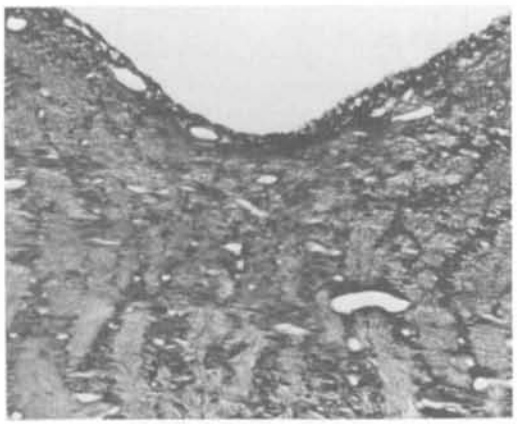

Fig. 3. This section (×100) of the optic nerve head of animal No. 30 shows the maximal extent of histologic change and cupping observed six weeks after severing the short posterior ciliary arteries. There is glial proliferation and replacement of lost neurons. No retrodisplacement of the cup, nor glaucomatous cupping is present. A small amount of saucerization is noted.

severed short ciliary arteries. In addition, spasm of the central retinal artery may have occurred as a result of the surgery. Histologic evidence of intraretinal hemorrhages in two eyes supports this. All probably contributed to the compromise in retinal nutrition during the period of protein synthesis.

Our results contrast with those of Anderson and Davis[10] who found no evidence of any histologic change in the optic nerve head of young squirrel monkeys following surgical interruption of the short posterior ciliary arteries. This may reflect their selection of the young squirrel monkey for study. Armaly and Araki[4] have found a 79 per cent reduction in blood flow to the retrolaminar optic nerve of rhesus following ligation of the short posterior ciliary arteries. A decrease in blood flow of this magnitude would probably be sufficient over a period of time to explain the 80 per cent reduction in axoplasmic transport and the histologic changes observed.

The technical assistance of Cheryl Curington is appreciatively acknowledged.

From the Veterans Administration Hospital (MRIS 5221-02) and the Department of Ophthalmology, University of Florida, Gainesville, Fla. This study was supported in part by Fight for Sight, Inc. (G-507), New York, N. Y. Submitted for publication Nov. 3, 1975. Reprint requests: Dr. N. S. Levy, 4020 Newberry Rd., Gainesville, Fla. 32607.

REFERENCES

1. Levy, N. S.: The effect of elevated intraocular pressure on slow axonal protein flow, INVEST. OPHTHALMOL. **13**: 691, 1974.
2. Anderson, D. R., and Hendrickson, A.: Effect of intraocular pressure on rapid axoplasmic transport in monkey optic nerve, INVEST. OPHTHALMOL. **13**: 771, 1974.
3. Levy, N. S.: The effect of elevated intraocular pressure on slow axoplasmic transport in the rhesus monkey. Doctoral Dissertation. University of Chicago, 1975.
4. Armaly, M. F., and Araki, M.: Optic nerve circulation and ocular pressure: Contribution of central retinal artery and short posterior ciliary arteries and the effect on oxygen tension, INVEST. OPHTHALMOL. **14**: 475, 1975.
5. Hayreh, S. S., and Baines, J. A.: Occlusion of the posterior ciliary artery. III, Br. J. Ophthalmol. **56**: 754, 1972.
6. Levy, N. S., and Adams, C. K.: Slow axonal protein transport and visual function following retinal and optic nerve ischemia, INVEST. OPHTHALMOL. **14**: 91, 1975.
7. Karlsson, J. O., and Sjöstrand, J.: Synthesis, migration, and turnover of protein in retinal ganglion cells, J. Neurochem. **18**: 749, 1971.
8. Williard, M., Cowan, W. M., and Bagelos, P. R.: The polypeptide composition of intraaxonally transported proteins. Evidence for four transport velocities, Proc. Natl. Acad. Sci. **71**: 2183, 1974.
9. Leone, J., and Ochs, S.: Reversibility of fast axoplasmic transport following differing durations of anoxic block in vitro and in vivo. Program/Abstracts, Annual Meeting of the Society for Neuroscience, **3**: 147, 1973.
10. Anderson, D. R., and Davis, E. B.: Retina and optic nerve after posterior ciliary artery occlusion: an experimental study in squirrel monkeys, Arch. Ophthalmol. **92**: 422, 1974.

Centrioles and cilia in the mesothelial cells of the pericanalicular region.

