M-K medium and postmortem cytologic damage

Perry S. Binder and M. Gary Wickham

Freshly enucleated rabbit eyes were refrigerated at +4°C under standard eye bank conditions for 2, 6, 9, and 21 days. One group of corneas with a scleral rim were excised and placed in M-K medium, stored for 18, 24, or 48 hr at +4°C; they were then removed, and endothelial cell viability was evaluated with nitroblue tetrazolium. The cells were examined by light microscopy and scanning and transmission electron microscopy. A second group of corneas were similarly obtained and then used as donor corneas from 6 mm transplants. Each recipient rabbit was evaluated daily by slit-lamp biomicroscopy and corneal pachometry. Fourteen days postoperatively, the rabbits were sacrificed, the eyes enucleated, and the excised corneas were evaluated in a fashion similar to those of group I. M-K medium storage protected the morphology and functional integrity of the rabbit corneal endothelium up to 48 hr beyond moist chamber storage for 2, 6, and 9 days. However, M-K medium appeared to have no such effect on corneas that had been moist chamber–stored for 21 days. These results suggest that some human corneas with a prolonged time from death to moist chamber storage may be utilized for corneal transplantation after further storage in M-K medium.

Key words: corneal preservation, corneal endothelium, corneal transplantation, M-K medium

Following the introduction of corneal storage in tissue culture medium 199 with 5% dextran (M-K medium) by McCarey and Kaufman, there was a rapid acceptance of this technique due both to its simplicity and to early reports touting the good quality of donor tissue. Initial laboratory studies using rabbit corneas demonstrated that M-K medium was able to maintain endothelial integrity for up to 14 days of storage at +4°C. With human corneas, the medium appeared to provide tissue whose morphology appeared better than that of corneas stored under standard moist chamber conditions for a similar period of time. More recent studies using cat corneas, rabbit corneas, and eye bank eyes also show that M-K medium is able to maintain endothelial structural integrity. Storage for up to 4 days at +4°C produced tissue which was morphologically and functionally superior to that stored in moist chamber, but storage longer than 96 hr did not maintain the integrity of the corneal endothelium. Some authors concluded that the most important aspect of this technique of storage is that corneas be excised from the globes (and thereby removed from the stagnant aqueous) and placed in the medium as soon as possible after death.

At present, moist chamber–stored corneas are utilized as soon as possible after donor death and no later than 48 hr post-mortem. Most of the M-K medium–stored human corneas which have been used for penetrating keratoplasty have been enucleated and stored in the medium soon after death. Although immediate M-K storage is the ideal...
Fig. 1. Two days' moist chamber storage. A, Scanning electron micrograph of the endothelium of rabbit cornea stored in a moist chamber for 48 hr. B, Scanning electron micrograph of rabbit endothelium stored for an additional 24 hr in M-K medium. C, Scanning electron micrograph of rabbit endothelium stored in M-K medium for an additional 48 hr. The normal endothelial mosaic is present. D, Transmission electron micrograph of the endothelium of the same cornea in A. E, Transmission electron micrograph of the endothelium of the same cornea as in C. There is mild swelling of the mitochondria, but otherwise, normal ultrastructure is maintained. The increased density along the posterior plasma membrane in D is due to NBT.

Materials and methods

In vitro studies. New Zealand white rabbits (2.5 kg) were euthanized with an overdose of intravenous sodium pentobarbital, and the eyes were immediately enucleated. All eyes were handled as pairs with one eye serving as a control. Whole eyes were stored with standard eye bank techniques after the globes had been immersed in Neosporin solution (Burroughs Wellcome Co., Research Triangle Park, N. C.) and rinsed with normal saline.

The whole eyes were stored in a refrigerator at +4° to +6° C for 2, 6, 9, and 21 days (moist chamber storage). After the storage periods, one cornea from each mate pair was evaluated morpho-
Fig. 2. A, Scanning electron micrograph of rabbit endothelium stored in moist chamber for 6 days. B, Scanning electron micrograph of rabbit endothelium stored for an additional 24 hr in M-K medium. C, Scanning electron micrograph of rabbit endothelium stored for an additional 48 hr in M-K medium. The normal endothelial mosaic is maintained. Scattered throughout the posterior surface are ruptured cells involving less than 1% the total population. D, Light micrograph of rabbit endothelium stored in moist chamber for 6 days. The cells are swollen, and large intracytoplasmic vesicles are easily seen. (Toluidine blue staining.) E, Transmission electron micrograph of endothelium stored for 6 days in moist chamber and 48 hr in M-K medium. There is edema of the cells, with intracytoplasmic vesicles and edema of the mitochondria. The posterior plasma membrane and tight junctions are intact.

