DEGENERATION OF THE INNER NUCLEAR LAYER OF THE RETINA FOLLOWING LESIONS OF THE OPTIC NERVE

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NEURONAL PATHWAYS AND FUNCTIONS have been studied for many years by observing the process of neuronal degeneration. Trans-synaptic (transneuronal) degeneration has been observed in the nervous system when degeneration of the injured neuron not only proceeds to the synapse but continues to include the contiguous neuron. Such degenerative changes may be observed in both anterograde and retrograde directions. Anterograde trans-synaptic degeneration continues centrally after there has been a loss of the afferent neuronal supply. In the visual system, anterograde degeneration of the lateral geniculate body has been documented following lesions of the optic nerve and retina.[3,4,6,7,15,24,25] This degenerative process may be species specific[23] and may vary with the age of the animal. Trans-synaptic retrograde degeneration has been noted in the ganglion cells and in the inner nuclear layer of the retina in animals and man. Atrophy of the optic nerves and tracts resulting from occipital lesions has been described by Moeli,[26] Dejerine,[8] Nissel von Mayendorf,[29] Fledelius,[9] Haddock and Berlin,[16] Ganser,[10] Kluver,[20] and Van Buren.[33,34] Van Buren has reported evidence of retrograde trans-synaptic degeneration of the inner nuclear layer after lesions of the optic nerve fibers in monkey and man. These changes were described as "cavitary" (cystic) degeneration of the inner nuclear layer. Atrophy of the inner nuclear layer was also observed by Haschke and Sickel[17] in a patient who had a tumor of the optic nerve for five years.

REVIEW OF THE LITERATURE

Schultze, in 1872,[32] observed the loss of ganglion cells after severance of the optic nerve. Nissl[28] established that a nerve's cells degenerate

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following injury to its axon. James[19] noted degeneration to the ganglion cells in the retina of rabbits after sectioning the optic nerve three millimeters behind the globe. These experimental animals demonstrated chromatolysis of the ganglion cells 10 days after section of the optic nerve; and after 20 days there was disorganization and fragmentation of the ganglion cells. Leinfelder[22] noted that ganglion cells of the retina react to injury by retrograde degeneration in proportion to the extent of the injury and to the distance of the injury from the nerve cells. Degeneration of the ganglion cells of the retina in a patient with chromophobe adenoma of the pituitary was reported by Gartner.[11] Kupfer[21] observed retrograde degeneration of the retinal ganglion cells in the eyes of three patients in which lesions of the chiasm had been present for less than one year. He concluded that the reaction of retinal ganglion cells to injury was similar to that of ganglion cells elsewhere in the central nervous system. In contrast, Greenfield[14] proposed that secondary changes (chromatolysis) do not occur in retinal ganglion cells after axonal injury, but that the ganglion cells undergo atrophy without chromatolysis.

Retrograde trans-synaptic degeneration of the optic nerve and ganglion cells had been noted in the primate visual system after occipital lesions. Kluver[20] reported a rhesus monkey in which the left occipital lobe was extirpated. Four years later there were defects in the staining properties in the ganglion cells of the corresponding halves of the retina. Van Buren demonstrated loss of ganglion cells of the retina four years following an occipital lesion in a monkey.[34]

Clinically, pallor of the optic disc occurs following occipital-lobe injuries of long standing (Walsh[35]). Haddock and Berlin[16] described a veteran who sustained a shotgun wound to both occipital lobes with early atrophy appearing three-and-one-half years later and complete atrophy of the optic nerves after five years. In three human cases with lesions of the cerebral hemisphere there was trans-synaptic degeneration of the ganglion cells (Van Buren[33]). In these cases, a quantitative loss of ganglion cells was noted as early as six and 13 months. Van Buren's detailed quantitative work has given objective support to a controversial topic (Walsh,[35] Polyak[30–31]). Optic atrophy and loss of the ganglion cells in the retina were present in five autopsy cases with destructive lesions of the hemisphere of over four years' duration (Gills[12]). Artifacts and difficulties in interpretation of histology in autopsy eyes prevented definitive evaluation of changes in the optic nerve and ganglion cells after hemispheric lesions, but the findings were generally in agreement with those of Van Buren.

Retrograde trans-synaptic degeneration of the inner nuclear layer after lesions in the optic nerve fibers has been described in monkey and man by Van Buren.[34,35] In two monkeys, the optic chiasm was sectioned 20 months before death; in two other monkeys, the optic tracts were sectioned 48 months before death (photomicrograph, Figure 1). "Cavitary" (cystic) degeneration was present in the inner nuclear layer of the retina of these eyes. Van Buren also noted degeneration of the inner nuclear layer of the retina in a patient in whom a chiasmal lesion had been known for 20 months before death, and in whom a surgical lesion had been made at the chiasm 18 months before death. Haschke and Sickel[17] followed a patient with a tumor of the optic nerve of a non-seeing eye for five years. Multiple ERG tracings were essentially normal. There was atrophy of the inner nuclear layer of the retina of the eye (Figure 2).

These changes observed by Van Buren and Haschke in the inner nuclear layer of the human retina warrant further investigation and documentation. In such a study, the following questions need to be considered.

1. Is there a decreased cellularity in the inner nuclear layer of the eyes after lesions involving the optic nerve fibers?
2. What are the types of cells which degenerate in the inner nuclear layer?
3. What is the time interval required for the degenerative process in the inner nuclear layer?
4. What are the various manifestations and ultimate extent of the degeneration?
5. What is the explanation for the elevated ERG responses in patients with sectioned nerves?

From a review of the literature on trans-synaptic degeneration, it is apparent that the phenomenon may occur in any neuronal system, including the visual system. However, retrograde trans-synaptic degeneration of the inner nuclear layer remains a controversial topic. Recognition of the retinal degenerative changes is difficult unless a quantitative study is carried out. The lack of quantitative studies makes it difficult to evaluate the findings of previous observers. The purpose of this investigation is to quantitate the cells in the inner nuclear layer of the retina in eyes with associated lesions of the optic nerve or chiasm and in normal eyes. Quantitation is accomplished by counting the number of cells in the inner and outer nuclear layers over specific areas and in definite meridians. This quantitative assessment will add objectivity to findings which previously had been based on impressions.

