Blood Vessels of the Glaucmatous Optic Disc in Experimental Primate and Human Eyes

Harry A. Quigley, Rebecca M. Hohman, Earl M. Addicks, and W. Richard Green

Experimental chronic glaucoma was produced in primate eyes and quantitative measurements were made of the capillary area in the optic nerve head. The percent of the nerve head occupied by capillaries remained normal despite considerable loss of disc tissue as glaucomatous excavation proceeded. This finding, identical to that in simple optic atrophy, suggests that the nerve head can maintain a stable capillary to tissue ratio despite substantial tissue loss. Further observations on human glaucoma eyes confirmed that there is not selectively greater loss of capillary volume out of proportion to the loss of neural tissue of the optic nerve head. Fluorescein angiography of primate and human glaucoma eyes showed that a change in the appearance of disc vessels occurred most frequently when there was little or no remaining neural or vascular tissue anterior to the scleral lamina cribrosa. Invest Ophthalmo Vis Sci 25:918–931, 1984

While the mechanism of glaucoma’s damage to the optic nerve has been studied extensively, no definitive explanation has emerged yet. Recent investigations have linked the regional structure of the optic nerve head to the pattern of nerve fiber loss,1–3 supporting the idea that intraocular pressure (IOP) elevation induces physical changes in the nerve head as an initial step in damage. Compression and distortion of the lamina cribrosa, the site of damage,4,5 occurs in human glaucoma eyes even prior to the stage of visual field loss.6

It is not established, however, whether these early changes in the laminar connective tissues directly compress nerve fibers or indirectly injure them by affecting the blood supply to axonal bundles carried in nerve head microvasculature. Two independent experiments in primates suggest that direct neural injury is the more likely mechanism,7,8 while another acute primate study indicates that elevated IOP causes abnormal vascular permeability.9 When blood flow is measured during acute IOP increase in animals, no decrease in vascular supply is detected at IOP levels that damage nerve fibers.8,10,11

The lack of ultrastructural damage to nerve head capillaries5,12,13 and the apparently normal blood flow with elevated IOP seem to contradict the conclusions of previous reports supporting an ischemic mechanism. Cristini14 and Francois15 each reported that blind glaucoma eyes have a disproportionate loss of disc capillaries by light microscopy and injection techniques. Kalvin and co-workers found poor filling of disc vessels in monkeys with acute experimental glaucoma.16 The purpose of our report is to study quantitatively the number and size of blood vessels in glaucomatous nerve heads. We examined nerve head capillaries in a number of human glaucoma eyes and performed quantitative measurements of capillary area in a chronic glaucoma model system in primates.

In addition, the fluorescein angiographic appearance of the optic disc is different from normal in the glaucomatous human eye17,18 and in acute monkey IOP experiments.19 This investigation compares the fluorescein angiography of human and primate eyes with the histology of the same eyes. In this way, some of the previously reported changes induced by elevated IOP can be understood better.

Materials and Methods

Two human eyes with iris melanomas that caused secondary glaucoma were studied both clinically and histologically.6 Informed consent was obtained from both. In both eyes, fluorescein angiography of the optic disc was performed at IOP between 20 and 30 mmHg. Both eyes were enucleated soon thereafter because of tumor growth and uncontrolled IOP. They were fixed...
in 4% glutaraldehyde in phosphate buffer, postfixed in 2% osmium tetroxide, and each optic nerve head was embedded in epoxy resin. One-micron thick sections were cut at multiple levels in each specimen and examined by phase contrast light microscopy.

Chronic experimental glaucoma was produced in the eyes of cynomolgus monkeys (Macaca fascicularis) by one of two methods: (1) anterior chamber injection of ghost red blood cells or (2) laser treatment of the trabecular meshwork. Investigations using animals in this study conform to the ARVO Resolution on the Use of Animals in Research. IOP was monitored with applanation tonometry or with a calibrated pneumotonomograph, under ketamine hydrochloride anesthesia. In these eyes, the percent normal figure is derived by comparing the media induced glaucoma had media too hazy to examine the optic disc; in the five eyes of this type included for histologic study (Table 1, eyes 1, 2, 4–6) the change in cup size was measured as a cup to disc ratio in the fixed tissue with calipers. Laser-induced glaucoma was produced in 15 eyes. These 15 had their discs photographed stereoscopically in color and in red-free light, and they had fluorescein angiograms, all performed at regular intervals under intravenous pentabarbital sodium anesthesia. In these eyes, cup/disc ratios were measured by overlaying a scale on stereophotographs (as was done in the human eyes reported as well). When IOP was measured after administration of pentabarbital, the IOP was usually lower than the value under ketamine sedation. In addition, the IOP during barbiturate anesthesia varied somewhat dependent upon the level of anesthesia. The effect of IOP on angiographic appearance was not studied systematically; however, the range of IOP of eyes photographed was between 15 and 40 mmHg.

