants develop persistent esotropia. This correction is postulated to be due to an exuberant convergence mechanism.25 If a similar change in vergence tone occurs in monkeys, one might postulate that visual deprivation within 24 hr of birth results in persistent exotropia by preventing a normally occurring increase in vergence tone. In contrast, visual deprivation begun at a later time during infancy may result in esotropia, by allowing vergence tone to increase without binocular visual feedback.

Nystagmus due to Visual Deprivation

Our study suggests that visual-pattern deprivation during the first 50 days of life consistently results in nystagmus in macaque monkeys. Reverse-eyelid suturing in cynomolgus monkeys causes monocular pendular nystagmus in the eye sutured first. This type of deprivation in rhesus monkeys causes binocular pendular nystagmus, binocular jerk nystagmus with velocity-increasing slow phases, and a latent nystagmus. Similar types of nystagmus have been described in patients treated for monocular and binocular congenital cataracts, although the waveforms have never been confirmed by eye movement recordings.4-9

Why do rhesus and cynomolgus monkeys develop different types of nystagmus despite being exposed to the same visual deprivation schedule? One possibility is that there is a species difference in the types of nystagmus that results from visual deprivation. Alternatively, the rhesus monkeys may have a longer critical period during which visual deprivation can alter the development of gaze-holding systems. For example, if the critical period in the rhesus monkeys extended beyond the time of the reverse-eyelid suturing...
A) VISUAL ACUITY
EYE DEPRIVED FIRST

![Graph A](image1)

B) VISUAL ACUITY
EYE DEPRIVED SECOND

![Graph B](image2)

Fig. 7. Visual spatial acuity in five experimental monkeys. Successive measurements for each monkey are connected by a line. Spatial acuity is plotted in cycles per degree (cpd) on the left vertical axis, and by Snellen measurements on the right vertical axis. Spatial acuity in the eye that was sutured first (A) was less than the spatial acuity of the fellow eye that was sutured second (B). The spatial acuity in monkey Rh4 (binocular-eyelid suture) had to be examined with both eyes opened because of the severity of the latent nystagmus that occurred during monocular viewing.

(25 days), these monkeys would have had each eye closed during the critical period. The consequence of this may be similar to binocular-eyelid suturing. If the critical period in the cynomolgus monkeys did not extend beyond the time of the reverse-eyelid suturing, then they in effect would have only received monocular deprivation. In other words, we are suggesting that if one eye is visually deprived during the critical period then monocular pendular nystagmus occurs; if each eye is visually deprived during the critical period, then binocular pendular and binocular jerk nystagmus occurs.

What is the critical period for the development of visual deprivation-induced nystagmus during infancy? Although there have been several studies done of visually deprived monkeys, only a few studies have reported nystagmus. In the studies reporting nystagmus, the visual deprivation was started in the first 2 weeks of life; in those that did not report nystagmus, the deprivation was usually not started until after 2 weeks of life. These data suggest that normal visual input during the first 2 weeks of life may be sufficient to prevent visual deprivation-induced nystagmus in monkeys.

What is the cause of nystagmus in visually deprived monkeys and why do up to three different types occur? We believe that specific types of nystagmus occur as a result of visual deprivation of specific structures in the brainstem, cerebellum, and cerebral cortex involved in gaze holding. Horizontal and/or vertical jerk nystagmus with velocity-increasing slow phases is probably due to visual deafferentation of the eye position neural integrator. Instability of the neural integrator results in jerk nystagmus with velocity-increasing slow phases. The neural integrator is mediated by neurons in the medial vestibular nucleus, nucleus prepositus hypoglossi, interstitial nucleus of Cajal, and the flocculus of the cerebellum. Furthermore, in monkeys, all of these areas receive visual motion input from the nucleus of the optic tract (NOT). In contrast, monocular and binocular pendular nystagmus is probably due to visual deafferentation of the flocculus by the inferior olivary nucleus. Low-intensity electrical stimulation of the flocculus in monkeys elicits vertical, horizontal, and torsional slow phases in the ipsilateral eye. One source of visual input to the flocculus is from the NOT and accessory optic system, which in turn project via the tectospinal tract to the inferior olive, which in turn projects to the flocculus. Lesions in the tectospinal tract and the adjacent central tegmental tract result in pendular nystagmus. Latent nystagmus may be due to visual deafferentation of occipital cerebral cortex involved in optokinetic nystagmus. Bilateral occipital lobectomy in monkeys results in a latent nystagmus.

Visual Spatial Acuity

At 3 weeks of age, the visual spatial acuity of normal monkeys is 2–9 cpd and increases with age to a maximum of 20–30 cpd by 50 weeks of age. The first 9 weeks of life is the critical period in which short-term monocular-eyelid suturing can cause severe amblyopia in rhesus monkeys. Our data demonstrates the first month of life is more sensitive to visual-pattern deprivation than the second month of life. The visual spatial acuity of the eye deprived during the first month peaked between 0.2–2.0 cpd by 1 yr, whereas the spatial acuity of the eye deprived second peaked between 5–10 cpd by 1 yr.
Key words: strabismus, nystagmus, amblyopia, visual deprivation, monkey development

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