logically, and the other cornea was placed in M-K medium (Warner Lambert Pharmaceutical Co., Plains, N. J.) containing 8 μg/ml gentamicin sulfate (Caramycin; Schering Corp., Bloomfield, N. J.) for 18, 24, and 48 hr at +4°C and then examined. The moist chamber-stored cornea (control) and the M-K medium-stored cornea were evaluated (after storage) in vitro with nitroblue tetrazolium (NBT). The percent of endothelial cell viability was calculated by dividing the estimated number of unstained cells by the estimated total number of cells in five different 40x fields.

Eight eyes were treated as 0 time controls, with four each stored for 18, 24, and 48 hr in M-K medium. In all other respects processing was as that used for the experimental eyes.

The corneas were fixed in 2.5% glutaraldehyde in 0.1M phosphate buffer at pH 7.15 for 30 min. They were then placed endothelial side up on a Teflon block, and two 6 mm trephine samples were taken. Both samples were then post-fixed in 2% osmium tetroxide in 0.1M phosphate buffer. One trephine sample from each cornea was dehydrated and processed for scanning electron microscopy (SEM) by the Freon critical point technique. After processing, the samples were mounted on aluminum studs, coated with 60:40 Au: Pd, and examined in an ETEC scanning electron microscope at 20 kV. The second 6 mm trephine specimen was embedded in Epon, sectioned, stained and evaluated by transmission electron microscopy (TEM) with a Zeiss EM-10 transmission electron microscope at 60 kV.

In vivo studies. Corneas stored similarly to
those in the in vitro experiment were used as donors for corneal transplantation. The stored corneas were transferred to a Teflon block, and a 6 mm trephine button was taken. All surgery was done under a Zeiss operating microscope with clean but not sterile techniques. Corneas were sutured in place with 12 interrupted 10-0 monofilament nylon sutures. All animals received 40,000 units of penicillin-streptomycin (Microbiological Associates, Inc., Bethesda, Md.) intramuscularly at the time of surgery. Postoperatively, the animals received topical atropine and neosporin daily. On occasion, 1% prednisolone acetate (Pred Forte; Allergan Pharmaceuticals, Irvine, Calif.) was applied when corneal vascularization occurred. The animals were evaluated daily with a Mackay-Marg tonometer, a slit-lamp biomicroscope, and a corneal pachometer. Although daily measurements were made, corneal clarity and thickness on the first postoperative day were used as indicators of endothelial cell function. Technical failures were eliminated from the study. External color photographs were taken the first and fourteenth postoperative days. Sutures were not removed. The animals were sacrificed on the fourteenth postoperative day with an intravenous overdose of sodium pentobarbital. The eyes were immediately enucleated and fixed in glutaraldehyde for 15 min, and corneas with a 3 mm scleral rim were excised and evaluated.

Results

In vitro studies

NBT. Moist chamber storage for 2 and 6 days did not produce a significant amount of staining. By 9 days, up to 20% of the endothelial cells were staining, and by 21 days almost all the cells stained (Table I). Treatment with M-K medium after moist chamber storage did not decrease the amount of NBT staining. The M-K medium controls all had approximately 5% staining cells.

SEM. There were progressive morphologic changes with each period of storage, but the majority of changes took place after 9 days (Table I). The most constant early change was cellular edema with folds in Descemet's membrane. There were no major changes...
noted in the corneas stored in moist chamber for 2 days (Fig. 1, A), but after 6 days of storage, an increased number of endothelial cells were found to be ruptured and the normally smooth posterior surface became slightly irregular and rough (Figs. 2, A, and 3, A). By 21 days of moist chamber storage, very few cells remained on the posterior corneal surface, and there was severe folding of Descemet's membrane (Fig. 4, A). Removal of 2-, 6-, and 9-day corneas from moist chamber and placement in M-K medium yielded endothelial cells whose morphological features appeared to remain the same over the period of 18 to 48 hr (Figs. 1, B and C, 2, B and C, and 3, B and C). That is, a cornea stored for 2 days in M-K medium following 2 days of moist chamber storage appeared the same as a cornea stored for 2 days in moist chamber alone. This relationship held for 2, 6, and 9 days, although corneas stored for 6 or 9 days plus an additional 48 hr in M-K medium appeared morphologically better than corneas stored in moist chamber only for 6 or 9 days (Table I).