M. GARY WICKHAM AND DAVID M. WORTHEN.

An evaluation of 70 trabecular meshwork biopsies obtained at the time of therapeutic surgery in glaucomatous and cataractous eyes revealed that the mesothelial cells in the iridocorneal angle had a marked abundance of cilia and centrioles. The distribution of cells showing cilia and/or centrioles is positively correlated with the apparent aqueous humor outflow pathway. The morphology and arrangement of the cilia-centriole complexes in the angle are highly variable and show many forms not previously reported in a single tissue. There were no obvious correlations between organelle abundance and the identifiable factors affecting the patients involved in this study.

Scherft and Daems[1] presented a review of the literature on the various types of cilia in vertebrates. Their data showed that most single or

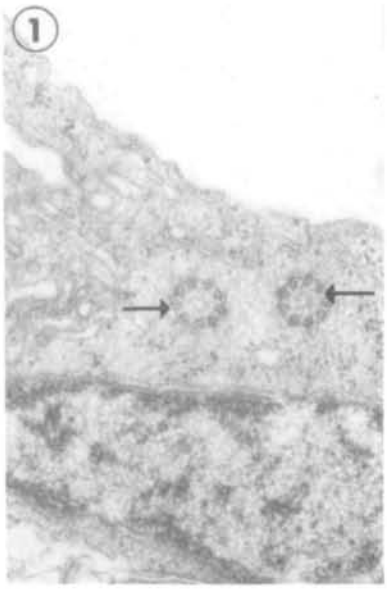

Fig. 1. A commonly occurring but atypical centriolar arrangement in a meshwork cell with two basal bodies shown in cross section. ×33,500.

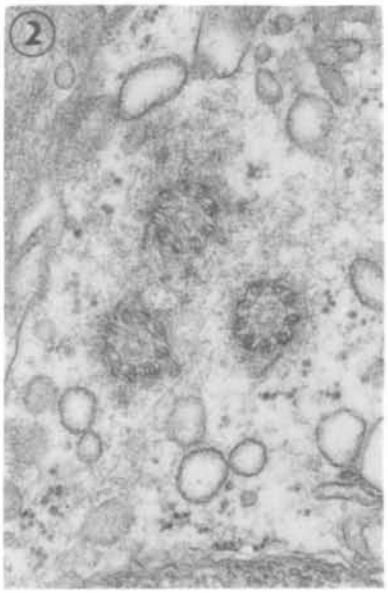

Fig. 2. An illustration of three cross-sectioned basal bodies present in a corneoscleral meshwork cell. ×64,000.

primary cilia originated from a centriolar pair and had a 9 by 2 + 0 structure consisting of nine peripheral bundles of two tubules each with no central bundles.

Single cilia have been identified in several tissues of the eye. Allen[2] found primary cilia in neurons of the inner nuclear and ganglion cell layers of the human and guinea pig retinas. These cilia were of unknown function, but Allen suggested that they might be sensory in nature. Shabo and Maxwell[3] and Inomata, Bill, and Smelser[4] briefly mentioned the occurrence of single cilia in the mesothelial cells of the non-human primate trabecular meshwork. Wulle[5] briefly described the occurrence of single cilia of the trabecular meshwork of the developing human eye, while Vegge[6] mentioned the occurrence of single cilia in the trabeculum of adult human eyes. Hogan, Alvarado, and Weddell[7] illustrated the presence of this organelle type in several anterior segment tissues. Several good studies of the fine structure of the trabecular meshwork of man and the non-human primate[8, 9] do not mention single cilia. This may be due to a number of factors such as species, age, disease state and the amount of the meshwork examined.

Since single cilia are distinctly marked by being ensheathed by the cell and associated with a single centriole pair, they are easily identified in any cell type in which they occur. In addition, since they have an unknown function and/or significance, their occurrence in number in any tissue type adds to the possible elucidation of their function. We wish to report on the occurrence of numerous typical and atypical single cilia occurring in the trabecular meshwork of human eyes suffering with either primary open-angle glaucoma or cataract.

Methods and materials. Two millimeter trephine biopsy samples of the trabecular meshwork were obtained at the time of filtering surgery in 100 patients. The samples were immediately fixed in the operating room using a wide variety of fixative schedules. Immediately after fixation, they were dehydrated in ethanol and embedded in an Epon 812 mixture. Thick sections (2 μ) were taken until a full-thickness, physically undisturbed portion of the trabecular meshwork canal of Schlemm region was found. Thirty of the biopsies taken in surgery were rejected because they did not possess such an undisturbed area of tissue. Following thick sectioning, the tissues were serially thin sectioned, mounted on 75 by 300 grids, stained with uranyl acetate and Reynolds lead citrate and examined in a Zeiss EM 9S2 or in a Zeiss EM 10 electron microscope.

