Experimental allergic optic neuritis in guinea pigs: preliminary report

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An experimental model for acute allergic optic neuritis was produced in adult strain 13 guinea pigs by sensitization with isogenic spinal cord emulsion in complete Freund's adjuvant. These animals exhibited two distinct clinical patterns: (1) "retrobulbar optic neuritis," with a diminished pupillary response to light despite a normal fundus, and (2) "neuroretinitis," with a diminished pupillary response associated with hyperemia and swelling of the disc and juxtapapillary retinal edema. Histopathologic study of those animals with "retrobulbar neuritis" revealed that some had no abnormalities in the optic nerve or chiasm, but showed foci of mononuclear cell infiltration in the brain. Others had a mononuclear cell infiltration localized to the retrobulbar portion of the optic nerve and chiasm with multiple foci of axial and periaxial demyelination. Similar pathologic changes were present in the animals with "neuroretinitis," but the lesions were located just behind the lamina scleralis. These animals also exhibited marked swelling of the axons at the lamina retinalis. On examination by light microscopy, the alterations in the region of optic nerve head appeared characteristic of papilledema.

Key words: allergic optic neuritis, neuroretinitis, retrobulbar optic neuritis, demyelination, optic nerve, strain 13 guinea pigs.

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Experimental allergic encephalomyelitis (EAE) has been studied extensively in the past and has been considered by some as an excellent model for examining acute demyelinating disease processes affecting the central nervous system. Several workers have observed optic neuritis, retinal vasculitis, and uveitis in guinea pigs, rabbits, and monkeys affected with EAE. Pathologic changes at the optic nerve head, however, have not been described in the guinea pig, which is considered by some to provide a better animal model for multiple sclerosis.

The purpose of this report is to describe the demyelinating optic neuritis in guinea pigs, to correlate the clinical findings with...
pathologic changes, and to detect effects of demyelination on the optic nerve head. Future reports will deal with the mechanisms of demyelination and ultrastructural changes at the optic disc in experimental allergic optic neuritis (EAON)—the optic neuritis that occurs in association with EAE.

Materials and methods

Twelve adult strain 13 guinea pigs, weighing 360 to 420 gm., were inoculated intracutaneously at five sites in the nuchal region with 0.5 ml. of an emulsion containing 0.25 ml. of a 50 percent spinal cord suspension. This suspension was prepared from the spinal cord of the same strain of guinea pigs, emulsified with 0.25 ml. of Arlacet-Bayol (Freund's adjuvant) incorporating 2.5 mg. of killed, dried Mycobacterium tuberculosis. The procedure was carried out under ketamine anesthesia and aseptic conditions.

In conducting the research described in this report, the investigators adhered to the Guide for laboratory animal facilities and care, as promulgated by the Committee on the Guide for Laboratory Animal Facilities and Care of the Institute of Laboratory Animal Resources, National Academy of Sciences–National Research Council.

For controls, four animals of the same strain were similarly inoculated with 0.5 ml. of complete Freund's adjuvant (1 ml. of Arlacet-Bayol incorporated with 10 mg. of killed, dried M. tuberculosis). All animals were observed daily, especially for gait disorders and for abnormal pupillary response to light. The optic disc and surrounding retina were examined by indirect ophthalmoscopy.

Twelve to 14 days following inoculations with spinal cord emulsion, the experimental group of animals developed disturbances of gait. About 24 to 48 hours after the onset of these disturbances, the animals were anesthetized with pentobarbital; the eyes were enucleated with 3 to 4 mm. of optic nerve attached and immediately fixed in a 2 percent solution of glutaraldehyde. The eyes from four control animals were also enucleated 13 to 15 days after inoculation and were similarly fixed. All animals were then put to death by an intraperitoneal injection of 2 ml. of pentobarbital solution, after which the brain, remaining optic nerve, and optic chiasm were removed from the cranium and fixed in a 2 percent solution of glutaraldehyde. The optic discs, optic nerves and chiasm, and about a 2 mm. segment of retina with choroid of each animal...
Fig. 2. Mononuclear cell infiltration is present in the periaxial region of the retrolaminar portion of the optic nerve. There is associated demyelination. Mononuclear cell infiltration is also seen within the pial sheath. (Epon section stained with toluidine blue, x165; AFIP Neg. 76-5831.)

were embedded in Epon. Sections 1 μm thick were studied after being stained with toluidine blue. Cerebral lesions were sectioned in paraffin, stained with hematoxylin and eosin, and examined by light microscopy.