TEM. Progressive postmortem changes occurred in corneas stored 2, 6, and 9 days, but there was essentially no difference between those corneas stored in M-K medium and the control (moist chamber) corneas for that same period (Table I). Corneas stored for 2 days had minimal changes (Fig. 1, D and E), whereas those stored for 6 and 9 days had changes noted by previous authors.\(^1\) - \(^11\) that included swelling of the endothelial cells with an intact posterior membrane, presence of intracytoplasmic vacuoles, decreased cytoplasmic density and mitochondrial edema (Figs. 2, D and E, and 3, D to F). Minimal changes were noted in the M-K medium con-
Table I. In vitro evaluation

<table>
<thead>
<tr>
<th>M-K storage (hr)</th>
<th>No. of eyes</th>
<th>% staining (NBT)</th>
<th>SEM</th>
<th>TEM*</th>
</tr>
</thead>
<tbody>
<tr>
<td>2 days' moist chamber:</td>
<td>0</td>
<td>2</td>
<td>&lt;5</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td>18</td>
<td>4</td>
<td>5-10</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td>24</td>
<td>4</td>
<td>5-10</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td>48</td>
<td>4</td>
<td>5-10</td>
<td>0</td>
</tr>
<tr>
<td>6 days' moist chamber:</td>
<td>0</td>
<td>2</td>
<td>5-10</td>
<td>+2</td>
</tr>
<tr>
<td></td>
<td>18</td>
<td>4</td>
<td>5-10</td>
<td>+1</td>
</tr>
<tr>
<td></td>
<td>24</td>
<td>4</td>
<td>&lt;5-10</td>
<td>+1</td>
</tr>
<tr>
<td></td>
<td>48</td>
<td>4</td>
<td>5-10</td>
<td>+1</td>
</tr>
<tr>
<td>9 days' moist chamber:</td>
<td>0</td>
<td>2</td>
<td>5-10</td>
<td>+3</td>
</tr>
<tr>
<td></td>
<td>18</td>
<td>4</td>
<td>5-15</td>
<td>+1</td>
</tr>
<tr>
<td></td>
<td>24</td>
<td>4</td>
<td>5-20</td>
<td>+1</td>
</tr>
<tr>
<td></td>
<td>48</td>
<td>4</td>
<td>5-20</td>
<td>+1</td>
</tr>
<tr>
<td>21 days' moist chamber:</td>
<td>0</td>
<td>2</td>
<td>50-100</td>
<td>+4</td>
</tr>
<tr>
<td></td>
<td>18</td>
<td>4</td>
<td>40-100</td>
<td>+3</td>
</tr>
<tr>
<td></td>
<td>24</td>
<td>4</td>
<td>60-80</td>
<td>+4</td>
</tr>
<tr>
<td></td>
<td>48</td>
<td>4</td>
<td>100</td>
<td>+4</td>
</tr>
<tr>
<td>M-K controls:</td>
<td>18</td>
<td>2</td>
<td>5</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td>24</td>
<td>2</td>
<td>5</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td>48</td>
<td>2</td>
<td>5</td>
<td>0</td>
</tr>
</tbody>
</table>

*Summarized morphologic changes, with 0 representing no change and +4 representing severe change.