Results. We found single cilia to be more common in the trabecular meshwork than expected on the basis of previous reports in the literature.[3-6] There was no association between disease state and either the type of or abundance of cilia. The highest density of cilia was in the presumed flow path of aqueous humor into the canal, but occurrence was highly variable sample to sample. In all cases, the uveal and corneoscleral meshwork contained the most cilia and/or

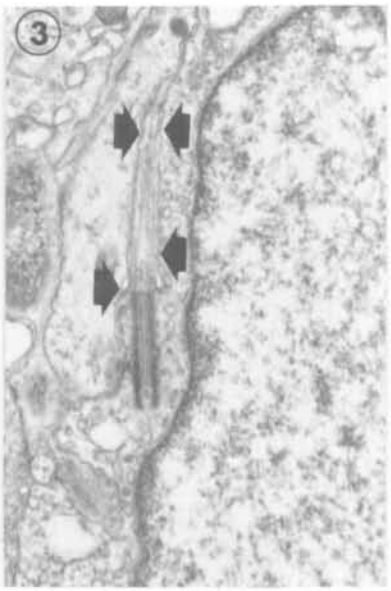

Fig. 3. Single cilium passing into the extracellular space between two adjacent meshwork cells. Notice the close apposition (arrows) of the plasmalemma of the cilium to that of both an adjacent cell and the cell from which it issues. ×17,000.

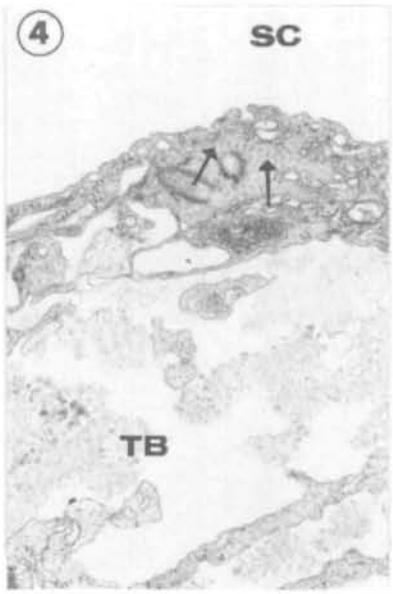

Fig. 4. Striations (arrows) surrounding a centriole pair in a cell of the inner wall of Schlemm's canal. ×12,500.

centrioles. There were fewer of each in the scleral fibrocytes, smooth muscle of the ciliary body, pericytes of the capillaries in the posterior angle, and Schwann cells accompanying innervation into the trabecular meshwork.

Several configurations of cilia and centrioles were observed in the meshwork region. The typical single cilium, identified by the projection of a cilium from one of two perpendicularly opposed centrioles, was often seen. In a few cases we were unable to confirm, by serial section, the occurrence of paired centrioles even though the pair should have a close spatial relationship. Thus, single cilia originating from unpaired centrioles apparently occur in the trabecular meshwork.

Another configuration commonly encountered was the apparent occurrence of two centrioles that are aligned parallel to one another rather than perpendicular to one another (Fig. 1). Only extensive serial sectioning would reveal whether or not these parallel cilia and centrioles are faced on end by a single or double perpendicular centriole, but from our data it is apparent that two cilia with parallel fibers can leave the cell surface adjacent to one another. The frequency of occurrence of parallel centrioles with any other microtubular structure nearby would indicate that there are single cilia arising from single centrioles while other observations indicate that two cilia may occur in conjunction with a single perpendicular centriole.

A more striking, but less common configuration, is three cross-sectioned centrioles located adjacent to one another in a single cell (Fig. 2). We have never seen a section in which three parallel cilia issue from a single area of a single cell. The significance of these special arrangements in terms of centriolar division and ciliation of paired bodies is not clear. The largest number of separate centrioles observed in close association in a single cell was four and this unusual case appeared to represent the division of a centriolar pair.

The cell-cilium spatial relationship is as important as centriole structure. Cilia passing from a free surface of the cell make contact with adjacent cells (Fig. 3), but do not appear to undergo extensive membrane specialization. In localized areas there are desmosome-like plaques. Although we have never seen a situation in which a cilium was located intracellularly, it is apparent from Fig. 3 that cilia can become enmeshed in cell-cell relationships. Single cilia typically have a flaccid cell membrane and tend to be short and relatively formless once they leave the cell. Cilia were seen to pass to the cell base where they associated with the basal lamina. We were not able to tell whether or not the cilia themselves are actively involved in the production of basal lamina, and whether or not they form true hemidesmosomes, however, localized cell membrane thickenings occur along the edge facing the basal lamina.