Results

The 12 guinea pigs inoculated with spinal cord emulsion showed a loss of weight 8 days after the injection; these animals developed gait disorders and excessive salivation and mastication 4 to 6 days later. Among these animals, six showed an absence of the pupillary reflex to light bilaterally, but indirect ophthalmoscopy revealed no abnormalities. They were classified clinically as having "retrobulbar neuritis," whereas in one other animal a pupillary response to light was absent only in the left eye, which had developed an elevated hyperemic disc and edema of the juxtapapillary retina. This eye was classified as having "neuroretinitis." Five experimental and the four control animals had no pupillary abnormalities, and the controls remained healthy.

Among the seven guinea pigs with abnormal pupillary reflexes, pathologic changes in the optic nerves were found in four eyes—three in the right eye and one in the left eye (the eye with the disc and retinal changes). The other eyes from these animals, as well as the eyes from the remaining five animals that had been sensitized to spinal cord emulsion, showed no lesions in the optic nerves or chiasms. Sections of the brains of all 12 guinea pigs, however, revealed foci of mononuclear cell infiltration. The four control guinea pigs were free of these changes.

Histopathologically, the optic neuritis in the four eyes revealed focal infiltration of mononuclear cells in the retrobulbar part of the optic nerve. These cells were present predominantly within the pial septa and extended into the adjacent axonal bundles. Occasionally, perivascular distribution of mononuclear cells was also noted. At the site of mononuclear cell infiltration, myelin staining was absent. A few axons with stainable myelin were irregularly swollen.
Fig. 3. Well-developed papilledema with marked swelling of the nerve fibers at the optic disc, obliteration of the optic cup, and lateral displacement of the juxtapapillary retina. (Toluidine blue, x180; AFIP Neg. 76-5834.)

The process of demyelination involving the nerve was focal, with a periaxial distribution in some (Fig. 2) and an axial pattern in others. In two eyes (one eye classified clinically as having neuroretinitis and another eye classified as having retrobulbar neuritis) the swollen axons at the lamina retinalis had obliterated the optic cups and produced lateral displacements of the juxtapapillary retina (Fig. 3). The peripapillary axons were more swollen than the axons situated in the central region of the disc. Mild, focal, mononuclear cell infiltration was present in the choroid as well as in the meninges in all four eyes.

Discussion

In our study of EAON, two distinct types of optic neuritis were observed clinically. One group of eyes, classified as having retrobulbar neuritis, revealed histopathologic alterations similar to the changes described by previous investigators, and these lesions were localized to retrobulbar, orbital, and intracranial portions of the optic nerve exhibiting foci of demyelination (Fig. 2). Mononuclear cell infiltration was seen with disrupted myelin sheaths and with phagocytosis of myelin fragments. Moderate axonal swelling was noted at the vicinity of the inflammation. The other group of eyes, exhibiting the features of neuroretinitis, however, revealed foci of demyelination and inflammation just posterior to the lamina scleralis. In these eyes, swelling in the lamina retinalis was observed with obliteration of the physiologic cup and lateral displacement of the juxtapapillary retina (Fig. 3). These changes are typical of those seen in papilledema produced by other pathologic processes. Such axonal changes at the lamina retinalis have not been described in previous papers on EAE. The axonal enlargement in the lamina retinalis was more marked than that noted at the lamina choroidalis, and this swelling extended into the juxtapapillary nerve fibers. The eye classified clinically as having neuroretinitis, however, histopathologically revealed no inflammatory
cell infiltration in the disc anterior to the lamina cribrosa, the juxtapapillary retina, or the vitreous.

Despite the minor differences in pathologic changes between EAON and multiple sclerosis, the focal demyelination with mononuclear cell infiltration observed in the optic nerve of these animals offers an experimental model for the study of pathogenetic mechanisms of demyelinating diseases in the optic nerve of human subjects.

REFERENCES