### Table 1. Clinical, optic nerve head and optic nerve data on glaucomatous monkeys

<table>
<thead>
<tr>
<th>Monkey no.</th>
<th>Duration of IOP elevation</th>
<th>IOP level (mmHg)*</th>
<th>Cup size increase</th>
<th>Neural area optic nerve, % normal</th>
<th>Nerve fiber layer atrophy</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mild damage</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>1 week</td>
<td>65 ± 5</td>
<td>no</td>
<td>87%†</td>
<td>no exam</td>
</tr>
<tr>
<td>2</td>
<td>1 month</td>
<td>41 ± 12</td>
<td>no</td>
<td>83%†</td>
<td>no exam</td>
</tr>
<tr>
<td>3</td>
<td>2 months</td>
<td>46 ± 13</td>
<td>to 0.5</td>
<td>107%†</td>
<td>no exam</td>
</tr>
<tr>
<td>Moderate damage</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>4</td>
<td>2 weeks</td>
<td>48 ± 8</td>
<td>no</td>
<td>79%</td>
<td>50–90% loss</td>
</tr>
<tr>
<td>5</td>
<td>2 weeks</td>
<td>64 ± 13</td>
<td>to 0.8</td>
<td>73%†</td>
<td>no exam</td>
</tr>
<tr>
<td>6</td>
<td>4 months</td>
<td>33 ± 8</td>
<td>to 0.9</td>
<td>77%</td>
<td>0–75% loss</td>
</tr>
<tr>
<td>Severe damage</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>7</td>
<td>3 months</td>
<td>39 ± 9</td>
<td>to 0.95</td>
<td>69%†</td>
<td>90–100% loss</td>
</tr>
<tr>
<td>8</td>
<td>11 months</td>
<td>46 ± 15</td>
<td>to 0.95</td>
<td>10%</td>
<td>90–100% loss</td>
</tr>
<tr>
<td>9</td>
<td>4 months</td>
<td>34 ± 6</td>
<td>to 0.95</td>
<td>15%</td>
<td>90–100% loss</td>
</tr>
<tr>
<td>10</td>
<td>4 months</td>
<td>34 ± 5</td>
<td>to 0.95</td>
<td>20%</td>
<td>90–100% loss</td>
</tr>
</tbody>
</table>

* Mean ± standard deviation. † In these nerves, the percent normal figure is derived by comparing the nerve area with the mean of normal monkey data for nerve area or percent capillary area (Table 2). In all others, control is fellow eye.

Ten glaucomatous eyes were studied by quantitative, histologic techniques. Five of these had had ghost-cell glaucoma (Table 1, eyes 1, 2, 4–6) and five had laser-induced glaucoma (Table 1, eyes 3, 7–10). All 10 animals were killed by intravascular perfusion of fixative, under deep anesthesia, to study their 10 glaucomatous eyes and 7 normal fellow eyes. (In three animals the fellow eye was utilized for a different experiment not reported here.) The IOP at fixation was set artificially at 15 mmHg by an anterior chamber needle connected to a reservoir. We felt that the measurements we wished to make should express the potential capillary space in all eyes, glaucomatous and normal, under standardized conditions of blood and eye pressures. While one might think that allowing the IOP to be set at the elevated level prevailing in the glaucoma eyes would allow an idea of how open the capillaries were in vivo, we do not feel that our method in this paper is capable of measuring the flow rate of blood or the relative number of capillaries that were actually open or closed under physiologic conditions. Rather, the measurement given here represents glaucoma eyes compared with nonglaucoma, normal eyes in terms of capillary volume with all conditions identical.

