Estimation of Corneal Endothelial Pump Function in Long-Term Contact Lens Wearers

William M. Bourne,1 David O. Hodge,2 and Jay W. McLaren1

PURPOSE. To study the effects of long-term contact lens wear on morphologic and physiologic properties of corneal endothelial cells.

METHODS. The endothelial permeability to fluorescein and the rate of corneal deswelling from hypoxia-induced edema were measured in 20 long-term (mean, 17 ± 9 years; range, 5–33 years) contact lens wearers and 20 age-matched control subjects. From these data, the relative endothelial pump rate in each subject was estimated, based on the pump-leak hypothesis of corneal hydration control. Corneal autofluorescence and the aqueous humor flow rate were determined by fluorescein fluorophotometry. Images of corneal endothelial cells were recorded by using specular microscopy, and morphologic indices (cell density, coefficient of variation of cell area, percentage of hexagonal cells, and skewness) were determined.

RESULTS. No statistically significant differences were found between the contact lens and control groups in endothelial permeability, corneal deswelling, relative endothelial pump rate (mean ± SD 1.07 ± 0.33 relative pump units versus 1.01 ± 0.25 relative pump units; contact lens versus control; P = 0.57), and endothelial cell density. Contact lens wearers had a significantly higher aqueous humor flow rate (3.57 ± 1.03 μl/min versus 2.77 ± 0.51 μl/min; P = 0.005), coefficient of variation of cell area (0.35 ± 0.09 versus 0.28 ± 0.04; P = 0.006), and corneal autofluorescence (3.1 ± 0.6 ng/ml versus 2.3 ± 0.3 ng/ml fluorescein equivalents; P < 0.001) than did non-contact lens wearers.

CONCLUSIONS. Despite the known effects of long-term contact lens wear on corneal endothelial morphometry, no effect on endothelial function was found. (Invest Ophthalmol Vis Sci. 1999;40: 603–611)
Twenty age-matched (within 5 years) subjects who had never worn contact lenses. All 40 subjects were examined, including slit lamp examination, tonometry, and undilated fundus examination, and had none of the following conditions: any ocular disease, previous ocular surgery, topical ocular medications in the previous month, systemic medications that can affect corneal thickness (e.g., birth control pills, diuretic drugs), ptosis, keratoconjunctivitis sicca, superficial corneal vascularization extending more than 2 mm inside the limbus, or diabetes mellitus. All subjects were at least 18 years old. All contact lens wearers had always worn similar lens types in both eyes. Types of lenses worn and duration of wear are given in Tables 1 and 2. This study followed the tenets of the Declaration of Helsinki and was approved by our Institutional Review Board; all subjects provided written informed consent to participate.

### MATERIALS AND METHODS

#### Subjects

We recruited 20 subjects from our contact lens clinic who had used daily-wear contact lenses in both eyes for at least 5 years. Twenty age-matched (within 5 years) subjects who had never worn contact lenses were also recruited from Mayo Clinic patients and staff and their families, to serve as control subjects. All 40 subjects were examined, including slit lamp examination, tonometry, and undilated fundus examination, and had none of the following conditions: any ocular disease, previous ocular surgery, topical ocular medications in the previous month, systemic medications that can affect corneal thickness (e.g., birth control pills, diuretic drugs), ptosis, keratoconjunctivitis sicca, superficial corneal vascularization extending more than 2 mm inside the limbus, or diabetes mellitus. All subjects were at least 18 years old. All contact lens wearers had always worn similar lens types in both eyes. Types of lenses worn and duration of wear are given in Tables 1 and 2. This study followed the tenets of the Declaration of Helsinki and was approved by our Institutional Review Board; all subjects provided written informed consent to participate.

#### Measurements

Subjects were studied on 2 days, and identical procedures were used in all subjects. Contact lenses were not worn during either of the study days. On the afternoon of day 1, autofluorescence of the central cornea was measured at an excitation wavelength of 488 nm (emission between 515 nm and 600 nm) by using a two-dimensional scanning ocular fluorophotometer. Corneal fluorescence was measured three times, and the mean was accepted as the autofluorescence. Anterior chamber volume of each eye was then measured by using a photogrammetric technique and 5 to 10 photographs of the central corneal endothelium of each eye were recorded by a wide-field contact specular microscope.

