Electroretinograms and Visual Evoked Potentials in Long-Term Monocularly Deprived Cats

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The effects of long-term monocular lid-suture deprivation on visual-evoked cortical potentials (VEPs) and flash- and pattern-evoked electroretinograms (FERGs and PERGs, respectively) were assessed in the cat. VEPs were virtually eliminated when recorded with the deprived eye, indicating that the lid suture produced a severe amblyopia in that eye. In contrast, FERGs and PERGs were more similar for both deprived and nondeprived eyes and comparable to those recorded in normal animals. The current findings demonstrate that long-term deprivation (3-4 yr) does not produce systematic changes in the electroretinogram. Invest Ophthalmol Vis Sci 31:1405-1409, 1990

Cats reared with monocular lid suture exhibit severe deficits in spatial and temporal sensitivity. Neural correlates of this stimulus-deprivation induced amblyopia are manifested at the level of the dorsal lateral geniculate nucleus and visual cortex (for reviews see References 1, 2). Not surprisingly, visual-evoked cortical potentials (VEPs) are attenuated for the deprived eye.3,4

Although deprivation clearly influences the development of cells in the geniculate and cortex, retinal deficits have not been reported in monocularly deprived cats. Sherman and Stone5 found no effect of monocular lid suture on the number, size, density, or conduction latency of ganglion Y-cells recorded in the retinas of animals that showed severe deficits in geniculate Y-cells. Kratz et al6 reported no loss in spatial or temporal resolution in the deprived eyes of monocularly lid-sutured cats.

In these studies, a relatively short period of deprivation (4-18 months) did not cause significant changes in retinal functioning, but did produce deficits in visual cortex. The purpose of the current study was to assess the effects of long-term (3-4-yr) lid suture on retinal and cortical activity by measuring flash- and pattern-evoked electroretinograms (FERGs and PERGs, respectively) and VEPs.

Materials and Methods. Animals: Two cats (C and F) were born and reared in the animal facility (Providence, RI, and St. Louis, MO). Each cat had the lid of one eye sutured shut 5-7 days after birth. The left eye of cat C remained closed for 46 months and was opened 7 months prior to the first recording session. The right eye of cat F remained closed for 34 months and was opened 13 months prior to recording. Recordings were made over a period of 18 months.

Stimulus generation: Pattern stimuli were generated by a Picasso Image Synthesizer (Innisfree, Cambridge, MA) on a cathode ray tube (CRT) display (Hewlett-Packard, Colorado Springs, CO, 1321B, P-31 phosphor) that subtended 44 x 31° of visual angle at a viewing distance of 48 cm. Patterns were vertical sinusoidal gratings that were varied in spatial frequency from 0.12 to 2.0 c/deg. Contrast, defined as the difference between the maximum and minimum luminance divided by the sum, was 0.34. Mean luminance of the display was 2.9 cd/m².

Flash stimuli were generated by a strobe light (Stroboslash, Type 1539-A, General Radio, Concord, MA) directed through a diffuser at a uniform white field the same size and viewing distance as the video display. The manual setting on the strobe light was used to vary the intensity of the flash between three levels: low, medium, and high intensity.

Preparation and recording procedures: VEPs: VEPs were recorded from area 17 from an awake, unanesthetized, restrained cat. Aseptic surgery was performed on anesthetized cats before training and recording sessions began. For details concerning surgery and the harness and apparatus used to restrain the cat, please refer to Baro and Lehmkuhle.7 All procedures were in compliance with the ARVO Resolution on the Use of Animals in Research.

A stainless steel screw secured in the skull over P4 at the midline served as an epidural electrode for cortical recordings. The reference electrode was placed over the frontal lobe approximately 5 mm lateral from the midline. The electroencephalogram (EEG) signal was amplified 10,000 times and band-pass filtered from 0.1 to 100 Hz. Stimulus presentations, on-line digitizing of potentials, artifact rejection, and data storage and analysis were controlled by an Apple Ile computer (see Reference 8). Each VEP record was recorded...
the average of 100 sweeps. The transient stimulus was turned on 5 ms after the beginning of each sweep and turned off after the last sample in the sweep was recorded. Sweep duration was 236 ms (i.e., a sampling rate of 1084 Hz). Approximately 1 sec elapsed between consecutive sweeps.

A video camera was used to observe the cat during recording, and sweeps were initiated only when the cat was attending to the display. Pureed baby food was dispensed periodically between sweeps to assist in maintaining the cat's attention in the direction of the display.

**Electroretinograms:** FERGs and PERGs were measured for the deprived and nondeprived eyes of each cat. Cats were anesthetized with an intramuscular injection of a mixture of ketamine hydrochloride (25 mg/kg) and xylazine HCl (2.5 mg/kg) during recording in order to minimize eye movements and their contamination of recordings. Additional injections were administered throughout the recording session as needed to maintain the level of anesthesia, which was indexed by the presence of eye movement artifacts in the recordings. The harness and apparatus used to secure the cat were the same as above.

Once the cat was anesthetized, each eye was dilated and accommodation was paralyzed with atropine. Alcaine (Alcon) solution was instilled to anesthetize the cornea and a corneal lubricant applied to prevent corneal abrasions. A clear contact lens with an implanted gold wire (Jet Electrode, Life-Tech, Inc., Houston, TX) was placed on the surface of one eye; the cat then was refracted and the appropriate specta-
icle lens placed in front of the eye. The lid of the other eye was closed and covered with soft, opaque material. Needle electrodes were inserted under the skin near the lateral canthus and posterior along the midline and served as reference and ground, respectively.