The fixed optic nerve heads were divided in half vertically, postfixed in osmium, embedded in epoxy resin, and both halves serially sectioned for phase-contrast light microscopy. The area of the nerve head and the area occupied by capillary lumens were measured with an image analysis system by the following method, previously described and illustrated. From the serial sections of each nerve head, we took a section every 50 μm, so that a total of at least 12 sections were
Table 2. Capillary area as percent of nerve head tissue

<table>
<thead>
<tr>
<th>Capillary area</th>
<th>% of fellow eye</th>
</tr>
</thead>
<tbody>
<tr>
<td>Normal eyes (10)</td>
<td>2.2 ± 0.4%</td>
</tr>
<tr>
<td>Glaucoma eyes (10)</td>
<td>2.2 ± 0.3%</td>
</tr>
<tr>
<td>Mild damage (3)</td>
<td>2.3%</td>
</tr>
<tr>
<td>Monkey 1</td>
<td>2.1%</td>
</tr>
<tr>
<td>Monkey 2</td>
<td>2.0%</td>
</tr>
<tr>
<td>Monkey 3</td>
<td>2.7%</td>
</tr>
<tr>
<td>Moderate damage (3)</td>
<td>2.4%</td>
</tr>
<tr>
<td>Monkey 4</td>
<td>2.1%</td>
</tr>
<tr>
<td>Monkey 5</td>
<td>2.4%</td>
</tr>
<tr>
<td>Monkey 6</td>
<td>2.8%</td>
</tr>
<tr>
<td>Severe damage (4)</td>
<td>2.0%</td>
</tr>
<tr>
<td>Monkey 7</td>
<td>2.2%</td>
</tr>
<tr>
<td>Monkey 8</td>
<td>1.9%</td>
</tr>
<tr>
<td>Monkey 9</td>
<td>1.9%</td>
</tr>
<tr>
<td>Monkey 10</td>
<td>1.9%</td>
</tr>
</tbody>
</table>

* Comparison is with pooled normal group due to lack of fellow eye data.

The glaucomatous nerve heads as a group had the most severe damage, histologic estimates of remaining neural tissue were performed. The optic nerve was removed from the globe and razor marks were placed to identify the superior and nasal zones. After osmium postfixation and epoxy-embedding, 1-μm cross-sections were performed on tissue from 1–2 mM behind the globe. The area of the whole nerve inside the pia mater excluding the central vessels was measured with an image analysis system. In similar fashion, the area occupied by neural bundles alone was measured. The latter represents a reasonable approximation of the proportion of remaining optic nerve fibers, and allows us to know at what stage of damage were the eyes with various capillary counts.

Nine selected specimens of fixed retina were taken from standard locations, and postfixed and embedded as above. In 1-μm sections, the thickness of the nerve fiber layer was measured and compared with the normal values previously obtained for this species. This data, along with the measurement of cup/disc ratio and the amount of remaining neural tissue in the optic nerve, were combined to assess the stage of damage (Table 1).

Results

Quantitative Capillary Measurements

In epoxy-embedded sections of perfusion-fixed tissue, capillaries are identified easily as clear zones surrounded by their single endothelial lining cells (Fig. 1, 2). Vessels with more than one layer of surrounding cells were not included in the counts of vascular or total area. In a study of normal and atrophic nerve heads, we found that the number of capillaries per unit area and the mean size of capillaries varied in different zones of the tissue, but the percentage of tissue occupied by capillaries was quite constant in normal eyes. Hence, it is this parameter, percent capillary area, that is given in the Tables.

The 10 eyes included 3 in which relatively brief exposure to elevated IOP had occurred and little optic nerve damage was detectable either clinically or histologically (Table 1). Three additional eyes clearly had suffered some damage since disc cup size had enlarged, nerve fiber layer atrophy was present, and the neural area of the optic nerve had declined compared with fellow eyes. Four eyes had severe loss of neural tissue in the retinal nerve fiber layer, large disc cups, and substantial loss of neural area. It should be noted that disc cup size increase sometimes occurs rapidly in the macaque eye, even at 2 months with IOP as low as 40 mmHg. This also was observed by Gaasterland.

