inhibits the appearance of PMNs in the aqueous humor undoubtedly inhibits inflammatory responses in the eye. However, because of the difficulty in obtaining representative samples of aqueous humor, it is conceivable that less potent but potentially useful anti-inflammatory compounds, which are evaluated in this way, could be overlooked.

Key words: inflammation, endotoxin, eye, polymorphonuclear leukocytes, myeloperoxidase, aqueous humor, iris-ciliary body

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References


Protective Barrier Effect of the Posterior Lens Capsule in Exogenous Bacterial Endophthalmitis—An Experimental Primate Study

Todd L. Beyer, George Vogler, Dian Sharma, and Francis E. O'Donnell, Jr.

Ten eyes of five Rhesus monkeys underwent extracapsular lens extraction. The right eye of each monkey was allowed to retain an intact posterior capsule. The left eye of each monkey had a wide primary capsulectomy with minimal anterior vitrectomy. In order to exclude operative contamination, we waited 2 to 3 weeks later to challenge the eyes with bacteria. Seventy-two hours after anterior chamber injection of equal numbers of Staphylococcus aureus, diagnostic cultures were obtained from the anterior chamber and vitreous and correlated with the clinical findings. Injection of 10,000 S. aureus produced culture-positive endophthalmitis in eyes that had undergone posterior capsulectomy, but it failed to produce endophthalmitis in fellow eyes with intact posterior capsules. This suggest that a significant barrier effect against the development of bacterial endophthalmitis exists in the eyes with intact posterior capsules. Invest Ophthalmol Vis Sci 25:108-112, 1984

Exogenous, bacterial endophthalmitis is a disastrous complication of intraocular surgery. Visual prognosis in this disease remains relatively poor even with aggressive therapeutic intervention using intracameral antibiotics and early vitrectomy.1-3 The best treatment remains prevention. Recently there has been renewed interest in extracapsular cataract extraction techniques, primarily because of the popularity of posterior chamber intraocular implants. It is the purpose of this investigation to evaluate the role that the primate posterior capsule may play in inhibiting the development of endophthalmitis by confining the infected material to the anterior chamber. A barrier effect of the posterior capsule in the rabbit model has been previously suggested by Katz and Forster in an unpublished communication.

Materials and Methods. Ten eyes of five monkeys underwent extracapsular cataract extraction. On the day prior to the operative procedures, each monkey was examined, while under ketamine sedation, for lens, retinal, or vitreous abnormalities with the direct and
indirect ophthalmoscopes. At the end of the examination, the periorbital hair was removed and the eyelashes trimmed. A generous amount of 0.5% chloramphenicol ointment was applied to each eye. On the day of surgery, each monkey was given a preoperative injection of ketamine and then administered a general anesthetic utilizing halothane and oxygen. The eyes of each monkey were then dilated with 2.5% phenylephrine and 1% cyclopentolate. The entire face, both eyelids and lid margins were then prepped carefully with povidone-iodine and the conjunctival fornices were irrigated with sterile saline. Next, sterile drapes were applied and a pediatric wire lid speculum inserted. A 4-0 silk suture was then passed beneath the superior rectus muscle and clamped to the drape with a sterile hemostat. A fornix-based conjunctival flap was fashioned under microscopic control and bleeders cauterized with bipolar wet-field cautery.

A grooved incision was made with a razor blade and two horizontal 8-0 silk mattress sutures were placed at the 1:00 and 11:00 o’clock positions. A small puncture into the anterior chamber was made with the razor blade and an irrigating cystotome was inserted for the large (7 mm) round anterior capsulotomy. The preplaced sutures were looped to the side and the incision was extended and completed with curved left and right corneal scissors. The soft nucleus was expressed as completely as possible, and then the wound was closed by tying the preplaced sutures. The anterior chamber was reformed with balanced salt solution, and the cortex was removed completely with the 0.2 mm aspiration tip McIntyre needle on a 6-cc Monoject syringe. The posterior capsule of each right eye was punctured 8-0 silk suture and the fornix-based conjunctival flap hooded over the incision and anchored with 6-0 plain gut suture. The anterior vitreous was left undisturbed if possible. No peripheral iridectomies were performed. The wound was then closed with interrupted 8-0 silk suture and the fornix-based conjunctival flap hooded over the incision and anchored with 6-0 plain gut suture. Each eye received gentamicin sulfate 10 mg and triamcinalone 2.5 mg, subconjunctivally, at the end of the procedure. A combination steroid/antibiotic and atropine 1% ointments were applied, and the animals were returned to their cages.