On day 2, subjects instilled 2% fluorescein in both eyes beginning at 2 AM. The drops were instilled at 5-minute intervals, and the number of drops was determined by the subject’s age: 5 drops were instilled by subjects less than 26 years of age, 4 drops by those 26 to 35 years of age, and 3 drops by those more than 35 years of age. Subjects were instructed to return to sleep. All subjects reported to the clinical research area by 8 AM for baseline pachometry and fluorophotometry of each eye.

Corneal thickness was measured by using a modified Haag–Streit optical pachometer equipped with fixation lights and a potentiometer that registered measurements directly into a computer memory. The operator was not aware of the individual thickness measurements as they were recorded. The mean of 10 consecutive measurements was accepted as the corneal thickness. The process was repeated if the SD of the 10 measurements was 10 μm or more. The instrument was calibrated daily by measuring a set of contact lenses of known thickness. We noted that some examiners used slightly differ-

---

**TABLE 1. Contact Lens Wearers**

<table>
<thead>
<tr>
<th>Type of Contact Lens</th>
<th>Years of Wear</th>
</tr>
</thead>
<tbody>
<tr>
<td>Soft (n = 10)</td>
<td>10.4 ± 4.8 (5-18)</td>
</tr>
<tr>
<td>Polymethylmethacrylate (n = 3)</td>
<td>21.7 ± 5.8 (15-25)</td>
</tr>
<tr>
<td>Rigid gas permeable (n = 1)</td>
<td>6 (--)</td>
</tr>
<tr>
<td>Mixed (n = 6)*</td>
<td>27.3 ± 3.7 (23-35)</td>
</tr>
<tr>
<td>Total (n = 20)</td>
<td>17.0 ± 9.2 (5-35)</td>
</tr>
</tbody>
</table>

* Values are means ± SD, with range in parentheses.

**TABLE 2. Lenses Worn by Mixed Contact Lens Group**

<table>
<thead>
<tr>
<th>Subject</th>
<th>Age</th>
<th>Polymethylmethacrylate</th>
<th>Rigid Gas-Permeable Contact Lens</th>
<th>Extended-Wear Soft Contact Lens</th>
<th>Daily-Wear Soft Contact Lens</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>39</td>
<td>—</td>
<td>22</td>
<td>—</td>
<td>1</td>
<td>23</td>
</tr>
<tr>
<td>2</td>
<td>42</td>
<td>14</td>
<td>—</td>
<td>11</td>
<td>—</td>
<td>25</td>
</tr>
<tr>
<td>3</td>
<td>45</td>
<td>4</td>
<td>—</td>
<td>21</td>
<td>—</td>
<td>25</td>
</tr>
<tr>
<td>4</td>
<td>47</td>
<td>24</td>
<td>—</td>
<td>—</td>
<td>6</td>
<td>30</td>
</tr>
<tr>
<td>5</td>
<td>51</td>
<td>18</td>
<td>15</td>
<td>—</td>
<td>—</td>
<td>33</td>
</tr>
<tr>
<td>6</td>
<td>61</td>
<td>22</td>
<td>6</td>
<td>—</td>
<td>—</td>
<td>28</td>
</tr>
</tbody>
</table>
ent alignment end points when measuring contact lenses than they did when measuring corneas. To compensate, we derived a correction factor for each cornea based on its thickness, measured with the specular microscope during the afternoon of study day 1.

At approximately 8:20 AM each eye was fitted with a soft contact lens (38% water content; +20.00 D; 8.3-mm base curve) and patched. After 2 hours, at approximately 10:20 AM, the patches and contact lenses were removed, and corneal thickness and corneal and cameral fluorescence were measured in both eyes. Starting at approximately 11:30 AM, corneal thickness was measured in each eye and repeated every 10 minutes for 1 hour, then every 20 minutes for 1 hour, and then every 30 minutes until 5 PM. Corneal and anterior chamber fluorescence were measured at 11:30 AM and noon, and then hourly until 5:00 PM. After the final fluorescence and thickness measurements were completed, intraocular pressure was measured three times by applanation tonometry, and the mean was recorded. The study was then complete for that subject. Autofluorescence was subtracted from each fluorescence measurement.