The glaucomatous nerve heads as a group had the same percent capillary area as the normal eyes (Table 2; P > 0.05, two-tailed t-test). In the seven pairs of eyes in which a normal fellow eye could be compared directly section for section to the glaucoma eye of the
same animal, there was no significant difference by analysis of variance ($P > 0.05$). These statistical facts are obvious from simple inspection of the data in Table 2. The lack of difference between glaucoma and normal eyes in this parameter was true whether the elevated IOP had been present for only a brief exposure or had been of several months duration leading to severe loss of nerve fibers. Histologically, the full range of tissue loss is represented in the eyes in this series, from nearly normal nerve heads to bean-pot cupping (Fig. 1). In each case, the glaucoma nerve head had a percent capillary area of approximately 2%, just as normal eyes do. In fact, the largest difference between a glaucoma nerve head and its fellow (monkey 10) was a 20% difference in percent capillary area, and the glaucoma nerve had proportionately more capillary area.

Capillaries were present throughout the remaining nerve head tissue with no consistent zones of local loss. The capillaries within the scleral lamina cribrosa were no less intact than elsewhere (Fig. 2). We found it both unreliable and impractical to attempt to subdivide the nerve head into local zones. The boundaries between such zones are arbitrary and ill-defined in many sections.

The macaque nerve head is quite similar to the human. Our detailed study of the nerve fiber layer and optic nerves of the glaucomatous primates in this study show a pattern of damage that simulates that in the human eye. In both monkey and human, the loss of nerve fibers as viewed in retrobulbar optic nerve cross-sections is a diffuse atrophy with selectively greater loss at the superior and inferior poles of the nerve (Fig. 3). In the clinical examination of the retinal nerve fiber layer in red-free light, the primates also followed a pattern seen in human glaucoma patients. We observed a total of 15 primate eyes with serial red-free photography (only five of whom were included in the quantitative capillary counting, the rest being used in other experiments). Of these, four eyes had no detectable abnormalities in the clinical nerve fiber layer exam;
all had low levels of IOP for short durations and had no cup size enlargement either. In the 11 eyes that had progressive cup size enlargement, all developed atrophy of the nerve fiber layer. In 7 of the 11, the initial abnormal finding was a local or slit-like defect in the arcuate nerve fiber layer (Figs. 4, 5), either superior or inferior to the disc. In the other four who developed nerve fiber layer atrophy, diffuse loss of all striations occurred so rapidly (within 2 months), that no local defect stage was recognized.

There was one eye in which the retinal nerve fiber layer was severely atrophic, both clinically and histologically, but the loss of neural area in the optic nerve cross-section was only moderate (Table 1, eye 7). This occurred in all probability because this animal's damage was relatively rapid. While the axons had degenerated in the retina, the myelin sheaths of these dead fibers had not yet been cleared from the retrobulbar nerve by phagocytes. This myelin debris artificially kept the neural area closer to a normal value explaining the discrepancy. In animals allowed to survive for longer periods, or whose degeneration proceeded more slowly (eg, Table 1 eyes 8–10), neural area was 10–20% of normal since more myelin debris had been cleared.

**Fluorescein Angiography**

Despite the elevated IOP during angiography in many primate eyes, the pattern of disc vessels was seen
easily and indistinguishable from its appearance prior to induction of glaucoma until major cupping had occurred. Even when the disc rim was narrowed severely, there were no large areas of the disc devoid of detectable vessels (Figs. 6, 7). The original branch arterioles and venules could be identified in their more posterior locations, and smaller caliber vessels were seen between them as in the normal state. However, since the resolution of angiography is limited, it is not possible to judge whether the smaller vessels seen are the same or different ones from those seen prior to cup size increase. Nor it is possible to detect quantitatively whether there are more or fewer vessels present.

One change from normal in the primate disc with enlarged cup size is loss of the diffuse glow behind the superficial vessels of the disc (Figs. 6, 7). In the deeply cupped disc, the small vessels appear to be outlined sharply against a darker background until 5–10 min after injection when the usual diffuse fluorescence leaking in from the surrounding choroid blurs the details as it does in the normal disc. Since we have some histologic correlations, it is possible to demonstrate that there are, of course, capillaries on the floor of the deeply cupped disc anterior to the scleral or collagenous portion of the lamina cribrosa that are surely the small vessels seen angiographically (Fig. 7). However, the

---

**Fig. 3.** Cross-section of optic nerve from primate eye (no. 8) with severe glaucoma damage. Black areas in nasal and temporal nerve (right and left, respectively) are intact, remaining neural bundles. Superior and inferior (upper and lower) nerve is light in color indicating no remaining fibers in an hourglass-shaped pattern similar to that seen in later stages of glaucoma damage in human eyes (X38, paraphenylenediamine).