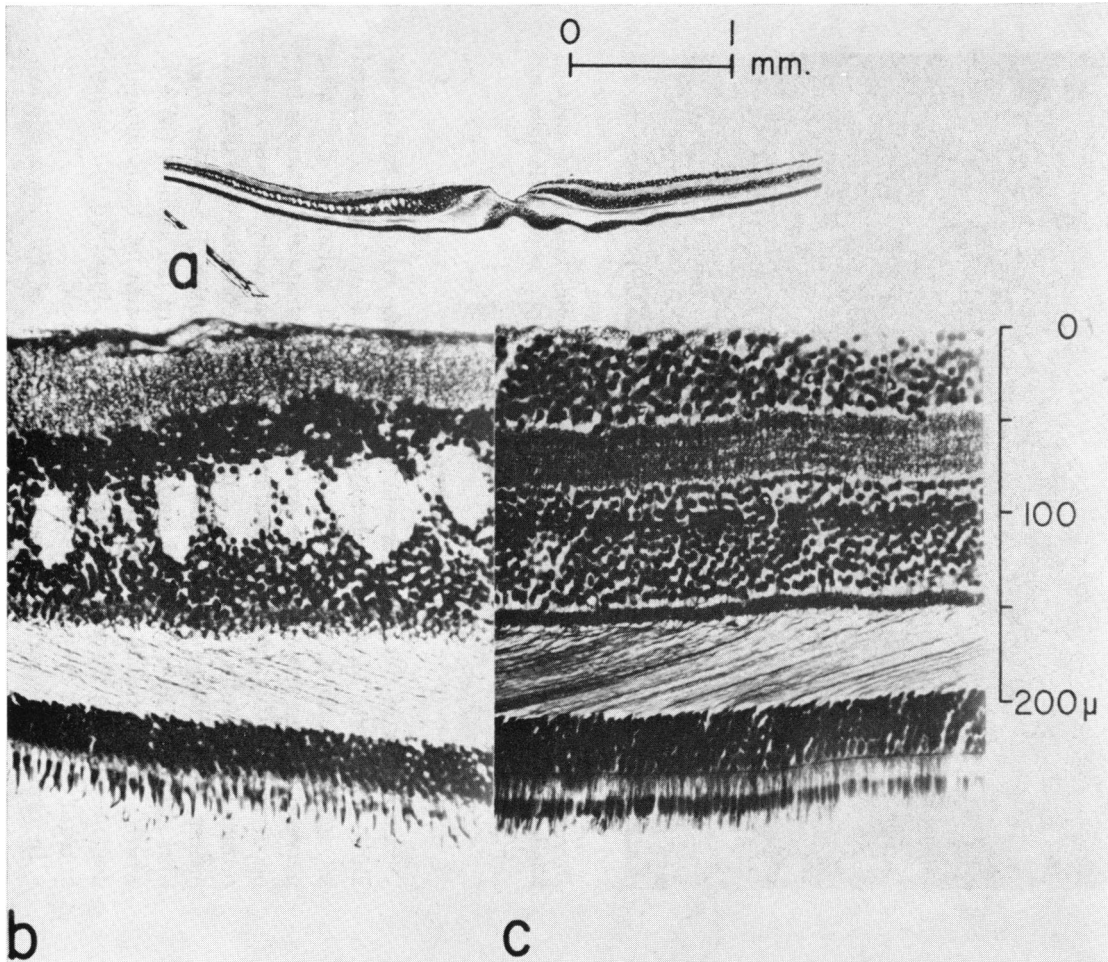

FIGURE 1

Horizontal section through the fovea of the right eye 20 months after section of the chiasm. In (a) the margin of the papilla is at the extreme left with loss of ganglion cells and cavitation in the bipolar layer of the medial side of the fovea. In (b) and (c) sections at higher power on the medial and lateral side of the fovea are shown. (a) cresyl violet, (b) and (c) phosphotungstic acid-hematoxylin stains (Van Buren[34]).

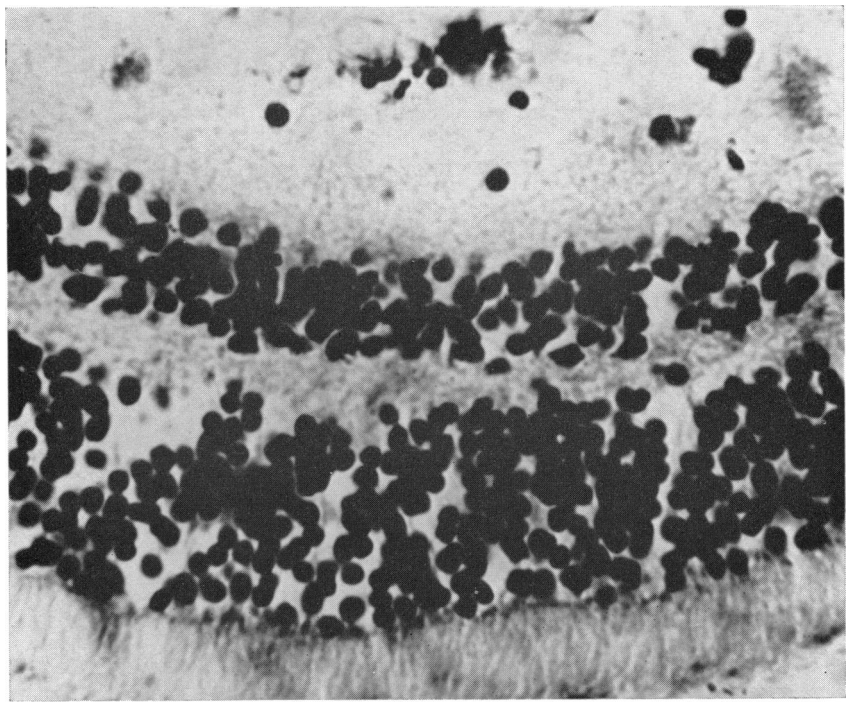

FIGURE 2

Decreased cellularity of the inner nuclear layer of the retina in a human eye with blindness for five years from a meningioma of the optic nerve (Haschke and Sickel[17]).

METHOD

In a review of 459 autopsies where ocular tissue was obtained at the Duke University Medical Center, 50 cases were found with associated intracranial lesions. Seven of the 11 cases with lesions of the optic nerve at or near the chiasm are presented to illustrate the changes that occur in trans-synaptic degeneration of the inner nuclear layer of the retina. (In the other four eyes, autolysis, other concurrent ocular disorders, and oblique sections prevented quantitation.) Sections from four additional eyes obtained surgically by one of the authors (Wadsworth) are included to help illustrate the process of trans-synaptic degeneration. In three cases the lesion primarily involved the chiasm; in eight cases the optic nerve was primarily involved.

In order to objectively assess the cellularity of the inner nuclear

layer, the cells were counted and the data statistically analyzed. Counts were made of 120 micron segments at meridians of 2, 4, 6, 10, and 12 mm. temporal to the edge of the disc and 2, 6, and 10 mm. nasal to the edge of the disc. The counts were made under 860 magnifications with the use of a Howard disc. In this study, approximately 200,000 cells were counted. The histologic sections were 10 microns in thickness. In each eye (11 eyes with lesions and five normal eyes), the cells in the inner nuclear layer of each meridian were counted five times and averaged. These averages were compared to the average counts of the inner nuclear layers from five normal eyes in the same meridian. In some eyes presented, the section was not exactly in the plane through the macula and disc; however, this did not appear to significantly affect the total cell count. The stains varied. The vascular supply was intact in all eleven eyes studied in detail. All counts recorded were by one author (Gills) and have been checked by other observers. Five separate counts were performed.