**Analysis**

The methods for calculating the deswelling rate, endothelial permeability, and aqueous humor flow rate have been described in detail elsewhere and are briefly outlined below:

**Deswelling Rate**

We assumed that after removal of the contact lens the stromal swelling, the thickness in excess of the OESS thickness, deswells as a first-order process, as described by Polse et al.: \[ q(t) = B + S e^{-Dt} \] where \( q(t) \) is the corneal thickness at time \( t \), \( B \) is the OESS thickness, \( S \) is the induced swelling (the thickness in excess of \( B \) at \( t = 0 \)), \( D \) is the deswelling rate constant, and \( t \) is time since removal of the contact lens. Corneal thicknesses were fitted to Equation 1 by nonlinear regression (S-PLUS; Statistical Systems, Seattle, WA) to obtain estimates of \( B \), \( S \), and \( D \) in each subject. Corneal thicknesses measured during the first 50 minutes after removal of the contact lenses were excluded because of the potential effects of decreased pH on the deswelling rate during this period. The PRPH, a more clinically meaningful parameter used to describe the deswelling rate, was also calculated: \[ \text{PRPH} = 100(1 - e^{-600}) \] \[ \text{(2)} \]

**Endothelial Permeability**

Endothelial permeability to fluorescein was determined by using the method described by Jones and Maurice: \[ \text{Permeability} = \frac{q_{r_e}(C_e(t_1) - C_e(t_0))}{(r_e C_e)(t_1 - t_0)} \] where \( t_0 \) and \( t_1 \) are time at the beginning and end of the interval, \( q_r \) is the mean corneal thickness on the interval, \( r_e \) is the steady state distribution ratio for fluorescein between the cornea and the anterior chamber (assumed to be 1.6 in corneas at normal thickness), \( C_e(t) \) is the concentration of fluorescein in the cornea at time \( t \), and \( C_e \) and \( C_a \) are the mean concentrations of fluorescein in the cornea and anterior chamber on the interval. Corneal fluorescence was adjusted for changes in corneal thickness. Mean concentrations \( C_e \) and \( C_a \) were determined from the initial and final concentrations on the interval by assuming that \( C_e \) and \( C_a \) decreased as a single exponential decay. We determined the permeability to fluorescein during two intervals. The first permeability (the AM, or hypoxic, permeability) was calculated from fluorescein concentrations at 8:00 AM, just before contact lens insertion, and at 11:30 AM, approximately 60 minutes after contact lens removal. The second permeability (the PM, or normoxic, permeability) was the mean of permeabilities during five 1-hour intervals between noon and 5 PM. The PM permeability is similar to the permeability measured when the cornea is not swollen.

**Aqueous Humor Flow Rate**

The clearance of fluorescein was used to calculate the aqueous humor flow rate: \[ \text{Flow} = \frac{\Delta M}{C_e \times \Delta t} - 0.25 \mu l/min \] where \( \Delta M \) is the loss of mass of fluorescein in the combined cornea and anterior chamber during an interval \( \Delta t \), and \( C_e \) is the average concentration in the anterior chamber during the same interval, estimated from the initial and final fluorescence and assuming a single exponential decay. The constant, 0.25 \( \mu l/min \), was subtracted to account for diffusional loss of fluorescein. We calculated aqueous humor flow between 12 noon and 5 PM, during the same intervals used to calculate PM endothelial permeability.

