


Endothelial damage from intraocular lens insertion. Herbert E. Kaufman and Jeffrey I. Katz.

Previous studies have shown that approximately 40 per cent of the corneal endothelial cells can be lost at the time of intraocular lens insertion. Momentary contact between the methacrylate surface and the endothelial cells causes an adhesion between these surfaces and results in extensive cell damage upon separation of the surfaces. This type of damage appears to be due to a biophysical interaction between these surfaces and may be avoided by altering the surface of the lens.

Recent studies at the University of Florida by Bourne and Kaufman and Forstot and associates have demonstrated that there is a 40 to 50 per cent cell loss of the central corneal endothelium after intraocular lens insertion. This loss was apparent immediately following surgery, did not progress with time, and was independent of the type of intraocular lens implanted. In contrast, standard cataract extraction in the fellow eyes of the pseudophakes caused a 7 to 8 per cent central endothelial cell loss. Although surgical manipulation must be responsible for some cell loss, there appears to be some other explanation for the marked difference between intraocular lens implantation and standard cataract extraction. We felt that there might be cell damage caused by the interaction of the methacrylate surface with the normal corneal endothelium. A study was therefore undertaken to assess the kind of damage that could be expected from brief contact between intraocular lenses and the corneal endothelium.

**Methods and materials.** Adult albino rabbits were killed by an intravenously administered overdose of pentobarbital sodium, and the eyes were enucleated immediately upon death. The corneas were removed with a 3 mm. rim of sclera, and the lens-iris diaphragm peeled from the cornea. The corneas were then placed endothelial side up on a Teflon block, and the central areas punched out with a 7 mm. trephine. An identical procedure was employed in isolating 7 mm. corneal buttons from fresh human eyes. Only paired (2 eyes from the same donor) human eyes were used, so that one eye could serve as a control for the other. Corneas were then gently placed with and without-rubbing on methyl methacrylate intraocular lenses as described below, and endothelial damage determined by either nitroblue tetrazolium (NBT) staining or scanning electron microscopy (SEM).

**NBT staining.** Corneas were incubated at 37° C. for 15 minutes in NBT stock solution with 0.3 mg./ml. diphosphopyridine nucleotide, reduced (DPNH) as a substrate. The solution stains oxidative enzymes in the cytoplasm of cells. Intact cells do not stain by this method, but cells with disrupted cell membranes stain dark blue. Cell damage was then estimated by observation under low-power microscopy.

**Scanning electron microscopy.** Corneas prepared for this study were fixed in 2.5 per cent cold glutaraldehyde for 1 hour and post-fixed in 1 per cent osmium tetroxide for 90 minutes. Corneas were dehydrated in increasing concentrations of ethyl alcohol and Freon TF, and finished in the Bomar critical point apparatus. Specimens were then coated with palladium-gold and viewed with a Zeiss Novascan 30.

**Results.**

**Normal control.** Six rabbit corneal buttons were isolated as described above, four of which were immediately stained with NBT and the remaining two prepared for SEM. The only damage identifiable in all six corneal buttons subjected to an isolation technique alone was minimal damage of the edges of the button. No corneas demonstrated, by either NBT staining or SEM, damage central to the edges of the buttons.

**Effect of contact between animal lens and endothelium.** Rabbit and human corneal buttons were grasped on the edge with a forceps and the endothelial side gently rubbed on isolated rabbit and human lens, respectively, for 15 and 60 seconds (Fig. 1). The only significant endothelial damage noted by NBT staining or SEM was a small peripheral area corresponding to forceps fixation. An occasional cell in the human corneas stained darkly with NBT.

**Rabbit endothelial damage following contact with methacrylate.** Five rabbit corneal buttons were grasped on the edge and gently placed on a 100 per cent methyl methacrylate surface which had first been dipped in balanced salt solution (Tis-U-Sol). Contact time varied from 1 to 60 seconds. An additional five rabbit corneas were placed on the methacrylate for similar periods of time and were continuously but gently rubbed back and forth during the time of contact. The methacrylate surface was a highly polished half-
sphere with a radius of curvature of 7.3 mm., mounted on a post so that it could be firmly secured. It had not been sterilized or treated with caustics. This surface facilitated total contact between the corneal buttons and the methacrylate and eliminated sharp edges which could cause endothelial damage.