CASE FINDINGS

All 11 cases will be individually presented with accompanying photomicrographs for seven of them.

CASE 1

A twelve-year-old male.

DIAGNOSIS. (1) Craniopharyngioma present for six years and partially resected two years prior to death. (2) Cut optic nerve, right eye, two years before death.

CLINICAL HISTORY. Six years prior to death, the patient began to experience severe headaches that were intermittent in nature, lasting about two days every month. His vision decreased three to four years prior to death. Three years before death he had to stop attending school because of inability to see gross objects. First Duke Outpatient Visit: April 17, 1940. Vision: light perception in both eyes. There was atrophy of both optic papillae. X-rays revealed erosions of posterior clinoids, enlargement of sella turcica, widening and separation of sutural lines. On May 16, 1940, Dr. Barnes Woodhall performed a right frontal craniotomy, partially resecting a large cystic craniopharyngioma. The right optic nerve was severed to obtain better exposure. Following surgery, radiation was given. The postoperative vision was: right eye, no light perception; left eye, light perception. Optic atrophy was present bilaterally. The retinal vessels were intact. The patient died from recurrence of the tumor on June 23, 1942.

GROSS PATHOLOGY. A large (5 cm. $\times$ 4.5 cm. sagittally) craniopharyngioma obliterated the chiasm and optic nerve (Figure 3).

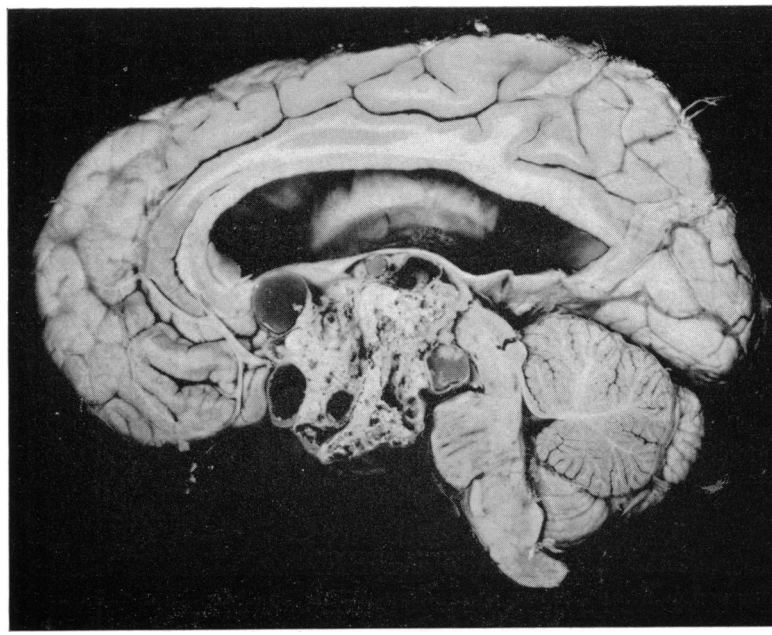

FIGURE 3. CASE 1.
Craniopharyngioma (5 cm. × 4.5 cm.) occupying the chiasmal area with obliteration of the chiasm and optic nerves.

MICROSCOPIC PATHOLOGY. Right eye: optic atrophy and loss of ganglion cells were present. Decreased cellularity and cystic changes were present in the inner nuclear layer of the retina. The decreased cellularity and cystic changes were particularly evident in the nasal retina (Figures 4–6).

CELL COUNT (Average of five counts). *Temporal*: 2 mm., 103; 4 mm., 109; 6 mm., 104; 10 mm., 65; 12 mm., 49. *Nasal*: 2 mm., 132; 6 mm., 84; 10 mm., 50.

Total number cells counted (five counts)	3471
Normal mean for all slides counted (five normal eyes)	6329
Probability by chance	.05

COMMENT. The decreased cellularity of the inner nuclear layer is compatible with trans-synaptic retrograde degeneration.

CASE 2

A forty-three-year-old female.

DIAGNOSIS. Adenocarcinoma, pituitary.

CLINICAL HISTORY. Eight years before death the patient stopped menses and had headaches, and for over two years had significant loss of vision. For

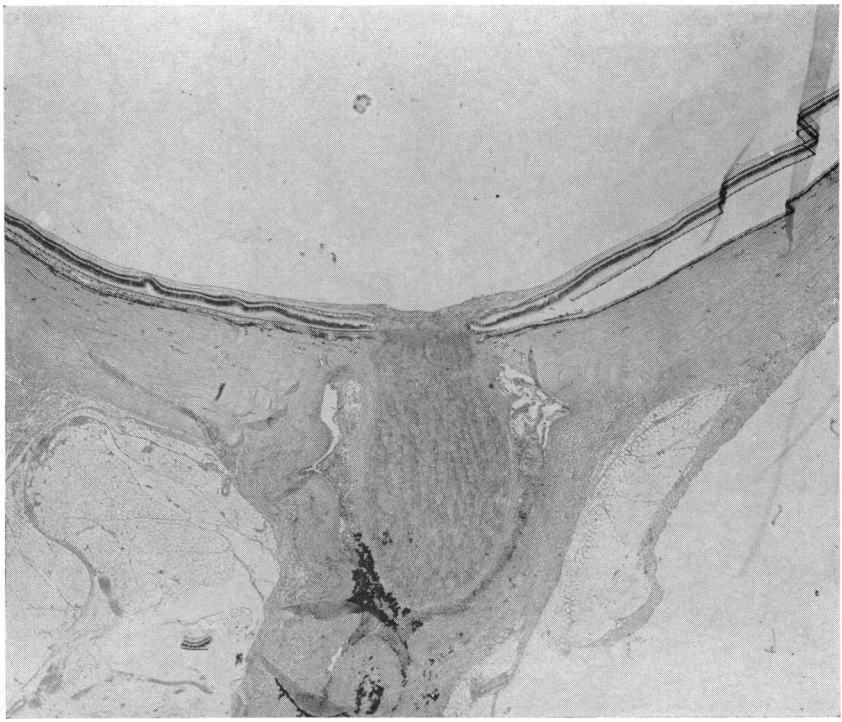

FIGURE 4. CASE 1.
Optic atrophy, loss of ganglion cells, and decreased cellularity of the inner nuclear layer is present.

18 months before death the patient was blind in her left eye. X-rays revealed complete destruction of the posterior clinoids and a large sella turcica. The patient died after an exploratory craniotomy.

GROSS PATHOLOGY. A large adenocarcinoma of the suprasellar area with compression of the optic nerve.

MICROSCOPIC PATHOLOGY. Left eye: optic atrophy, decreased number of ganglion cells, and loss of cellularity of the inner nuclear layer of the retina were present. The retinal vasculature appeared normal (Figure 7).

CELL COUNT (Average of five counts). *Temporal*: 2 mm., 78; 4 mm., 100; 6 mm., 70; 10 mm., 43; 12 mm., 48. *Nasal*: 2 mm., 69; 6 mm., 68; 10 mm., 42.