**Relative Endothelial Pump Rate**

The endothelial solute pump rate cannot be measured directly in humans, but the ratio of pump rate to normal pump rate can be estimated if we assume that the deswelling rate is directly proportional to the pump rate and inversely proportional to the endothelial permeability to fluorescein. This simple model can be expressed in terms of the pump rate:

\[ \text{Pump rate} = K \times \text{Deswelling rate} \times \text{Permeability} \]

where \( K \) is a constant. The absolute value of the pump rate can only be determined if we know \( K \). However, if \( K \) is constant from subject to subject, we can determine the ratio of pump rate of one person's cornea to normal pump rate:

\[ \text{Relative endothelial pump rate} = \frac{\text{deswelling rate}_i}{\text{deswelling rate}_n} \times \frac{\text{permeability}_i}{\text{permeability}_n} \] where the subscript \( i \) indicates an individual, and the subscript \( n \) indicates the normal values, for which we used the mean for the control group.
The relative endothelial pump rate was determined for each subject. Deswelling rate, determined by using Equation 1, and PM, or normoxic, permeability, determined by using Equation 3 for the period between noon and 5 PM, were used in Equation 6.

**Endothelial Morphologic Analysis**

Apices of 100 central corneal endothelial cells were digitized from photographic negatives magnified 500 times. The mean, SD, coefficient of variation (SD/mean), skewness\(^3\) of cell area, and the percentage of cells with six sides were determined by use of a commercial algorithm (Bambi system; Bio-Optics, Arlington, MA). The endothelial cell density in cells per square millimeter was expressed as the reciprocal of the mean cell area.

**Statistical Analysis**

The mean of the values for the left and right eyes was considered to be one observation in each subject. This summary was used for each subject to avoid the problems associated with the analysis of correlated data. For example, the correlation in PRPH between left and right eyes in this study was 0.56 (\(P = 0.0002\)). We compared contact lens wearers with control subjects by using a two-tailed Student’s \(t\)-test for means if the data were distributed normally and by using a Wilcoxon rank sum test if they were not. We also tested correlations between PRPH, induced swelling, permeability, relative pump rate, and the morphologic indices by using a Pearson’s correlation (\(r_p\)) for normal data and Spearman’s rank correlation (\(r_s\)) for non-normal data. A two-tailed \(P \leq 0.05\) was considered statistically significant in all tests.

**RESULTS**

We found no statistically significant differences between the two groups in any functional measurement except for an increase in aqueous humor flow rate in the contact lens wearers.

---

**TABLE 3. Results**

<table>
<thead>
<tr>
<th>Measurement</th>
<th>20 Control Subjects*</th>
<th>20 Contact Lens Wearers*</th>
<th>(P)‡</th>
<th>MDD(\dagger)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (years)</td>
<td>38 ± 14</td>
<td>37 ± 13</td>
<td>0.72</td>
<td>14</td>
</tr>
<tr>
<td>PRPH (%/h)</td>
<td>65.7 ± 12.3</td>
<td>72.5 ± 9.7</td>
<td>0.06</td>
<td>11.3</td>
</tr>
<tr>
<td>Open-eye steady state thickness ((\mu m))</td>
<td>552 ± 53</td>
<td>557 ± 37</td>
<td>0.76</td>
<td>47</td>
</tr>
<tr>
<td>8:00 AM thickness ((\mu m))</td>
<td>556 ± 55</td>
<td>554 ± 39</td>
<td>0.89</td>
<td>49</td>
</tr>
<tr>
<td>Induced swelling ((\mu m))§</td>
<td>11.1 ± 2.1</td>
<td>10.8 ± 1.7</td>
<td>0.68</td>
<td>2.0</td>
</tr>
<tr>
<td>Endothelial permeability, AM ((10^{-4}) cm/min)</td>
<td></td>
<td></td>
<td>3.95 ± 0.74</td>
<td>3.39 ± 1.05¶</td>
</tr>
<tr>
<td>Endothelial permeability, PM ((10^{-4}) cm/min)</td>
<td></td>
<td></td>
<td>4.04 ± 0.51</td>
<td>3.87 ± 1.08</td>
</tr>
<tr>
<td>Relative endothelial pump rate</td>
<td></td>
<td></td>
<td>1.01 ± 0.25</td>
<td>1.07 ± 0.33</td>
</tr>
<tr>
<td>Aqueous humor flow rate ((\mu l/min))</td>
<td>2.77 ± 0.51</td>
<td>3.57 ± 1.03</td>
<td>0.005¶</td>
<td>—</td>
</tr>
<tr>
<td>Intraocular pressure (mm Hg)</td>
<td>15.5 ± 2.8</td>
<td>15.6 ± 2.4</td>
<td>0.95</td>
<td>2.6</td>
</tr>
<tr>
<td>Corneal autofluorescence (ng/ml fluorescein equivalents)</td>
<td>2.3 ± 0.3</td>
<td>3.1 ± 0.6</td>
<td>&lt;0.001</td>
<td>—</td>
</tr>
<tr>
<td>Endothelial cell density (cells/mm(^2))</td>
<td>2736 ± 373</td>
<td>2688 ± 492</td>
<td>0.92¶</td>
<td>447</td>
</tr>
<tr>
<td>Coefficient of variation of cell area</td>
<td>0.28 ± 0.04</td>
<td>0.35 ± 0.09</td>
<td>0.006</td>
<td>—</td>
</tr>
<tr>
<td>Skewness of cell area</td>
<td>0.70 ± 0.53</td>
<td>0.77 ± 0.44</td>
<td>0.51¶</td>
<td>0.50</td>
</tr>
<tr>
<td>Hexagonal endothelial cells (%)</td>
<td>62.4 ± 7.0</td>
<td>55.8 ± 14.1</td>
<td>0.07</td>
<td>11.4</td>
</tr>
</tbody>
</table>