Table II. Transplant evaluation

<table>
<thead>
<tr>
<th>M-K storage (hr)</th>
<th>No. of eyes</th>
<th>Clinical evaluation*</th>
<th>Corneal thickness (mm)</th>
<th>NBT % staining</th>
</tr>
</thead>
<tbody>
<tr>
<td>2 days' moist chamber:</td>
<td>0</td>
<td>5</td>
<td>3 clear/2 cloudy</td>
<td>0.50-0.65</td>
</tr>
<tr>
<td></td>
<td>24</td>
<td>6</td>
<td>6 clear</td>
<td>0.36-0.52</td>
</tr>
<tr>
<td></td>
<td>48</td>
<td>4</td>
<td>4 clear</td>
<td>0.44-0.65</td>
</tr>
<tr>
<td>6 days' moist chamber:</td>
<td>0</td>
<td>4</td>
<td>3 clear/1 cloudy</td>
<td>0.48-0.67</td>
</tr>
<tr>
<td></td>
<td>24</td>
<td>5</td>
<td>5 clear</td>
<td>0.41-0.62</td>
</tr>
<tr>
<td></td>
<td>48</td>
<td>5</td>
<td>5 clear</td>
<td>0.45-0.62</td>
</tr>
<tr>
<td>9 days' moist chamber:</td>
<td>0</td>
<td>4</td>
<td>2 clear/1 cloudy</td>
<td>0.42-0.49</td>
</tr>
<tr>
<td></td>
<td>24</td>
<td>5</td>
<td>5 clear</td>
<td>0.40-0.59</td>
</tr>
<tr>
<td></td>
<td>48</td>
<td>5</td>
<td>4 clear/1 cloudy</td>
<td>0.46-0.68</td>
</tr>
<tr>
<td>21 days' moist chamber:</td>
<td>0</td>
<td>2</td>
<td>2 cloudy</td>
<td>0.46-0.72</td>
</tr>
<tr>
<td></td>
<td>24</td>
<td>3</td>
<td>1 clear/2 cloudy</td>
<td>0.48-0.66</td>
</tr>
<tr>
<td></td>
<td>48</td>
<td>3</td>
<td>2 clear/1 cloudy</td>
<td>0.52-0.58</td>
</tr>
</tbody>
</table>

*Recorded on first postoperative day
†Performed on fourteenth postoperative day.

Endothelial cells stored 21 days were disrupted (Fig. 4, B and C).

In vivo studies

Clinical evaluation. Corneas stored for 21 days were very edematous whether or not they had been placed in the M-K medium, but all corneas used for the 2-, 6-, and 9-day categories were relatively clear and were technically much easier to handle (Table II). With the use of the parameters of corneal thickness and clarity (in the absence of increased intraocular pressure), corneas stored 2 to 9 days appeared to function better than those stored for 21 days (Table II). Only one of 30 corneas stored in M-K medium after moist chamber storage for 2 to 9 days was cloudy and thick on the first postoperative day (Table II). Only three of eight corneas moist chamber–stored for 21 days and receiving M-K medium storage up to 48 hr appeared to be clear and thin on the first postoperative day (Table II).

NBT staining did not appear to be useful in evaluating transplantation success, since most of the donor corneas tended to have a minimum amount of staining (Table II).

Morphologic evaluation

TWO DAYS' MOIST CHAMBER STORAGE. There was no evidence of light microscopic changes, and endothelial cells were present in all specimens. SEM revealed an intact endothelial mosaic with prominent microvilli (Fig. 5, A to C). Endothelial cells were of normal size and distribution throughout the whole posterior corneal surface. TEM demonstrated some morphologic alterations at the junction between Descemet's membrane and endothelium, but the cells themselves appeared to have minor organelle alteration (Fig. 5, D and E). There was no difference in the quality of the cells stored in moist chamber and those stored up to an additional 48 hr in M-K medium (Table III).

SIX DAYS' MOIST CHAMBER STORAGE. Light microscopy demonstrated some endothelial swelling, although endothelial cells were present on all specimens. With SEM, endothelial...
cells appeared to be normal and to have an even
distribution throughout the posterior corneal
surface (Fig. 6, A to C). Cellular and extracellular
features in transmission micrographs were
similar to those seen in 2-day corneas (Fig. 6, D
to F). There was no morphologic difference
between corneas stored in moist chamber alone and those which were stored an additional period in M-K medium (Table III).

NINE DAYS' MOIST CHAMBER STORAGE. Intact
endothelial cells were identified by both light
microscopy and SEM (Fig. 7, A to C) in all
but one specimen. Many cells had an irregular
surface and an increased prominence of
microvilli, with obvious cell apposition lines.
TEM of control corneas revealed noticeable

Fig. 5. In vivo studies. Two days' storage in moist chamber. A, Scanning electron micrograph
of rabbit endothelium stored for 2 days in moist chamber. B, Scanning electron micrograph of
rabbit endothelium stored for an additional 24 hr in M-K medium. C, Scanning electron
micrograph of rabbit corneal endothelium stored for an additional 48 hr in M-K medium. The
normal endothelial mosaic is present in all specimens. D, Transmission electron micrograph of
rabbit corneal endothelium stored for 2 days in moist chamber. There is some irregularity to
Descemet's membrane. The endoplasmic reticulum is minimally dilated, and the mitochondria are edematous. E, Transmission electron micrograph of rabbit endothelium stored an additional 48 hr in M-K medium. There is minimal engorgement of the endoplasmic reticulum, and the mitochondria are edematous; otherwise, the cell appears normal.

cellular edema with mitochondrial swelling,
intracellular cyst formation, and decreased
cytoplasmic density (Fig. 7, D). Tight junc-
tions appeared intact, as did the posterior
plasma membrane. Corneas stored in M-K
medium following moist chamber storage
appeared to have less edema and better preser-
vation of cytoarchitecture (Fig. 7, E and F,
and Table III).