After a period of 2 weeks, all eyes were re-examined and clinical findings recorded. All eyes were found to be quiet with clear media. Conjunctival chemosis and/or hyperemia were absent. No eye had any clinical sign of endophthalmitis.

The inocula for intraocular injection were prepared in the following manner. A strain of Staphylococcus aureus (25923) was obtained from the American Type Culture Collection. The culture was monitored periodically for purity. Organisms were maintained on Blood Agar plates, (St. Louis Biological Laboratories, Inc., St. Louis, Missouri) and subcultured every 72 hours to assure viability. Twenty-four-hour-old cultures were used in preparing the inocula for all animal experiments. At prescribed time intervals, 24 hour cultures of the above described organisms were suspended in sterile saline. To assure a single cell suspension, the preparations were subjected to a vortex mixer for 30 seconds. Cell counts were performed by diluting the vortexed suspensions in sterile saline with a Tri-Lyne (Dade) erythrocyte pipette. The diluted suspensions were counted in a Neubauer hemocytometer. Appropriate dilutions were made accordingly to obtain $10^3$ or $10^4$ organisms/0.05 cc sterile saline. Microbial concentrations were verified at the time of preparation by inoculating known volumes of the suspension on blood agar plates and performing counts after 24 hours. An additional inoculation was made onto blood agar plates at the same time as anterior chamber injection to assure viability of the inocula.

After ketamine HCL injection, each monkey was brought to the operating suite, where a topical anesthetic agent was administered and a wire pediatric speculum inserted. A bevel up injection was carefully made into the anterior chamber through the surgical wound adjacent to one of the silk mattress sutures with a 27-gauge needle on a 0.5 cc calibrated tuberculin syringe (Fig. 1). Both eyes of monkey #1 received a single injection of a suspension of 1,000 Staphylococcus aureus organisms in 0.05 cc saline into the anterior chamber. The needle was carefully withdrawn, and a cotton tip applicator gently applied to minimize leakage. The remaining four monkeys (#2-#5) received an injection of 10,000 organisms/0.05 cc at 2½ to 3 weeks post-lens extraction.

After inoculation, each animal was examined by two observers who had no previous knowledge as to which eye had been capsulectomized. Their clinical handlight exams were recorded every 24 hours for 72 hours (Fig. 2). At 72 hours after inoculation, all animals were sacrificed, and the anterior chamber and central vitreous cavities were separately cultured using aseptic techniques.

Results. (Tables 1 and 2). Monkey #1 had inoculation of each eye with 1,000 Staphylococcus aureus organisms and failed to develop anything more than a mild anterior chamber reaction in both eyes, which cleared totally within 48 hours. Cultures of both the anterior chamber and vitreous in both eyes were sterile at 72 hours.

The remaining 8 monkey eyes were inoculated with 10,000 Staphylococcus aureus organisms, and each left eye developed a clinically obvious endophthalmitis within 72 hours (Fig. 2). Most often this was obvious
Fig. 1. Technique of anterior chamber inoculation with Staphylococcus aureus. A, in right eye with intact posterior capsule and B, in the left eye, which had a posterior capsulectomy.

at 48 hours, and it was characterized by the presence of marked edema, chemosis, purulent conjunctival drainage, corneal haze with vitreous opacification, and pupillary membranes. As time progressed, all left eyes got much worse. In contrast, all right eyes developed an anterior chamber reaction greatest at 24 hours, which proceeded to clear rapidly by 72 hours. The vitreous remained clear. No lid edema, purulent conjunctival drainage, corneal haze, pupillary membrane, or vitreous opacification developed in any of the right eyes. At the time of sacrifice (72 hours post injection), all eyes (right and left) had light growth or no growth from anterior chamber aspirates. Eyes with intact posterior capsules had no growth from the central vitreous.