* Values are means ± SD, with median in parentheses when data are not distributed normally.
† Two-tailed Student’s \(t\)-test for means (except # below).
‡ MDD, minimum detectable difference with 90% power (\(a = 0.05, \beta = 0.10\)).
§ Increase in thickness from 8:00 AM to 10:30 AM, from just before contact lens insertion to just after removal. \(n = 19\) in control group (10:30 AM thickness was not recorded in one subject).
|| \(n = 19\) in control group (no fluorophotometry on study day 2 for one subject).
¶ Significantly less than the evening permeability in the same subjects (\(P = 0.02\); paired \(t\)-test).
# Wilcoxon rank sum test.

**FIGURE 1. Relative corneal endothelial pump rate (in relative pump units) in long-term contact lens wearers and control subjects.** Horizontal lines indicate mean values. There was no significant difference between the groups (\(P = 0.57\)).
FIGURE 2. Aqueous humor flow rate in long-term soft contact lens wearers, nonsoft lens wearers (polymethylmethacrylate, rigid gas-permeable, and mixed lens wear), and control subjects. Horizontal lines indicate median values. The flow rate was significantly higher in the soft lens wearers than in the control subjects ($P < 0.001$) and nonsoft lens wearers ($P = 0.04$). The differences remained statistically significant if the high outlier value in the soft lens group was not included in the analyses.

(From Table 3). The deswelling rate, OESS thickness, induced swelling, endothelial permeability, and relative endothelial pump rate (Fig. 1) were all similar in the two groups. The AM permeability was significantly less than the PM permeability in the contact lens wearers, but not in the control subjects. Among morphologic measurements, the coefficient of variation was increased in the contact lens group. Corneal autofluorescence was also increased in the contact lens wearers.

TABLE 4. Results in Wearers of Soft Contact Lenses versus Wearers of Nonsoft Lenses