Total number of cells counted (five counts)	2580
Normal mean for all slides counted (five normal eyes)	6329
Probability by chance	.02

COMMENT. These findings suggest trans-synaptic degeneration in this eye after a lesion of the chiasm for about two years.

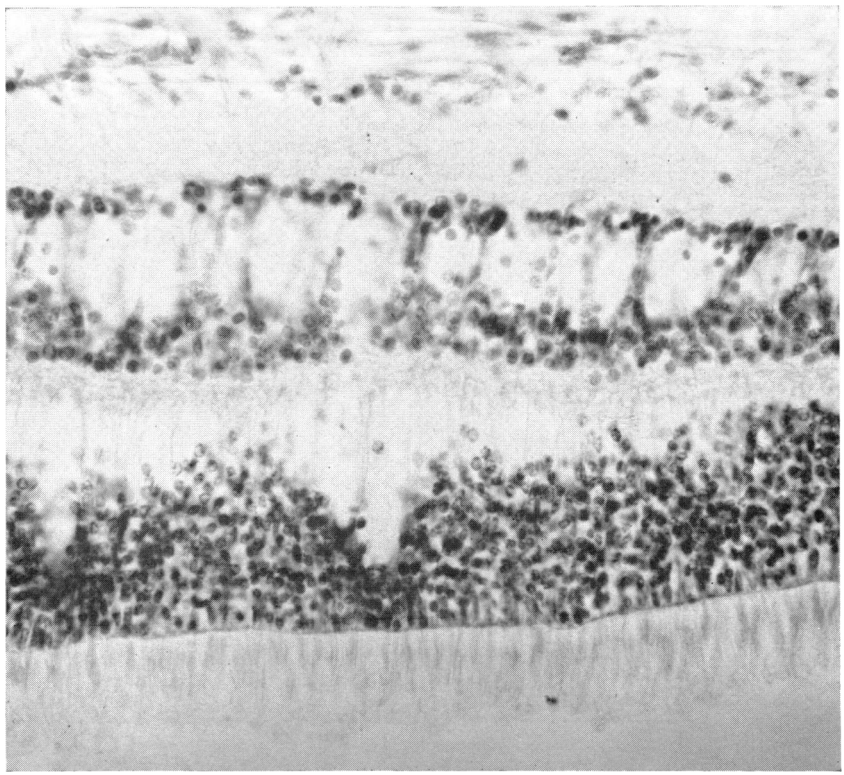

FIGURE 5. CASE 1.

Decreased cellularity with cystic areas which are more marked in the inner nuclear layer and present in the external nuclear layer of the retina between the disc and macula.

CASE 3

A sixty-seven-year-old female.

DIAGNOSIS. Meningioma of sphenoid ridge.

CLINICAL HISTORY. Twenty-one years before enucleation, a sphenoid ridge meningioma was removed along the roof of the right orbit. Optic atrophy in the right eye was "absolute." Recurrent proptosis occurred secondary to orbital meningoma. The retinal arterioles appeared normal.

MICROSCOPIC PATHOLOGY. Optic atrophy and loss of ganglion cells of the retina were present. There was decreased cellularity of the inner nuclear layer. The outer plexiform layer was atrophic (Figure 8).

CELL COUNT (Average of five counts). *Temporal*: 2 mm., 79; 4 mm., 105; 6 mm., 82; 10 mm., 69; 12 mm., 51. *Nasal*: 2 mm., 62; 6 mm., 67; 10 mm., 60.

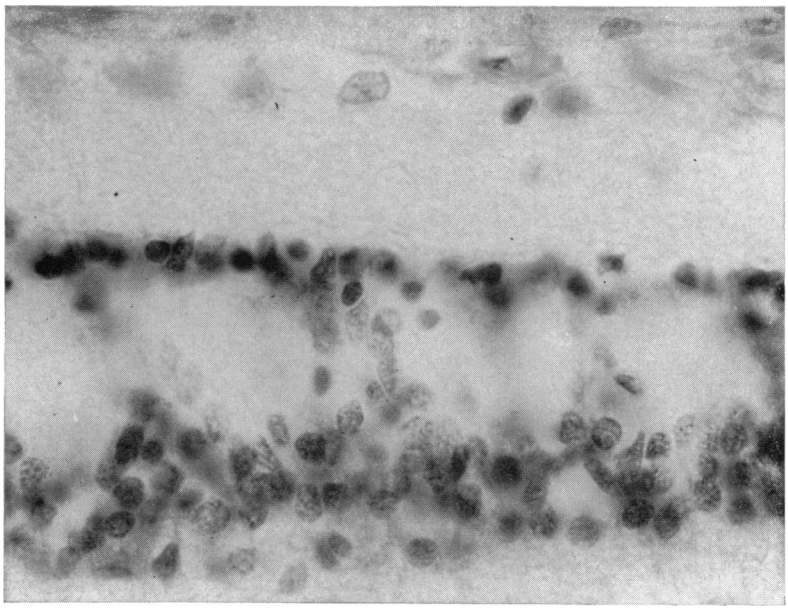

FIGURE 6. CASE 1.
Decreased cellularity and cystic spaces within the inner nuclear layer between macula and disc.

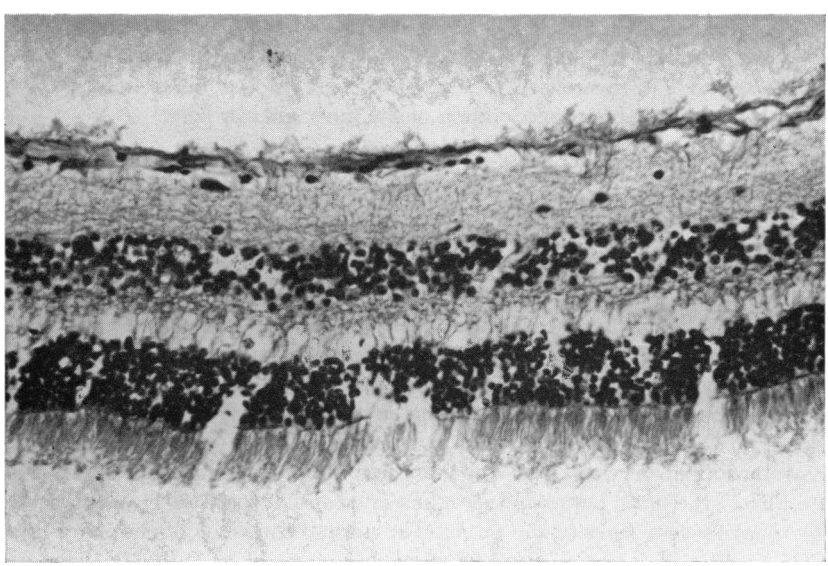

FIGURE 7. CASE 2.
Photomicrograph from mid-distance between disc and macula shows marked loss of ganglion cells and decreased cellularity of the inner nuclear layer.