**Fig. 4.** Rapid and progressive loss of nerve fiber layer striations in primate eye with higher range IOP. Normal appearance, upper left. In upper right, early phase of cupping has begun and subtle diffuse atrophy of nerve fiber layer pattern is suggested. Lower left, further loss of nerve fiber pattern diffusely and single local defect with loss of all striations inferiorly (arrow). Lower right, loss of all visible nerve fibers (see cross-section of this eye's optic nerve in Fig. 3).
Fig. 5. Photograph taken in red-free light of nerve fiber layer of primate before (upper) and after (lower) development of local defect in striations in inferior arcuate area (arrow) after six months of IOP elevation. Such local defects in nerve fiber layer are commonly observed in primates with extended IOP elevations to moderate levels, just as in human glaucoma eyes.

depth of these in the anteroposterior dimension is reduced greatly from the normal primate disc rim, and only a thin layer of such tissue made up of astrocytic glial cells and capillaries separate the vitreous cavity from the scleral lamina.

The two human glaucoma eyes present similar degrees of nerve fiber loss, but somewhat different angiographic appearance and nerve head histology. In both eyes, there was nearly normal visual acuity and mildly abnormal psychophysical testing. In one eye, the vertical cup to disc ratio was approximately 0.8 and the degree of undermining of the rim was minor (Fig. 8). The angiogram showed filling of small vessels throughout the disc though the rim at 12 o’clock had the poorest filling. When the disc was examined by light microscopy, the surface of the nerve head was collapsed backward (Fig. 9). However, anterior to the scleral laminal cribrosa, there was still a variable amount of remaining tissue containing nerve fibers, astrocytes and capillaries.

The second eye had more severe rim loss and excavation with vertical cup to disc ratio of 0.9 (Fig. 10). Angiography of this disc showed a reduced number of vessels, and those that were seen appeared blurred despite good focus of the remaining structures. Histologically, this eye had much more excavation and loss of anterior disc tissue, with few remaining vessels anterior to the collagenous portion of the nerve head (Fig. 11). In the collagenous lamina, however, there were capillaries seen throughout the specimen. In neither of the human examples could the number of capillaries be measured adequately. Quantification is impossible because the vessels have substantial variation in the patency of their lumens and also have red cells present in some vessels and not in others. Since it is not possible to standardize the IOP and fixative perfusion pressure, as in the primates, too great a variability arises for reproducible counts.

Discussion

In the first portion of this study, we found that the percent capillary area is normal in primate nerve heads with chronic glaucoma damage. The same is true of nerve heads with optic atrophy induced by orbital optic nerve transection. If nerve fibers are lost from the nerve head in an atrophic process, and the number of capillaries that was originally present remained the same, the percent area that they occupied would actually increase. The fact that the percent capillary area is remarkably constant indicates that as nerve fibers atrophy, so do capillaries, at just the appropriate rate to keep the percent vascular area stable. This regulation of capillary volume is just as efficient in nerve heads damaged by experimental glaucoma as in nonglaucomatous optic atrophy. We already had suspected that this was the case, based on more than 50 examinations of human glaucoma nerve heads by light and electron microscopy (including the two examples shown here). However, it is impossible to quantify and control the human eye observations as precisely as those in primates.

It is difficult retrospectively to determine the reason for the difference between our observations and previous reports of capillary loss in glaucoma nerve heads. In the other human reports, little clinical information was available and the eyes were predominantly blind. In Cristini’s report, eyes were “enucleated in a very advanced phase of the disease,” and in the
other cited report, they are studied with visual acuities of no light perception, light perception, and 9/10. Second, none of the previous reports used quantitative methodology. We have found it impossible to detect reliably the number of vessels present without quantitation. In the case of acute experimental primate studies using injection techniques to identify capillaries, several methodological problems could explain the difference. First, the IOP levels were extremely high in these studies, possibly preventing marker entry into vessels that were present. In fact, one of the authors of this study later detected vessels present in trypsin digest preparations that had not filled with india ink. Furthermore, a number of these animals had IOP high enough to cause central vein occlusion and retinal infarction. This has not been the case in our model systems. Hence, the previous reports may apply only to the extreme limits of IOP effects.