Table 1. Clinical findings

<table>
<thead>
<tr>
<th>Monkey</th>
<th># Bacteria injected into AC</th>
<th>24 hours after inoculation</th>
<th>48 hours after inoculation</th>
<th>72 hours after inoculation</th>
</tr>
</thead>
<tbody>
<tr>
<td>#1</td>
<td>OD 10^3</td>
<td>Moderate AC reaction</td>
<td>Mild AC reaction</td>
<td>Media clear</td>
</tr>
<tr>
<td></td>
<td>OS 10^3</td>
<td>Moderate AC reaction</td>
<td>Mild AC reaction</td>
<td>Media clear</td>
</tr>
<tr>
<td>#2</td>
<td>OD 10^4</td>
<td>Moderate AC reaction</td>
<td>Moderate AC reaction</td>
<td>Endophthalmitis</td>
</tr>
<tr>
<td></td>
<td>OS 10^4</td>
<td>Moderate AC reaction</td>
<td>Endophthalmitis</td>
<td>Endophthalmitis</td>
</tr>
<tr>
<td>#3</td>
<td>OD 10^4</td>
<td>Moderate AC reaction</td>
<td>Media clear</td>
<td>Media clear</td>
</tr>
<tr>
<td></td>
<td>OS 10^4</td>
<td>Moderate AC reaction</td>
<td>Endophthalmitis</td>
<td>Endophthalmitis</td>
</tr>
<tr>
<td>#4</td>
<td>OD 10^4</td>
<td>Moderate AC reaction</td>
<td>Media clear</td>
<td>Media clear</td>
</tr>
<tr>
<td></td>
<td>OS 10^4</td>
<td>Moderate AC reaction</td>
<td>Endophthalmitis</td>
<td>Endophthalmitis</td>
</tr>
<tr>
<td>#5</td>
<td>OD 10^4</td>
<td>Moderate AC reaction</td>
<td>Media clear</td>
<td>Media clear</td>
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<tr>
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<td>OS 10^4</td>
<td>Moderate AC reaction</td>
<td>Endophthalmitis</td>
<td>Endophthalmitis</td>
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</tbody>
</table>
cavity. All eyes with posterior capsulectomy had positive cultures from the central vitreous cavity.

**Discussion.** It is apparent from this experimental study that the posterior lens capsule inhibits the development of vitreous infection. It would seem likely that this is due to a barrier effect, which helps to confine the bacteria to the anterior chamber. As in the human situation, we found that the anterior chamber aspirate could be negative despite heavy growth from the vitreous aspirate. Apparently, the anterior chamber is better able to control bacterial infection than is the vitreous cavity. Noteworthy is the fact that three right eyes (animals 2, 3, 5) had positive cultures from the anterior chamber aspirate despite a negative vitreous culture and negative clinical appearance for endophthalmitis. This finding suggests that it is valuable to obtain both anterior chamber and vitreous cultures, especially in cases of early suspected endophthalmitis after extracapsular surgery where microorganisms may yet be confined to the anterior chamber.

Although this experimental study demonstrates the ability of the intact posterior capsule to inhibit spread of infection, it would be wrong to extrapolate from this study to the clinical situation and to conclude that planned extracapsular surgery with an intact posterior capsule is less likely to cause bacterial endophthalmitis than is intracapsular surgery. In general, when authors have compared intracapsular infection rates to extracapsular rates, they were comparing intracapsular to unplanned extracapsular rates. Allen, Mangiaracine, and others reported the incidence of postoperative infection was increased with extracapsular surgery, the majority of which were unplanned. Similarly, Christy and Lall reported an unplanned extracapsular infection rate of 1.31% that was three times higher than their intracapsular series of 0.41%. They felt that increased intraocular manipulation associated with an unplanned extracapsular extraction predisposed to a greater risk of exogenous endophthalmitis.

It seems doubtful that the infection rate for unplanned, extracapsular extractions is equivalent to that of modern, planned extracapsular extraction. For example, infection rates have been reviewed in the extracapsular technique of phacoemulsification. Reynolds reported an incidence of 0.4% (2 in 500 cases) of infectious endophthalmitis following phacoemulsification. These cases were associated with a vitreous wick syndrome. Kratz reported an incidence of bacterial endophthalmitis of 0.1% in a large series of 2,000 cases of phacoemulsification, of which an unspecified number had received a primary capsulotomy or capsulectomy. It may be that modern, planned extracapsular surgery, performed using appropriate prepping and draping, which excludes contaminated lashes and lid margins from the operative field, and executed under microscopic control, thus allowing for more complete removal of cortical material while reducing the possibility of inadvertent disruption of the posterior lens capsule and resultant admixture of formed vitreous, does reduce the chances of bacterial endo-