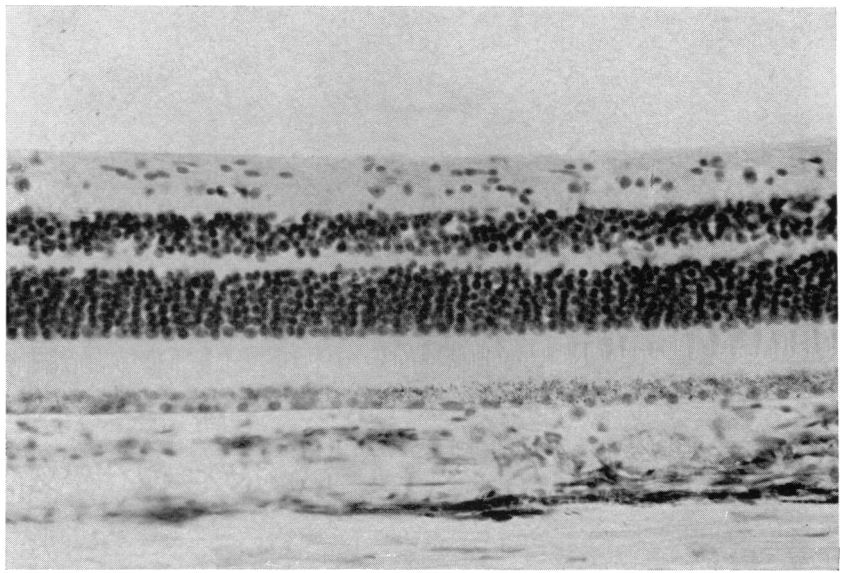

FIGURE 8. CASE 3.
Photomicrograph from 1.5 mm. temporal to disc.

Total number of cells counted (five counts)	2970
Normal mean for all slides counted (five normal eyes)	6329
Probability by chance	.02

COMMENT. The decreased cellularity present in the inner nuclear layer suggests trans-synaptic degeneration of the inner nuclear layer.

CASE 4

A fifty-three-year-old female.

DIAGNOSIS. Meningioma of the optic nerve of the right orbit for 11 years.

CLINICAL HISTORY. Loss of vision of the right eye was known for 11 years and light perception vision was present for nine years. Proptosis of the right eye had also been present for nine years. A right frontotemporal craniotomy was performed with the removal of a meningioma which extended into the orbit. Reoperation with enucleation was performed for progressive proptosis from a recurrence of the meningioma in the right orbit, frontal lobe, and globe.

MICROSCOPIC PATHOLOGY. The vascular supply was intact. There was atrophy of the optic nerve and ganglion cells and a decrease in cellularity of the inner nuclear layer of the retina. The normal thickness of the inner layer (five to six cells) in the posterior pole was reduced to a two-to-three cell thickness. A small melanoma was present in the choroid (Figure 9).

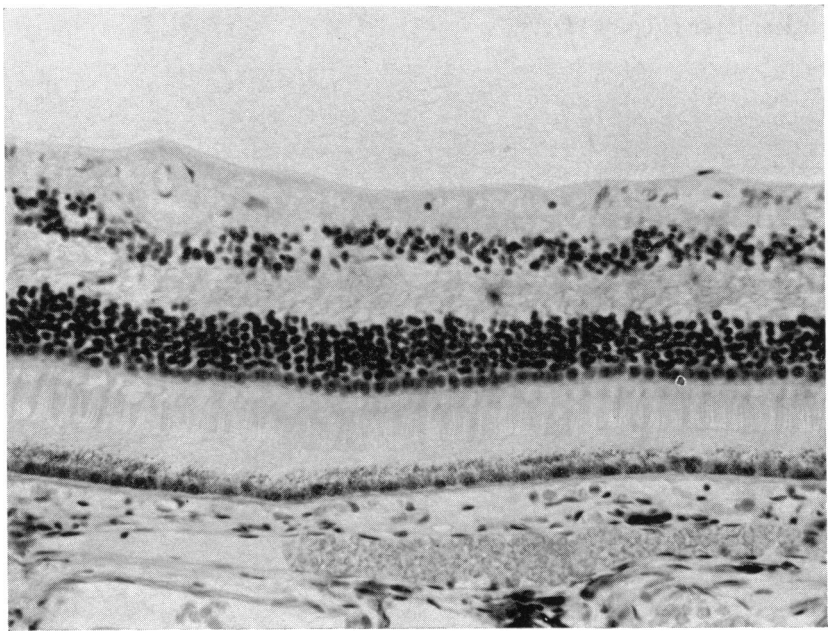

FIGURE 9. CASE 4.
Photomicrograph: area between macula and disc with loss of ganglion cells and decreased cellularity in the inner nuclear layer.

CELL COUNT (Average of five counts). *Temporal*: 2 mm., 66; 4 mm., 71; 6 mm., 86; 10 mm., 64; 12 mm., 25. *Nasal*: 2 mm., 61; 6 mm., 102; 10 mm., 24.

Total number of cells counted (five counts)	2514
Normal mean for all slides counted (five normal eyes)	6329
Probability by chance	.02

COMMENT. These degenerative changes in the inner nuclear layer appear to be secondary to trans-synaptic retrograde degeneration.

CASE 5

A three-year-old female.

DIAGNOSIS. Spongioblastoma of the optic nerve for at least two years.

CLINICAL HISTORY. At age one, the right eye was noted to turn out. At 18 months, the right eye began to proptose forward and downward. The vision was decreased. The right eye became painful at age two and was removed at age three.

MICROSCOPIC PATHOLOGY. The glioma extends into the optic papilla. The vascular supply was intact. The ganglion cells were atrophic. There were

cystic areas in the inner nuclear layer. There was loss of cells in the inner nuclear layer (Figure 10).

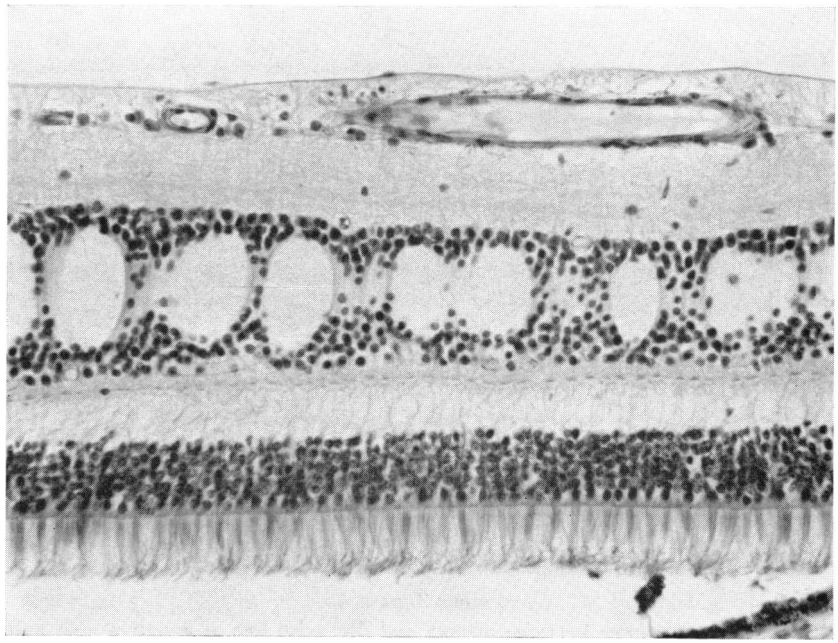

FIGURE 10. CASE 5.
Photomicrograph between macula and disc.