The corneal recordings were amplified 10,000 times (0.1–100 Hz) to generate PERGs or 2000 times (0.1–3000 Hz) to measure FERGs. Each PERG record was the average of 200 sweeps and each FERG record was the average of 10 sweeps. Artifacts were automatically rejected to prevent their addition into the accumulating waveform. Approximately 1 sec elapsed between consecutive sweeps.

Sweep duration for the recording of PERGs was 1072 ms (ie, a sampling rate of 239 Hz). The grating stimulus was counterphased in a squarewave fashion at 2 Hz (ie, 2 reversals during each sweep). Sweep duration for the recording of FERGs was 236 ms (ie, a sampling rate of 1084 Hz). The flash occurred 27 ms into the sweep.

Results. Cortical evoked potentials: Averaged VEP waveforms recorded from the deprived and nondeprived eyes of cat F at five spatial frequencies are shown in the top panel of Figure 1. Potentials produced by stimulation of the nondeprived eyes are comparable in amplitude and latency to those recorded from normal animals. As previously reported for normal animals, the bottom panel of Figure 1 shows that amplitude of the N1–P1 components tended to be largest at intermediate spatial frequencies (0.25 c/deg), whereas their latency increased with increasing spatial frequency. Components could not be identified consistently in potentials recorded during stimulation of the deprived eyes. Only at 0.25 c/deg could a small potential be discerned in the deprived eye records.

Electroretinograms: FERGs recorded from the deprived and nondeprived eyes of cats C and F at three intensity levels are shown in Figure 2. As is evident in the records, no differences were observed between the deprived and nondeprived eyes. In addition, FERGs recorded from these monocularly deprived animals are comparable to those recorded from normal animals.

Sample PERG waveforms recorded from the deprived and nondeprived eyes of cat C at four spatial frequencies are shown in Figure 3. PERGs were recorded in five separate recording sessions over a period of five weeks. The amplitude and phase of the second harmonic (4 Hz) was estimated by fast Fourier transform (FFT) analyses for each PERG waveform. In Figure 4, the average amplitude and phase and associated variabilities are plotted as a function of spatial frequency for the deprived and nondeprived eyes of each cat. Analyses of variance were performed on the repeated amplitude and phase measures for each cat.

Both amplitude (F(3,12) = 5.027, P = 0.017) and phase (F(3,12) = 8.025, P = 0.003) tended to decrease with increasing spatial frequency for cat C. Only phase changes (F(3,12) = 3.842, P = 0.038) across spatial frequency were significant for cat F. There were two effects of deprivation revealed with this analysis of PERG waveforms, which were different.
across cats. For cat C, the PERG amplitude was attenuated for the deprived eye relative to the nondeprived eye at higher spatial frequencies (spatial frequency × deprivation interaction, F(3,12) = 7.21, P = 0.005). For cat F, phase values were longer for the deprived eye (F(1,4) = 8.43, P = 0.043). This differ-

Fig. 3. PERGs at each spatial frequency for the nondeprived and deprived eyes of cat C. Stimulus contrast was 0.34. Squarewave modulation of the pattern began 1275 msec before the beginning of each 1072-msec sweep. Two pattern reversals occurred during each sweep. Each record is the average of 200 sweeps.

Fig. 4. Fourier amplitude and phase of the second harmonic of PERGs as a function of stimulus spatial frequency for cats C and F. Each data point represents the mean of five PERGs, recorded on different days. Error bars indicate ± one standard error of the mean. Open symbols represent data obtained from the nondeprived eye and filled symbols represent data obtained from the deprived eye.
ence between the average phase of the deprived and nondeprived eyes corresponds to a difference in latency of about 15 msec at 0.12 c/deg. All other effects were not statistically significant.

Discussion. The VEP records indicate that the long periods of monocular lid suture produced a severe functional amblyopia in the deprived eye of each cat. The attenuation of the VEP produced during stimulation of the deprived eye was larger than that reported in earlier studies, and was due probably to a longer period of deprivation (3-4 yr of deprivation in the current study vs 9-12 months of deprivation in earlier studies).

Although 3-4 yr of monocular lid suture produced a formidable reduction in the amplitude of the VEP, there were no consistent effects of the monocular lid suture on the FERG and PERG. FERGs were nearly identical for the deprived and nondeprived eyes and comparable to those obtained in normal animals. Some changes in PERGs were observed for the deprived eye, but those changes were small and inconsistent relative to the pervasive effects of monocular lid suture on the cortical VEP.*

The current results are consistent with earlier observations that deficits in the dorsal lateral geniculate nucleus and in the visual cortex occur after rearing with monocular lid suture, whereas retinal ganglion cells in the deprived eye seem to function normally. Moreover, the current data indicate that, even after long periods of monocular lid suture, the cat's retina remains relatively immune to the deleterious effects of visual deprivation. This pattern of results in cats suggests that the PERG, as an electrodiagnostic tool, would not delineate the severity of an amblyopia.

Key words: electroretinogram, visual evoked potential, visual deprivation, cat

References

*The large field size used in the current study precludes an assessment of any localized retinal deficits that may have been produced by the deprivation. It is conceivable that more consistent effects of deprivation on the PERG may be revealed with the use of smaller field sizes.