It is important to distinguish between our study that quantifies the number of capillaries present and one that measures important functional vascular properties: flow, intravascular pressure, oxygen delivery, etc. Our study shows that capillaries are not disproportionately lost in glaucoma. It does not demonstrate that microvascular function was normal, but merely that there is no reason to believe that such function would be deficient because of an insufficient capillary volume. It could be speculated that initial physical distortions
Fig. 7. Correlation of fluorescein angiogram and nerve head histology in primate eye with severe glaucoma damage from 11 months of increased IOP. Angiogram shows filling of large and small vessels throughout the deep cup floor. The major difference between this and the normal angiographic appearance is the lack of a diffuse glow between vessels here. In low and higher power micrographs of the cup floor of this eye (left and right, respectively), it is apparent that in front of the scleral or collagenous lamina (its anterior limit is designated by arrowheads), lies a thin layer of glia and capillaries (small arrows). These vessels are the small ones seen angiographically. Posterior to this, there are more capillaries, but these are surrounded by collagen of the lamina. (Lower left, X270; lower right, X425, paraphenylenediamine).

of lamina cribrosa tissues could compress capillaries in this area and depress the blood supply to neurons without causing capillary loss. In this case, the findings of our study would be compatible with a glaucoma damage mechanism involving poor vascular nutrition. However, most recent experiments that have attempted experimentally to measure blood flow in the optic nerve head with elevated IOP had found no decrease in flow until IOP approaches mean ocular blood pressure. One might argue that blood flow must be measured in the human glaucoma nerve head and that any experiment with primate eyes is unsatisfactory. We have shown that the primate glaucoma model simulates the disc, nerve fiber layer, and optic nerve damage patterns of the human eye. Since it is presently not feasible to measure blood flow in the human eye, we are presently measuring it in the chronic primate glaucoma model with attention to detailed microvascular function in zones as small as single nerve bundles.

Past studies of fluorescein angiography of the optic disc have suggested the possibility of vascular compromise in the glaucoma process. Our data helps to place some of these previous findings in perspective by comparing the angiograms of a nerve head with its histology. Both our primate and human eyes show that a considerable loss of nerve fibers is possible without any definite angiographic defect. To be sure, there are major shifts in the course of the arterioles on the nerve head surface as the cup widens and deepens, but the number of small vessels that are seen can remain remarkably constant. We found that some anterior disc rim atrophy occurs without detection in the angiogram,
as long as enough capillary bed remains at the cup floor in front of the scleral or collagenous lamina to present itself to the clinical view. However, in those circumstances where the anterior layer of remaining tissue becomes very thin, one visible change is the loss of a deep glow behind the superficial vessels early in the angiogram. This is a glow that presumably comes from capillaries below those on the disc rim surface but anterior to the scleral lamina. When only a tiny layer of vessels is present in front of the scleral lamina, this glow is not present and the remaining vessels are seen in sharp relief against a dark background. The background is dark, because the deeper vessels of the scleral lamina are surrounded completely by collagenous beams. These are invisible to clinical viewing in angiography because the surrounding collagen reflects light, preventing excitation of the fluorescein in the capillary, as well as hindering fluorescent emission from returning to the detecting film. Only those most superficial scleral lamina vessels with thin collagen coats might be seen and, then, in poor focus.

Hence, angiography seemed insensitive to the early phases of disc rim tissue loss. A phase of poor visibility of vessels did develop in advanced damaged eyes when
Fig. 9. Histology of human eye seen in Figure 8. Significant loss of anterior disc tissue lead to backward collapse of nerve head (upper right). Upper right shows disc rim with reduced amount of tissue but each of elements, nerve fibers, glia, and capillaries (arrows) remain. Lower left, nerve head opposite level of choroid with column of glial cells (G), thinned nerve bundle (N), and many capillaries (arrows). Lower right, middle of cup floor with no remaining nerve fibers; glia and capillaries occupy remaining tissue. These surface capillaries probably make up most of those visible angiographically. (Upper left, X50; upper right, X300; lower left, X450; lower right, X420, paraphenylenediamine).

A substantial loss of neural and vascular tissue had occurred. In our material, this change in the apparent number of vessels was not consistently present until considerably more than one-half the optic nerve tissue was gone. We had expected that in eyes with somewhat localized neural loss indicated by local nerve fiber layer defects (eg, Fig. 5), that the local loss of disc rim tissue also would be associated with a lack of visible vessels on the disc rim angiographically. This was not the case; several eyes had local defects in nerve fiber layer and even neural rim narrowing without apparent loss of vessels angiographically.