CELL COUNT (Average of five counts). *Temporal*: 4 mm., 167; 6 mm., 119; 10 mm., 56; 12 mm., 42. *Nasal*: 6 mm., 65; 10 mm., 43.*

COMMENT. The cystic areas of the inner nuclear layer were large, probably partly a result of retinal edema or venous engorgement. The decreased cellularity of the inner nuclear layer may represent retrograde trans-synaptic degeneration.

CASE 6

A fifty-three-year-old male.

DIAGNOSIS. Bilateral post-traumatic optic atrophy for 26 years.

CLINICAL HISTORY. Twenty-six years prior to death, the patient suffered a fractured skull in an automobile accident. The patient had sudden, almost complete blindness in both eyes.

MICROSCOPIC PATHOLOGY. Complete atrophy was present in the chiasm, both optic tracts, both geniculate bodies, the optic nerves, and the ganglion

*No counts were made at either 2 mm. temporal or 2 mm. nasal to the edge of the disc.

cells of the retina. There was loss of cells in the inner nuclear layer, particularly in the posterior fundus (Figure 11).

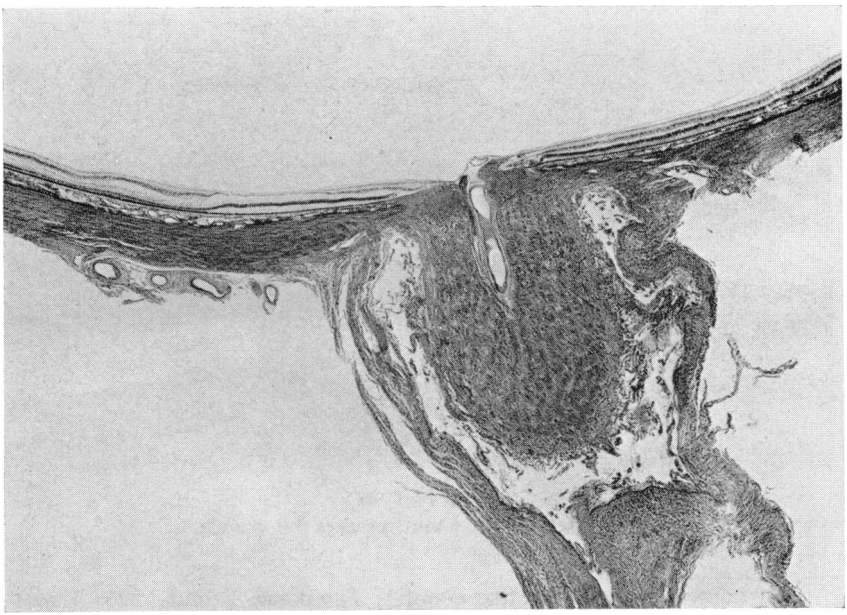

FIGURE 11. CASE 6.
Photomicrograph showing optic atrophy, loss of ganglion cells, and decreased cellularity of the inner nuclear layer.

CELL COUNT (Average of five counts). *Temporal*: 2 mm., 34; 4 mm., 135; 6 mm., 130; 10 mm., 87; 12 mm., 78. *Nasal*: 2 mm., 11; 6 mm., 102; 10 mm., 88.

Total number of cells counted (five counts)	3736
Normal mean for all slides counted (five normal eyes)	6329
Probability by chance	.05

COMMENT. The loss of cells of the inner nuclear layer and in the geniculate bodies represents both retrograde and anterograde trans-synaptic degeneration.

CASE 7

A sixty-two-year-old female.

DIAGNOSIS. Meningioma of left sphenoid ridge for 25 years.

CLINICAL HISTORY. The patient noted progressive proptosis of the left eye and optic atrophy. The left eye had been blind for years.

MICROSCOPIC PATHOLOGY. There was optic atrophy, loss of ganglion cells, and loss of cells in the inner nuclear layer (Figure 12).

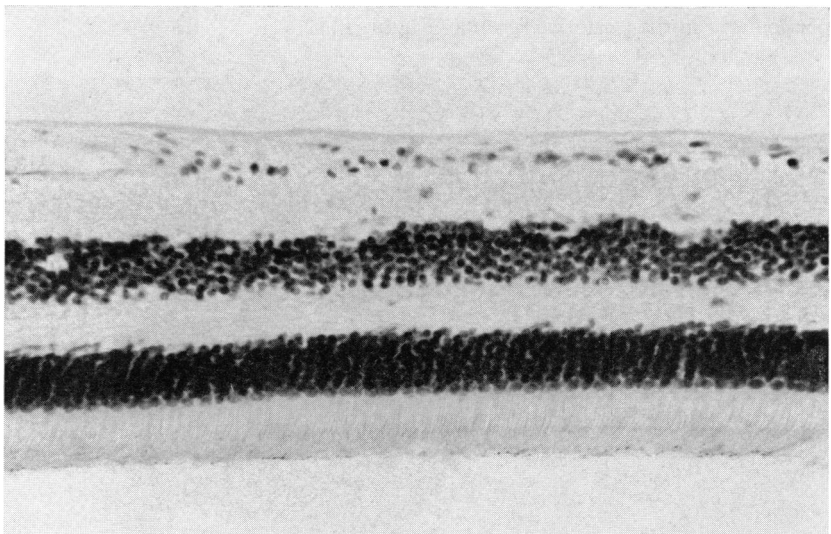

FIGURE 12. CASE 7.
Photomicrograph: 1 mm. nasal to the macula.

CELL COUNT (Average of five counts). *Temporal*: 2 mm., 226; 4 mm., 216; 6 mm., 153; 10 mm., 86; 12 mm., 58. *Nasal*: 2 mm., 122; 6 mm., 107; 10 mm., 81.

Total number of cells counted (five counts)	5368
Normal mean for all slides counted (five normal eyes)	6329
Probability by chance	.20

COMMENT. The loss of cellularity of the inner nuclear layer is probably secondary to trans-synaptic degeneration. The small cystic changes are normal.

CASE 8

A forty-seven-year-old male.

DIAGNOSIS. Chromophobe adenoma of the pituitary.