<table>
<thead>
<tr>
<th>Monkey</th>
<th># Bacteria inoculated into AC</th>
<th>Anterior chamber (AC) culture at 72 hours</th>
<th>Vitreous culture at 72 hours</th>
</tr>
</thead>
<tbody>
<tr>
<td>#1</td>
<td>OD $10^3$</td>
<td>Negative</td>
<td>Negative</td>
</tr>
<tr>
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<td>Negative</td>
<td>Negative</td>
</tr>
<tr>
<td>#2</td>
<td>OD $10^4$</td>
<td>Light growth</td>
<td>Negative</td>
</tr>
<tr>
<td></td>
<td>OS $10^4$</td>
<td>Light growth</td>
<td>Moderate growth</td>
</tr>
<tr>
<td>#3</td>
<td>OD $10^4$</td>
<td>Light growth</td>
<td>Negative</td>
</tr>
<tr>
<td></td>
<td>OS $10^4$</td>
<td>Light growth</td>
<td>Light growth</td>
</tr>
<tr>
<td>#4</td>
<td>OD $10^4$</td>
<td>Negative</td>
<td>Negative</td>
</tr>
<tr>
<td></td>
<td>OS $10^4$</td>
<td>Negative</td>
<td>Heavy growth</td>
</tr>
<tr>
<td>#5</td>
<td>OD $10^4$</td>
<td>Light growth</td>
<td>Negative</td>
</tr>
<tr>
<td></td>
<td>OS $10^4$</td>
<td>Light growth</td>
<td>Heavy growth</td>
</tr>
</tbody>
</table>

Fig. 2. Clinical appearance at 72 hours of monkey #4, demonstrating the absence of external signs of endophthalmitis in the right eye, which has an intact posterior capsule, and the presence of obvious clinical endophthalmitis in the left eye, which had a posterior capsulectomy.
Endophthalmitis in comparison to an intracapsular operation. The answer to this very important question would require a large clinical series, since the incidence of bacterial endophthalmitis is fortunately low. There is a significant barrier effect produced by an intact posterior lens capsule, which inhibits the spread of bacterial infection from the anterior chamber to the vitreous cavity in the primate model. It is not possible from the data obtained in this study to answer the controversy over whether or not modern extracapsular surgery is superior to intracapsular surgery in the prevention of bacterial endophthalmitis. Despite the demonstration of a significant barrier effect to endophthalmitis produced by the posterior lens capsule, the two techniques probably vary in the frequency and/or size of inadvertent bacterial inocula introduced during surgery. A large, multicenter clinical study is necessary to answer this important question.

Key words: Endophthalmitis, cataract, posterior capsule

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References

Characterization of Somatostatin-like Immunoreactivity in Vertebrate Retinas

David Marshak* and Tadaraka Yamada†

Large differences in retinal concentration of somatostatin-like immunoreactivity (SLI) were observed even among closely related species. Hog and chicken retinas, like those of goldfish and frog described previously, contained roughly equal amounts of SLI coeluting with somatostatin tetradecapeptide (S14) and octacosapeptide (S28) on Sephadex G 50 chromatography. In contrast, virtually all of the SLI from rat retina coeluted with S14, and nearly all of the bovine retinal SLI coeluted with S28. These species differences may reflect differences in post-translational processing of the various molecular forms of retinal SLI. Invest Ophthalmol Vis Sci 25:112–115, 1984.

Somatostatin-like immunoreactivity (SLI) in the retina has many of the properties of neurotransmitters SLI is synthesized in the retina1 and stored in its intrinsic neurons (review in reference 2). Retinal SLI can be released by depolarizing stimuli in the presence of calcium, apparently by the same mechanism as neurotransmitters and other secretagogues.3 The retina is an excellent system to test whether somatostatin duplicates the natural transmitter's actions on postsynaptic cells. Such studies would be facilitated by elucidation of the structures of retinal SLI. This survey of retinal SLI in various vertebrates provides preliminary information about the structures of these molecules.

Materials and Methods. Eyes from cows, rabbits, chickens and carp were dissected within 5 min of death.