Single cilia occurring in some cell types have been described as having ciliary rootlets, but none

of the cilia seen in our study of the trabecular meshwork have such a configuration. The centriole pair may occur in a cell region marked by definite striations (Fig. 4), but there is no indication that these are associated with the centriole pair. The cell region containing the centriole may be highly filamentous and these filaments may show periodic increased densities, but none of these appear to be true ciliary rootlets. Microtubules are also commonly seen in the cell region containing the centriole pair.

Discussion. Although the density distribution of single cilia in the meshwork region could be an artifact of cell distribution, that is not apparent from our data. The mesothelial cells of the uveal and corneoscleral meshwork seem to exhibit more types of single cilia and a wider variety of morphologic configuration of these cilia than do other cells described in the literature.

The importance of all the combinations of cilia and centrioles is difficult to approach in a study of pathological material alone. However, it is apparent that the concept of the typical mesothelial primary cilium, one arising only from the centriole pair, does not hold in the case of the material reported here. In fact, there appears to be such a great variety of forms within single areas of single biopsies that the only logical definition lies in a reconsideration of the potentials of cells noted to produce single cilia.

Factors responsible for this variation might be aging, disease state, drug use, or perhaps some environmental factor correlated with the patients undergoing surgery. On the basis of our biopsy study there appears to be no difference due to the two disease states compared, and the occurrence of cilia does not appear to be a technical artifact due to a particular fixative schedule.

The occurrence of several configurations of cilia in the trabecular meshwork casts doubt on their potential functional role. If these are sensory cilia as indicated by other authors, how can their widely differing morphology and cell-cilium spatial contact explain a single sensory function? The cell membrane surrounding each cilium appears to be capable of behaving in the formation of cell-cell contacts and in the interaction of the cell to the basal lamina. That fits nicely with the hypothesis that the single (primary) cilium is a "primitive" remnant of generalized cell morphology.

We are grateful for the technical assistance given us by Ms. Carole Ames, Veterans Administration Hospital, Gainesville, Fla.

From the Veterans Administration Hospital, and the Division of Ophthalmology, University of California at San Diego Medical School, San Diego, Calif. Supported by and conducted at the Veterans Administration Hospital, Gainesville, Fla. Submitted for publication Sept. 8, 1975.

REFERENCES

1. Scherft, J. P., and Daems, W. F.: Single cilia in chondrocytes, J. Ultrastruct. Res. **19**: 546, 1967.
2. Allen, R. A.: Isolated cilia in inner retinal neurons and in retinal pigment epithelium, J. Ultrastruct. Res. **12**: 730, 1965.
3. Shabo, A. L., and Maxwell, D. S.: The structure of the trabecular meshwork of the primate eye: a light and electron microscopic study with peroxidase, Microvasc. Res. **4**: 384, 1972.
4. Inomata, H., Bill, A., and Smelser, G. K.: Aqueous humor pathways through the trabecular meshwork and into Schlemm's canal in the cynomolgous monkey *(Macaca irus),* AM. J. Ophthalmol. **73**: 760, 1972.
5. Wulle, K. G.: The development of the productive and draining system of the aqueous humor in the human eye, Adv. Ophthalmol. **26**: 296, 1972.
6. Vegge, T.: The fine structure of the trabeculum cribiforme and the inner wall of Schlemm's canal in the normal human eye, Z. Zellforsch. **41**: 193, 1967.
7. Hogan, M. J., Alvarado, J. A., and Weddell, J. R.: Histology of the Human Eye, Philadelphia, 1971, W. B. Saunders.
8. Fine, B. S.: Observations on the drainage in man and rhesus monkey: a concept of the pathogenesis of chronic simple glaucoma, INVEST. OPHTHALMOL. **3**: 609, 1964.
9. Holmberg, A. S.: Schlemm's canal and the trabecular meshwork. An electron microscope study of the normal structure in man and monkey *(Cercopithecus ethiops),* Doc. Ophthalmol. **19**: 339, 1965.

The adrenergic receptors of the intraocular muscles of the human eye.

G. W. H. M. VAN ALPHEN.

To the Memory of Dr. Ludwig von Sallmann.

The adrenergic receptors in man were analyzed using isolated sphincter, dilator, and ciliary muscle strips, dissected from eyebank eyes. The dilator is mainly α, the sphincter both α and β, and the ciliary muscle predominantly β adrenergic.

In previous work[1-3] we analyzed the distribution of the adrenergic receptors of the internal muscles of the eye in three species. The results are shown in Table I.

The receptors in the human eye remain unknown since fresh material is hard to obtain. When it appeared that muscles of cat eyes would still respond after 12 to 24 hours of refrigeration at 4° C., a trial was made with eyebank eyes. It was found that from some eyes muscular responses could be obtained up to four days after death. There were enough eyes that adequately