CLINICAL HISTORY. Optic atrophy of the left eye for years. The patient was known to have progressive decrease in visual acuity in the left eye for eight years and blindness for at least five months. It is also of note that there was mild atrophy of the right disc, but vision in the right eye was 20/25 and the fields were full.

GROSS PATHOLOGY. The posterior clinoids were destroyed; the sella turcica eroded and enlarged to 3–4 cm. There was a 2 × 2 × 4 cm. encapsulated mass present in the left middle fossa in the Sylvian fissure between the left temporal lobe and frontal lobe. It was connected to the hypothalmus by a

1.5-cm. long × .3-cm. diameter stalk. The tumor was continuous with the mass of the pituitary fossa. The left optic nerve was completely displaced and stretched. The tumor had eroded the pituitary fossa and was present in the left sphenoid ridge. The right optic nerve was identified.

MICROSCOPIC PATHOLOGY. There was optic atrophy, a decreased number of ganglion cells, and loss of cellularity in the inner nuclear layer of the left eye.

CELL COUNT (Average of five counts). *Temporal*: 2 mm., 84; 4 mm., 82; 6 mm., 72; 10 mm., 51; 12 mm., 31. *Nasal*: 2 mm., 72; 6 mm., 71; 10 mm., 43.

Total number of cells counted (five counts)	2530
Normal mean for all slides counted (five normal eyes)	6329
Probability by chance	.02

COMMENT. The changes in the temporal retina of the left eye are compatible with retrograde trans-synaptic degeneration of the inner nuclear layer.

CASE 9

A forty-three-year-old male.

DIAGNOSIS. Glomus jugulare tumor of the right temporal bone. Intracranial compression of the right optic nerve.

CLINICAL HISTORY. This patient had a glomus jugulare tumor present for many years and it had recurred causing erosion of the greater wing of the sphenoid and compression of the right optic nerve. It is known that the vision had been decreasing in the right eye for many years. Vision was 20/60 in the right eye several years before death. Bilateral papilledema was present.

MICROSCOPIC PATHOLOGY. Optic atrophy, loss of ganglion cells, and decrease in the inner nuclear layer.

CELL COUNT (Average of five counts). *Temporal*: 2 mm., 95; 4 mm., 92; 6 mm., 83; 10 mm., 67; 12 mm., 57. *Nasal*: 2 mm., 191; 6 mm., 157; 10 mm., 83.

Total number of cells counted (five counts)	4115
Normal mean for all slides counted (five normal eyes)	6329
Probability by chance	.10

COMMENT. The decreased cellularity in the nuclear layer suggests retrograde trans-synaptic degeneration of the inner nuclear layer.

CASE 10

A twenty-eight-year-old female.

DIAGNOSIS. Meningioma compressing the left optic nerve at the chiasm.

CLINICAL HISTORY. The patient noted blurred vision of the left eye two-and-one-half years ago. She had not been able to see out of the left eye for one year.

GROSS PATHOLOGY. An intracranial psammous meningioma compressed the optic nerve.

MICROSCOPIC PATHOLOGY. Atrophy of the optic nerve and loss of ganglion cells were present. The inner nuclear layer appeared normal except for minimal rarefaction of the inner aspect.

CELL COUNT (Average of five counts). *Temporal*: 2 mm., 337; 4 mm., 231; 6 mm., 172; 10 mm., 63. *Nasal*: 2 mm., 132; 6 mm., 118; 10 mm., 81.

Total number of cells counted (five counts)	6279
Normal mean for all slides counted (five normal eyes)	6329

COMMENT. In this case the inner nuclear layer did not appear to be abnormal. Sufficient time had elapsed in this case for changes of retrograde trans-synaptic degeneration to be readily detectable.

CASE 11

A sixteen-year-old female.

DIAGNOSIS. Chondroma compressing the left optic nerve.

CLINICAL HISTORY. A nasopharyngeal chondroma, causing pain in the left eye, had been present for four-and-one-half years before enucleation. Complete loss of vision in the left eye was noted 18 months before enucleation (possibly related to trauma). Proptosis was progressive with loss of ocular movements. In the left eye, no pupillary response to direct light was present, but pupillary response was active consensually. Optic atrophy was present. The left eye was removed.

MICROSCOPIC PATHOLOGY. Optic atrophy and loss of ganglion cells were present. The inner nuclear layer appeared normal, except for rarefaction and cystic changes in the inner aspect of the inner nuclear layer, which may be within normal limits.

CELL COUNT (Average of five counts). *Temporal*: 2 mm., 252; 4 mm., 323; 6 mm., 210; 10 mm., 92; 12 mm., 109. *Nasal*: 2 mm., 139; 6 mm., 97; 10 mm., 85.

Total number of cells counted (five counts)	6753
Normal mean for all slides counted (five normal eyes)	6329

COMMENT. Sufficient time had not elapsed for trans-synaptic retrograde degeneration to be readily evident.

RESULTS

Decreased cellularity of the inner nuclear layer was noted in the first nine cases. In these cases, there was a statistically significant loss of cells. The cell counts at specific meridians are tabulated in Table 1. In all the cases with decreased cellularity of the inner nuclear layer, the lesion had been present for two years or longer. By analysis of variants, the following factors were related to the loss of cells in the inner nuclear layer.

TABLE 1. CELL COUNT, INNER NUCLEAR LAYER
(Average of Five Counts)

	Temporal					Nasal		
Case	2 mm.	4 mm.	6 mm.	10 mm.	12 mm.	2 mm.	6 mm.	10 mm.
1	103	109	104	65	49	132	84	50
2	78	100	70	43	48	69	67	42
3	79	105	82	69	51	62	67	60
4	66	71	86	64	25	61	102	24
5	—	167	119	56	42	—	65	43
6	34	135	130	87	78	111	102	88
7	226	216	153	86	58	122	196	81
8	84	82	72	51	31	72	71	43
9	95	92	83	67	57	191	157	83
10	337	231	172	122	63	132	118	81
11	252	323	210	92	109	139	97	85
Normal								
12	233	234	203	172	131	200	156	107
13	326	313	169	109	95	166	127	107
14	186	203	153	63	66	105	106	55
15	239	291	209	118	125	146	121	107
16	221	283	184	108	76	129	109	90

1. The pattern of cell distribution within the inner nuclear layer remained the same after lesions of the optic nerve in the eyes with decreased cellularity.

2. Degeneration in the inner nuclear layer was detectable by this technique only in cases where optic nerve or chiasm lesions were present for two years or longer.

3. The probability of the loss of cells within the inner nuclear layer occurring by chance was less than .02 for cases with blindness for longer than two years.

4. In Cases 10 and 11, the lesions have been present for less than two years, the inner nuclear layer appeared normal, and the cellularity was not significantly reduced.

In the 28 eyes studied in which a lesion of the optic nerve had caused blindness for longer than two years, there were no eyes with normal inner nuclear layers. No significant reduction in the size of the cells in the inner nuclear layer was detectable.