TWENTY-ONE DAYS' MOIST CHAMBER STORAGE.
All the corneas not receiving M-K medium
storage failed (Table II) and showed fibrin on
the posterior surface when examined mor-
phologically. There were few endothelial
cells on the posterior surface, and those that
remained were quite altered (Fig. 8, A).
Corneas receiving M-K medium treatment and that appeared to function clinically were covered posteriorly with enlarged and irregularly shaped endothelial cells (Fig. 8, B to E). Since the moist chamber-stored corneas failed, a comparison with additional M-K medium storage could not be made (Table III).

**Comparison of Donor Cells with Recipient Cells.** Comparison of the size and shape of donor cells to those of recipient cells at each storage period revealed no essential difference between corneas stored for 2, 6, and 9 days regardless of M-K medium treatment. However, corneas stored for 21 days and then kept in M-K medium appeared to have much larger endothelial cells. These were unusually shaped and comprised fewer cells per square millimeter (Fig. 8, C and E). A comparison of cell counts per square millimeter before and after keratoplasty was not performed, because of the wide variation between specimens.

**Discussion**

Evaluations of M-K medium–stored corneas have revealed that although there is no difference in the quality of tissue up to 48 hr post-mortem as compared with moist chamber storage, the medium appears to provide better quality tissue between 48 and 96 hr. It is clear from recent work that immediate immersion of donor tissue in the M-K medium provides the best quality tissue.

Studies using only rabbit corneas have clearly demonstrated the ability of M-K medium treatment to provide a functional corneal endothelium for up to 9 to 14 days of storage, but the reports are conflicting when M-K medium–stored corneas were compared with companion corneas stored in moist chambers.

Evaluation of cat corneas stored in M-K medium for 4 to 5 days have demonstrated that the preserved endothelium behaves much the same as human endothelium morphologically and functionally. More importantly, these studies have shown that structural preservation does not necessarily reflect the ability to resume normal function under more physiologic conditions. For example, although cat corneas stored in M-K medium for 5 days appeared to be morphologically
more altered than similarly stored moist chamber corneas, the M-K medium--stored corneas functioned better after keratoplasty.\textsuperscript{9}

In vitro evaluations of human corneas have shown that storage in M-K medium longer than 7 days produces morphologic changes that are probably irreversible, and the best evidence to date suggests these changes become most severe after 4 days of storage.\textsuperscript{11} Poor results with corneas stored longer than 96 hr have been reported.\textsuperscript{2,5}

Our results suggest that M-K medium can preserve the cytoarchitecture and function of the endothelium of corneas stored 9 days in moist chambers for an additional 2 days. These results are based upon the clinical evaluation of the corneas after keratoplasty, endothelial cell viability, and structural analysis of the endothelium. Placement of corneas stored in moist chamber for 21 days and then placement in M-K medium failed to

Table III. Transplant evaluation

<table>
<thead>
<tr>
<th>M-K storage (hr)</th>
<th>No. of eyes</th>
<th>Light microscopy*</th>
<th>SEM*</th>
<th>TEM*</th>
</tr>
</thead>
<tbody>
<tr>
<td>2 days' moist chamber:</td>
<td>0</td>
<td>5</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>24</td>
<td>4</td>
<td>5</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>48</td>
<td>5</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>6 days' moist chamber:</td>
<td>0</td>
<td>4</td>
<td>+1</td>
<td>+1</td>
</tr>
<tr>
<td>24</td>
<td>5</td>
<td>0</td>
<td>+1</td>
<td>+1</td>
</tr>
<tr>
<td>48</td>
<td>5</td>
<td>0</td>
<td>0</td>
<td>+1</td>
</tr>
<tr>
<td>9 days' moist chamber:</td>
<td>0</td>
<td>4</td>
<td>+1</td>
<td>+3</td>
</tr>
<tr>
<td>24</td>
<td>5</td>
<td>0</td>
<td>+1</td>
<td>+3</td>
</tr>
<tr>
<td>48</td>
<td>5</td>
<td>0</td>
<td>0</td>
<td>+1</td>
</tr>
</tbody>
</table>

* Morphologic changes, with 0 representing no change and +1 representing severe change.