All rabbit corneas which were placed on the methacrylate dipped in balanced salt solution showed significant cell damage both by NBT staining and SEM. Although the corneas exposed to the methacrylate for 60 seconds showed the greatest damage (approximately 30 to 35 per cent of the endothelial cells stained darkly with NBT), even corneas which had made contact for only 1 second suffered extensive damage (approximately 20 to 25 per cent). Rubbing the corneas on the methacrylate increased the damage slightly, but it was clear that the majority of endothelial injury was sustained immediately upon contact. When viewed by SEM, the corneas appeared as if the cell membranes had been torn off the damaged endothelial cells, thus exposing the cell nuclei.

Human endothelium following touch to methacrylate. One human cornea was placed on methacrylate (dipped in balanced salt solution) for 15 seconds and another one for 45 seconds. SEM revealed that the two corneas placed on methacrylate dipped in balanced salt solution suffered extensive damage, with the cell membranes of the endothelium having been ripped off (Fig. 2, A and B).

Touch to commercial intraocular lenses. Rabbit and human corneas were placed on several types of commercially available intraocular lenses. These consisted of (1) Medallion lens (Medical Workshop, Holland); (2) McGhan No. 52 lens (Santa Barbara, Calif.); (3) Binkhorst 2-loop and 4-loop lens (McGhan); and (4) Copeland lens (CopeLand Intra Lenses, New York, N. Y.). Contact times varied between 1 and 60 seconds. Lenses were dipped in balanced salt solution prior to contact with the corneal button. Both NBT staining and SEM were performed.

Results in this portion of the investigation were quite similar to the earlier results obtained with the methacrylate, in that all intraocular lenses employed caused extensive endothelial damage. Occasionally, the edge of an intraocular lens rubbed the endothelium while the cornea was being placed on the lens, and this caused a ring of damage conforming to the edge of the lens. In several instances we noted holes through the endothelium and Descemet's membrane which were not consistent with the type of damage caused by the smooth methyl methacrylate surface, and, therefore, scanning microscopy of one
intraocular lens was performed. This type of mechanical injury was totally different from that of the methacrylate contact in that cells were wiped off in mechanical injury but cell remnants persisted after contact with methacrylate.

Examination of the lens surface by scanning electron microscopy.
1. A platinum-type intraocular lens was prepared by mounting it on a scanning stub and coating it with palladium-gold. SEM revealed that the iris loop, which appeared to be fixed to the posterior surface of the lens, actually protruded through the anterior surface. It is apparent that these protuberances on the anterior surface of the lenses are capable of producing endothelial damage, with corneal contact, and this probably explains the occasional holes seen in Descemet's membrane. The same problem of anterior protrusion of the loops was seen in a number of other commercially available intraocular lenses.
2. A rabbit corneal button was placed on a Binkhorst 2-loop intraocular lens for 15 seconds and then gently removed. SEM showed that the endothelial cell membranes remained adherent to the lens surface (Fig. 3).

Prevention of endothelial damage with soft contact lens. In order to determine if substances other than methacrylate were capable of causing endothelial damage such as was observed in the previous portions of the investigation, rabbit and human corneal buttons were placed on a glass sphere and a stainless steel surface for periods varying between 1 and 45 seconds. NBT staining revealed significant staining of the endothelial cells in areas of contact with both the glass and the stainless steel.

The central portion of a Bausch and Lomb T series soft contact lens was trephined with a 3 mm corneal trephine. This circular portion of the soft lens was then dipped in tissue culture Medium 199 and placed on the glass sphere. Rabbit and human corneal buttons were gently placed on the lens-glass surface for periods varying between 1 and 45 seconds. NBT staining revealed that the central corneal endothelium which had made contact with the soft lens suffered practically no damage, whereas the peripheral area of the corneal button which was in contact with glass only, suffered extensive endothelial damage.