<table>
<thead>
<tr>
<th>Measurement</th>
<th>10 Soft Lens Wearers*</th>
<th>10 Nonsoft Lens Wearers†</th>
<th>$P$‡</th>
<th>MDD§</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (y)</td>
<td>29 ± 8</td>
<td>44 ± 13</td>
<td>0.006</td>
<td>—</td>
</tr>
<tr>
<td>Years of contact lens wear</td>
<td>10 ± 5</td>
<td>24 ± 8</td>
<td>0.002</td>
<td>—</td>
</tr>
<tr>
<td>PRPH (%/h)</td>
<td>76.6 ± 8.4</td>
<td>68.4 ± 9.5</td>
<td>0.06</td>
<td>13.0</td>
</tr>
<tr>
<td>Open-eye steady state thickness (µm)</td>
<td>559 ± 41</td>
<td>555 ± 32</td>
<td>0.79</td>
<td>55</td>
</tr>
<tr>
<td>8:00 AM thickness (µm)</td>
<td>558 ± 41</td>
<td>550 ± 38</td>
<td>0.68</td>
<td>58</td>
</tr>
<tr>
<td>Induced swelling (µm)</td>
<td>60 ± 4</td>
<td>59 ± 10</td>
<td>0.60</td>
<td>11</td>
</tr>
<tr>
<td>Induced swelling (%)</td>
<td>10.9 ± 1.4</td>
<td>10.7 ± 2.0</td>
<td>0.80</td>
<td>2.5</td>
</tr>
<tr>
<td>Endothelial permeability, AM ($\times 10^{-4}$ cm/min)</td>
<td>4.10 ± 0.90</td>
<td>2.68 ± 0.65§</td>
<td>&lt;0.001</td>
<td>—</td>
</tr>
<tr>
<td>Endothelial permeability, PM ($\times 10^{-4}$ cm/min)</td>
<td>4.42 ± 0.98</td>
<td>3.33 ± 0.91</td>
<td>0.02</td>
<td>—</td>
</tr>
<tr>
<td>Relative endothelial pump rate (relative units)</td>
<td>1.26 ± 0.27</td>
<td>0.87 ± 0.27</td>
<td>0.004</td>
<td>—</td>
</tr>
<tr>
<td>Aqueous humor flow rate (µL/min)</td>
<td>4.04 ± 1.09</td>
<td>3.09 ± 0.74</td>
<td>0.04*</td>
<td>—</td>
</tr>
<tr>
<td>Intraocular pressure (mm Hg)</td>
<td>14.6 ± 2.5</td>
<td>16.5 ± 2.0</td>
<td>0.07</td>
<td>3.3</td>
</tr>
<tr>
<td>Corneal autofluorescence (ng/ml fluorescein equivalents)</td>
<td>3.0 ± 0.4</td>
<td>3.2 ± 0.8</td>
<td>0.32</td>
<td>0.90</td>
</tr>
<tr>
<td>Endothelial cell density (cells/mm²)</td>
<td>2941 ± 271</td>
<td>2435 ± 543</td>
<td>0.04*</td>
<td>—</td>
</tr>
<tr>
<td>(2966)</td>
<td>(2710)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Coefficient of variation of cell area</td>
<td>0.34 ± 0.10</td>
<td>0.36 ± 0.09</td>
<td>0.59</td>
<td>0.14</td>
</tr>
<tr>
<td>Skewness of cell area</td>
<td>0.74 ± 0.48</td>
<td>0.81 ± 0.41</td>
<td>0.63§</td>
<td>0.65</td>
</tr>
<tr>
<td>(0.50)</td>
<td>(0.71)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hexagonal endothelial cells (%)</td>
<td>60.7 ± 14.7</td>
<td>50.9 ± 12.3</td>
<td>0.12</td>
<td>19.7</td>
</tr>
</tbody>
</table>

*Values are means ± SD, with median in parentheses when data are not distributed normally.
† Nonsoft: polymethylmethacrylate, rigid gas-permeable, or mixed lens wear (see Table 1).
‡ Two-tailed Student's $t$-test for means (except * below).
§ MDD, minimum detectable difference with 90% power ($\alpha = 0.05$, $\beta = 0.10$).
¶ Increase in thickness from 8:00 to 10:30 AM, from just before contact lens insertion to just after removal.
# Significantly less than the evening permeability in the same subjects ($P = 0.04$; paired $t$-test).
* Wilcoxon rank sum test.
variation of cell area ($r_p = -0.45$; $P = 0.05$), and age ($r_p = -0.50$; $P = 0.03$; Fig. 3). Relative endothelial pump rate was correlated with years of contact lens wear ($r_p = -0.52$; $P = 0.02$) and age ($r_p = -0.49$; $P = 0.03$; Fig. 4). Years of contact lens wear was also associated with age ($r_p = 0.85$; $P < 0.001$) and percentage of hexagonal cells ($r_p = -0.64$; $P = 0.002$), but not with coefficient of variation ($r_p = 0.36$; $P = 0.12$).