COMMENT

The decreased cellularity of the inner nuclear layer after lesions of the optic nerve and chiasm is statistically significant. The probability of the changes occurring by chance is less than .02 for the group of

eyes with lesions causing blindness for longer than two years. This degeneration of the inner nuclear layer may represent trans-synaptic degeneration in a retrograde direction. The time required (two years) for the degeneration to occur, and the consistent decreased cellularity of the inner nuclear layer after long-standing lesions of the optic nerve, strongly suggest retrograde trans-synaptic degeneration as the explanation for these findings.

Thinning of the outer plexiform layer was present in Cases 1, 3, and 6. In Case 1, there was loss of cells in the outer nuclear layer (Figure 5). The changes in the outer nuclear layer may represent a further step in the retrograde trans-synaptic degeneration process. Anterograde trans-synaptic degeneration of both geniculate bodies was present in Case 6.

Decreased cellularity of the inner nuclear layer appeared to be the essential manifestation of retrograde trans-synaptic degeneration. Cystic degeneration of the inner nuclear layer may be present in some eyes with cut optic nerves, but is probably not the essential change of retrograde trans-synaptic degeneration. The cystic changes of the inner nuclear layer may result from unassociated retinal changes. Bahn[1] presented a patient with cystic changes in the inner nuclear layer in whom the optic nerve had been cut five months before enucleation. The cystic changes present in the inner nuclear layer of this eye were not distinguishable from those associated with retrograde trans-synaptic degeneration. These cystic changes are probably not related to trans-synaptic degeneration, however, because of the short interval. Cystic degeneration of the inner nuclear layer may possible be the result of degeneration of Müller cells. Trans-synaptic degeneration of the inner nuclear layer may occur without cystic changes as observed in seven of the cases presented in this paper. Small cystic areas may normally be present in the inner nuclear layer of the retina.

The large cystic spaces in the inner aspect of the inner nuclear layer of the retina, associated with retrograde trans-synaptic degeneration, occur where the Müller and amacrine cells are known to be located. Amacrine cells are thought to be inhibitory (Cajal[5]). With a loss of amacrine cells and their inhibitory influence, an elevated ERG response would be expected. Elevated ERG responses obtained long after sections of the optic nerve (three and 37 years later) have been reported in two human cases (Gills[13]); these ERG findings suggest a loss of centrifugal inhibitory fibers. However, there is controversy over whether centrifugal fibers exist and whether ERG responses after section of the optic nerves are elevated (Polyak[30,31] and Brindley[2]).

The loss of cellularity in the inner nuclear layer, evident in Cases 1 through 9, is more marked than would be expected with loss of only the amacrine and horizontal cells. Loss of bipolar cells, which compose most of the inner nuclear layer, is therefore probably the principal reason for the decreased cellularity of the inner nuclear layer after lesions of the optic nerve. However, the entire layer undergoes degeneration as can be seen in the comparative slides of Van Buren. The bipolar, Müller, amacrine, and horizontal cells probably all degenerate.

In four patients with optic atrophy associated with multiple sclerosis for long periods of time, there was decreased cellularity of the inner nuclear layer. The number of cells in the inner nuclear layer compared with normal values, and the probability of the decreased cellularity resulting by chance was .02. The loss of cells in the inner nuclear layer in eyes of patients with advanced multiple sclerosis is compatible with trans-synaptic degeneration of the inner nuclear layer.

These objective observations strongly suggest that retrograde trans-synaptic degeneration of the inner nuclear layer occurs. Several years are required after lesions of the optic nerve and chiasm before the changes are readily visualized. Further investigation of this most interesting and controversial subject is warranted.

SUMMARY

Eleven eyes with lesions of the optic nerve and chiasm and five normal eyes were histologically studied and cells of the inner nuclear layer were counted in specific meridians. There was consistent decreased cellularity in the nine eyes in which lesions of the optic nerve had been present for two years or longer. The probability that the decreased cellularity occurred by chance was less than .02 in five eyes with blindness for longer than two years. Retrograde trans-synaptic degeneration as the explanation for the degeneration in the inner nuclear layer is discussed. These case findings give statistical significance to the controversial concept of retrograde trans-synaptic degeneration of the inner nuclear layer of the retina following lesions of the optic nerve.

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DISCUSSION

DR. J. REIMER WOLTER. Using pathologic conditions as nature's own nerve-cutting experiments the authors have presented in this excellent study very convincing evidence for the occurrence of retrograde trans-synaptic degeneration involving neurons of the inner nuclear layer of the human retina. This trans-synaptic degeneration was observed two years or more following interruption of the ganglion cell neurites in the optic nerve or chiasm and was preceded by simple retrograde degeneration of the retinal ganglion cell layer. The well-documented study fits in well with much less complete earlier observation. It also fits in well with the pattern of similar retrograde trans-synaptic degeneration that has been reported to occur in the retinal ganglion cell layer following lesions in optic radiation.

The occurrence of trans-synaptic degeneration in the visual system is a most interesting fact. It may indicate that one neuron in this chain does not only supply the next neuron downstream with an impulse, but that it also gets something important back in some kind of an exchange without which the neuron would die. There is room, however, for many other possible explanations. Trans-synaptic degeneration of bipolar cells in the cases of the present study was observed after extensive destruction of the optic nerve or chiasm. This destruction, thus, involved not only the afferent (centripetal) neurites of the visual system, but must also have destroyed efferent (centrifugal) neurites. These are now known to be present in man in great numbers. The functions of these efferent nerves in the retina are as yet unknown. However, they could be trophic in nature and their interruption could well be a factor in a diffuse atrophy of retinal neurons. The role of the retinal blood vessels in this late trans-synaptic degeneration of retinal neurons is another open question. The clinician knows that the retinal

blood vessels soon become very narrow in cases with complete atrophy of nerve fiber layer and optic nerve and that the neurons of the inner nuclear layer are thus entirely dependent on the decreased nutrition supplied by these narrowed vessels. Some of the retinal photomicrographs presented by Drs. Gills and Wadsworth show the very advanced vascular narrowing and sclerosis that typically develop after a few years of atrophy of the inner retina and optic nerve. Was there a relation of the degree of vascular sclerosis to the presence or absence of cystic degeneration in the inner nuclear layer in the cases of this study?

It would be interesting to know whether the authors have tried to differentiate the neuronal cells of the inner nuclear layer from the radial cells of Müller. It seems that the results of their cell counting would have been even more significant if they would have considered the neurons only—and not the glial cells of this layer.

May I finally congratulate the authors on a very important contribution. Their detailed quantitative work has settled an uncertain and controversial issue.

DR. JAMES P. GILLS, JR. Retinal vascular attenuation begins three to five years after optic nerve lesions and the attenuation progresses with time. The attenuation appears to occur following loss of cellularity and may thus be a trophic phenomenon.

The type of cells of the inner nuclear layer that degenerate is not known. We will continue to pursue this interesting aspect by examining monkey eyes with chronic lesions of the optic nerves by special histochemical stains.