Discussion. Although there are certainly many factors which can cause endothelial cell damage at the time of surgery, such as corneal manipula-

Fig. 2, A. Scanning electron micrograph of human corneal endothelium following touch to intraocular lens (dipped in balanced salt solution) for 15 seconds. Anterior membranes of cells have been torn off. (x600.)
Fig. 2, B. Different area. (×700.) For legend see Fig. 2, A.

Fig. 3. Scanning electron micrograph of the surface of platina-type intraocular lens after touch to rabbit corneal endothelium. Endothelial cell membranes are adherent to lens surface. (×350.)
tation and irrigation of the anterior chamber, and although lens design with surface protuberances may be of some importance, it is clear that a biophysical interaction between methacrylate and the corneal endothelial surface produces extensive cell damage. It appears that the methacrylate adheres instantaneously to the endothelial surface, and with separation of the two surfaces the anterior membranes of the endothelial cells are torn off. The membranes are seen, by SEM, to remain adherent to the methacrylate surface.

Other substances such as glass and stainless steel are capable of producing similar endothelial damage. In contrast, the natural lens appears to cause no damage when it is touched to the endothelium. We feel, therefore, that this is a surface problem of these materials and is not due to the chemical makeup of the materials per se. When a portion of the glass sphere is covered with a soft lens, corneal endothelial damage occurs only in areas of direct contact between the endothelium and the glass.

If, in fact, the intraocular lens could be kept away from the endothelium and the chamber never lost, during lens implantation, the type of damage described in this investigation would not occur. It has been our experience, however, that placement of the intraocular lens into the eye with removal of the lens holder and suturing of the wound, is not always possible without brief touch between the endothelium and the lens. The risk of such touch is greatly increased when the vitreous face bulges forward. Because of the conclusive evidence that contact (no matter how brief) between lens and corneal endothelium can produce significant endothelial damage, we undertook a second investigation to see if we could modify the surface of the lens. We found that a variety of substances could prevent endothelial damage when the lens was dipped into the substance prior to being touched to the endothelium. Two patients have undergone uneventful lens implantation after the lens was dipped into protective solutions and cell loss was 11 per cent with a methylcellulose solution and 9 per cent with a polyvinyl pyrrolidone solution.3

The importance of endothelial damage at the time of surgery is not absolutely certain, but it is clear that if damage is extensive enough, corneal edema supervenes. Since the human corneal endothelium seems not to regenerate, it may be that if enough endothelial cells are lost at the time of surgery, even if the cornea clears, the death of additional endothelial cells with aging may result in an area of endothelial surface that can no longer be covered by the spreading cells. It seems likely, therefore, that late corneal edema may result in at least some of the patients.

If endothelial contact with the methacrylate surface is a major cause of endothelial damage, and if such damage can be easily eliminated, it may not be necessary to withhold intraocular lens insertion from patients with mild endothelial changes. Also, the age criterion for lens insertion may become more flexible if further studies confirm our findings that intraocular lenses do not cause progressive damage to the endothelium and that the major cause of endothelial cell death at the time of surgery can be eliminated.


REFERENCES


A new technique for the vital staining of the corneal endothelium. DAVID J. SPENCE AND GHOLAM A. PEYMAN.

A new technique for the staining of the corneal endothelium combines the actions of the vital stain trypan blue and the intercellular stain alizarin red S. The technique is an improvement over former staining methods, because it defines both viable and nonviable cells and thereby permits an investigator to accurately quantify endothelial cell damage.

The vital stain trypan blue1-4 has been used for many years in the assessment of corneal endothelial viability. The technique is inexpensive and simple, and it is well suited for an overall determination of the endothelial status.

The disadvantage of the trypan blue method is that it is not as specific as is desirable. The dye enters and stains only those cells whose membrane permeability has been increased by cell wall damage. Healthy cells are therefore not delineated, and an accurate quantification of cell damage by grid counts is impossible. Using trypan blue, an investigator can only roughly estimate the