Although age and years of contact lens wear were significantly related to PRPH and to relative endothelial pump rate, age and years of wear contained similar information ($r_p = 0.85$; $P < 0.001$). After adjusting for age, therefore, the years of contact lens wear were not significantly related to PRPH or to relative endothelial pump rate (multiple regression analysis). Statistically significant correlations were not found in the control group.

**Discussion**

In this study, as in other studies, long-term contact lens wearers had a higher than normal coefficient of variation of endothelial area, but normal cell density, compared with subjects who had never worn contact lenses. In contrast to these changes, there were no significant functional differences in their corneal endothelia; neither recovery from swelling (PRPH), nor permeability to fluorescein, nor relative endothelial pump rate in this group was significantly different from normal subjects. It is not clear why these physiologic properties did not change. Perhaps the increase in paracellular area resulting from the greater variability of cell size and shape was not great enough to change the pump and barrier. Alternatively, the density of active pump sites may have decreased with age and the years of wear.

Recovery from swelling induced by contact lenses has also been measured in long-term contact lens wearers by others. Nieuwendaal et al. found that PRPH was significantly less in contact lens wearers than in control subjects, whereas we found that it was more, although not significantly so ($P = 0.06$). It is not clear why results from our experiment and that of Nieuwendaal et al. differ, although in our protocol, thickness during the first hour of deswelling was not used to calculate PRPH, whereas Nieuwendaal used thickness every 15 minutes after the lens was removed. We purposely did not include fluorescein or corneal thickness during this period because of the potential changes in fluorescein and deswelling rate induced by the lower stromal pH. As a test of whether this difference in methods could account for the differences in the results of these two studies, we reanalyzed our data and included the thickness measured immediately after removal of the contact lens. By this method, PRPH values for contact lens wearers and control subjects were $58.0\% \pm 7.3\%$ and $47.7\% \pm 11.6\%$, respectively. These values are lower than those shown in Table 3, presumably because of the effect of decreased pH during the first 50 minutes after contact lens removal. However, PRPH in the contact lens wearers was still not significantly lower than PRPH in the control subjects when calculated in this way. The decreased PRPH found by Nieuwendaal et al. must have resulted from either an increased endothelial permeability or a decreased pump rate. Nieuwendaal et al. did not measure endothelial permeability, but it has not been abnormal in other studies of long-term contact lens wearers.

Another possible explanation for the differences between the results of our study and that of Nieuwendaal et al. is that more of their subjects wore polymethylmethacrylate lenses (17/21) than in our study (8/20) and therefore may have been exposed to more severe hypoxia. Also, the average wearing time of polymethylmethacrylate or soft contact lenses was longer in their study (19.5 years) than in ours (14.2 years in 19 subjects). Therefore, we tested separately the 9 subjects in the polymethylmethacrylate- and mixed-lens-wear groups (Table 1) and the 12 subjects who had worn contact lenses for more than 12 years (mean wearing time, 23.3 years). Neither group differed significantly from their age-matched control subjects in PRPH or relative endothelial pump rate. These findings are explained by the data in Figures 3 and 4, in which the contact lens and control groups are seen to overlap considerably.

Others have found a delay in the return to normal thickness after cataract extraction in eyes with preoperative polymegathism and pleomorphism that were present in our sample of contact lens wearers apparently do not limit the pump function. The slower recovery of corneal thickness after cataract extraction may have been the result of slower recovery of the polymegathous endothelium after surgical trauma or inflammation.

The OESS thickness was similar between the two groups, as was the 8 AM thickness. These findings confirm those of Nieuwendaal et al. Holden et al. found a decrease in stromal thickness of $11 \mu m$ (2.3%) in the lens-wearing eye of unilateral contact lens wearers, but it was manifest only after discontinuing lens wear for 7 days. When the lenses were initially removed, the stromal thickness was increased 2.5% in the lens-wearing eyes. Our subjects had discontinued their lenses only overnight, and residual edema may therefore have been present.

The contact lens and control groups had similar swelling induced by two hours of hypoxic contact lens wear. Nieuwendaal et al. found less induced swelling in their contact lens wearers. This difference in findings, similar to the difference in PRPH, remains unexplained. Erickson et al. found a strong correlation between the percentage of induced swelling...
and endothelial cell density in 15 adapted lens wearers. We found no correlation between these two parameters in the 20 contact lens wearers in the present study (r = 0.05; P = 0.83).