These specimens had fewer cells per square millimeter than all other specimens.
Fig. 8. For legend see opposite page.
improve the quality of the tissue. The morphologic changes observed in the corneas stored for 9 days in moist chamber and 2 additional days in M-K medium were somewhat less severe than those changes reported to occur in corneas stored for up to 14 days in M-K medium alone.

Recently, the corneal specular microscope has been utilized to evaluate the quality of donor tissue before keratoplasty and the condition of the transplanted cornea after keratoplasty. These studies have demonstrated that when endothelial cells are lost postoperatively, the remaining cells spread and enlarge to replace lost cells and that a relatively few number of cells per square millimeter are still able to maintain a cornea of normal thickness and clarity. In our study, we attempted to compare the recipient and donor endothelium as far as number of cells per square millimeter, size, and shape, but a direct arithmetic comparison was impossible because of the wide variation in cell counts. Only corneas stored for 21 days had enlarged and distorted endothelial cells (Fig. 8).

Another way to evaluate the number and shape of the donor endothelial cells would be through the use of in vivo specular microscopy, but such a microscope was not available to us at the time of this study. Similarly, the use of temperature reversal to evaluate the function of the donor corneas would be useful, but once such a cornea has been temperature-reversed, it may not be suitable for transplantation.

Manipulation of the electrolyte and energy sources and other factors in the storage medium may provide a better medium for preservation. It is clear that the pH of stored corneas in the M-K medium may fluctuate widely, and it is possible that a change in the storage fluid every 48 hr could improve the quality of the donor tissue. However, this additional step requires additional cost and technical expertise, and all additional handling subjects corneas to the risks of mechanical trauma and infection.

Until an ideal medium becomes available, we believe that the following principles should be applied to the utilization of M-K medium–stored corneas:

1. Donor corneas should be stored in M-K medium at the first opportunity after death and should be used within 96 hr.
2. If the death-to-storage time is prolonged, then the maximum storage time in M-K medium should be decreased.
3. However, on the basis of the results of this study, there appears to be some leeway in maintaining the integrity and function of the corneal endothelium after moist chamber storage, by the use of M-K medium storage of excised corneas for up to 48 hr more. This may allow better surgical planning and better use of a limited number of donor corneas.

We thank Keith Deibert, Patti Hagiwara-Akers, and Harvey Sternberg for their capable assistance.

---

Fig. 8. In vivo study. A, Transmission electron micrograph of rabbit corneal endothelium stored for 21 days in moist chamber and then used as a donor for corneal transplantation. The posterior plasma membrane appears to be broken, the normal cytoarchitecture of the cell appears disrupted, and only the nucleus remains visible. B, Scanning electron micrograph of cornea stored an additional 24 hr in M-K medium. The donor cornea on the right appears to have an intact posterior surface. The wound is intact, and the recipient cornea on the left appears to have a smooth surface. The sites of the nylon sutures are easily visualized. C, Scanning electron micrograph of cornea stored an additional 24 hr in M-K medium. The entire surface is covered with endothelial cells, which are irregular in shape and size. D, Scanning electron micrograph of rabbit cornea stored an additional 48 hr in M-K medium. There are more folds in Descemet's membrane in the donor cornea on the right than in the recipient cornea on the left. The wound is intact. E, Scanning electron micrograph of cornea stored an additional 48 hr in M-K medium. There is a more normal pattern to the endothelial mosaic, although irregularly shaped and enlarged cells can easily be seen.
REFERENCES


Information for authors

Most of the provisions of the Copyright Act of 1976 became effective on January 1, 1978. Therefore all manuscripts must be accompanied by the following statement, signed by each author: “The undersigned author(s) transfers all copyright ownership of the manuscript entitled (title of article) to The Association for Research in Vision and Ophthalmology, Inc., in the event the work is published. The author(s) warrant that the article is original, is not under consideration by another journal, and has not been previously published.” Authors will be consulted, when possible, regarding republication of their material.