Not only does neural loss substantially precede angiographic defects, but discs with zones of no apparent filling still have capillaries present, especially within the scleral lamina cribrosa. It is especially important to note that angiography can show no vessels at the scleral lamina when they are present, since substantial evidence exists that the scleral lamina is the site of damage in glaucoma. It is at this location, in acute and chronic experimental glaucoma and in human glaucoma eyes, that the nerve fibers show the initial signs of injury both physiologically and anatomically. Therefore, the capillaries of greatest interest in glaucoma pathogenesis are hidden from clinical and angiographic view. The visible pattern of more anterior vessels is only an indirect expression of what might be occurring below. Though one might assume that the superficial and laminar capillaries behave similarly, this may be incorrect. Both the data of Geijer and Bijn and Soosi and Anderson suggest that the retinal capillary circulation is unable to maintain normal flow
at extremely elevated IOP, while the laminar vessels do so. In this case, angiography might suggest poor filling or no flow in anterior vessels when normal flow was present at the site of major interest in the scleral lamina. Our study does not show by angiography whether flow is normal or abnormal in the disc. It does suggest that one should interpret with great caution data from angiography as a measure of flow or vessel patency in the scleral lamina cribrosa.

It seems established that apparent lack of vascular filling of optic disc sectors occurs in some glaucoma eyes.17,18 Our data suggest that this finding results when loss of disc rim tissue is severe enough that few nerve fibers and capillaries remain anterior to the scleral lamina. We find no support for the idea that it results from disproportionate loss of capillaries prior to later neural atrophy. But, is it possible that the filling defects observed in human eyes represent a lack of flow caused by elevated IOP in capillaries in an intact disc rim? If this were the case, one might expect that the higher the IOP, the more extensive would be the filling defects. However, while filling defects were seen more frequently in glaucoma suspects with elevated IOP than in normals by Loebl and Schwartz,29 there was no

Fig. 10. Angiogram of second human glaucoma eye, with clinical photographs of normal right disc (bottom left) and glaucomatous left disc with 0.9 cup/disc ratio (bottom right). The disc rim is nearly absent, hence vessels seen in cup are those of disc floor only. They are apparently fewer than in normal state, seen against a dark background, and appear in poor focus (though photographs are in optimum focus for this level).
Fig. 11. The deep cup of same eye as Figure 10 is seen at low power above, with slightly deeper and narrower appearance than in life due to preparation artifact. Lower left, disc floor showing only glia and capillaries (arrows) remaining. Note that here the only capillaries remaining are those of the superficial scleral lamina and are surrounded by a clear zone that contains collagenous connective tissue. Possibly this collagen covering leads to the blurred appearance of capillaries in the angiogram. Just below this thin covering layer, the thicker sheets of scleral lamina surround many remaining capillaries, some still containing red blood cells (arrows). These vessels are part of the dark background of the angiogram, since fluorescence is either poorly stimulated or poorly transmitted out of these vessels or both because of the dense connective tissue around them (Top, ×35; bottom left and right, original magnification, ×315, paraphenylenediamine).

correlation between the extent of filling defects and the patient's IOP at the time of angiography. Spaeth likewise found that "the relationship between level of intraocular pressure and perfusion of the optic nerve . . . by fluorescein angiography is complex. Angiography suggests . . . that at pressures lower than the systolic the responses are surprisingly variable." Our observations qualitatively support those of Schwartz and Spaeth that the filling of capillaries on the disc is not highly correlated with IOP. This indicates that blood flow is relatively independent of IOP in the range studied (under 50 mmHg) or that angiography is a weak indicator of actual blood flow. Clearly, angiographic defects do occur as part of glaucoma's damage in some eyes. Whether they represent a major loss of anterior disc rim tissue as suggested by our data or represent poor flow in capillaries supplying an intact zone of nerve fibers, it may be possible to utilize them to predict those eyes proceeding to further damage. As yet, only a small number of such prospectively studied cases has been illustrated. Further examinations will be important in establishing the clinical usefulness of the finding.

Key words: glaucoma, pathology, optic disc, fluorescein angiography, capillary, optic nerve, retina, blood flow, axon

References