In this study we assumed that all fluorescein and fluid was transferred across the endothelium and that losses to the epithelial surface were negligible. The epithelial contribution to corneal hydration control is probably small in normal eyes. It is possible that changes in the epithelium affected our measurement of endothelial permeability in contact lens wearers, because their epithelia are more fragile. Because epithelial permeability is not elevated in contact lens wearers, because their epithelia are more fragile.36 Because epithelial permeability is not elevated in contact lens wearers, our estimates of endothelial permeability should be valid.

The AM permeability was significantly less than the PM permeability in the contact lens wearers, but not in the control subjects. The AM permeability in the contact lens wearers was also less than that in the control subjects, but the difference did not reach statistical significance (P = 0.07). The AM permeability was calculated for the period when the eye was wearing an aphakic soft contact lens with the lids closed, and therefore the cornea was hypoxic and acidic.38 The AM, or hypoxic, permeability is also decreased in diabetes mellitus.14,15 The mechanism of this abnormal response to acidosis in contact lens wearers and people with diabetes is unknown; swelling of endothelial cells with narrowing of the paracellular pathway has been suggested.14,15 Contact lens wear and diabetes induce endothelial cell polymegethism, so that this morphologic change may be associated with the decrease in hypoxic permeability. The AM permeability was not decreased, however, in seven phakic eyes with corneal transplants that also had polymegethism, although the endothelial cells were much larger.39

There was no significant difference in PM permeability between the contact lens wearers and the control subjects. These results exclude, with 90% confidence, a difference of 0.88 x 10^-4 cm/min (Table 2). The PM, or normoxic, permeability was measured during the period when the cornea was exposed to normal oxygen concentrations and was similar to that measured during the unworn state.26 These findings confirm those of earlier studies, in which no effect was found of long-term contact lens wear on endothelial permeability to fluorescein.10,11

We calculated a relative endothelial pump rate by using normalized values and assuming that the deswelling rate was directly proportional to the pump rate and inversely proportional to the permeability. These assumptions arise from the pump-leak hypothesis of corneal hydration control:1 In steady state, the rate of active solute (and passive fluid) transfer from the stroma to the aqueous humor by the endothelial pump is balanced by the passive leak of solute and fluid across the endothelium into the stroma. This analysis ignores the movement of solutes and fluid across the epithelium and limbus, which is thought to be of minor importance. The endothelial permeability to the small molecule, fluorescein, is assumed to be proportional to the leak. It follows that when the swollen cornea deswells, the rate of deswelling is slower in the presence of a decrease in the pump rate or an increase in permeability.

Two fluorophotometric parameters were elevated in contact lens wearers, corneal autofluorescence and aqueous humor flow rate. Similar increases in corneal autofluorescence have been found in patients with diabetes mellitus13–15,40 and in those who have had corneal transplants.39,40 Autofluorescence is thought to arise from mitochondrial flavoproteins,44 and its major portion most likely originates in the epithelium.41 Elevated autofluorescence suggests increased native fluorescence or a change in local conditions that affect its fluorescence, such as an alteration in oxidative state, pH, temperature, light-filtering characteristics of the overlying stroma, or interactions with other molecules.

The increased aqueous humor flow confirms our earlier results, although a smaller study of 11 extended-wear contact lens wearers did not show an elevated flow rate in contact lens wearers.37 At least three factors that may be altered in contact lens wearers could elevate aqueous humor flow. First, larger eyes may require a higher flow rate for adequate function. This possibility is consistent with the absence of difference in intraocular pressure in the two groups, despite the increased aqueous humor flow (Table 2). Our control subjects were not matched for refractive error, and the contact lens wearers were more myopic (mean spherical equivalent correction, −4.6 ± 2.3 D; n = 19) than the control subjects (mean spherical equivalent, +0.1 ± 1.0 D; n = 12; P < 0.0001). The contact lens group also had larger anterior chamber volumes (215 ± 37 μl) than the control group (178 ± 37 μl; P = 0.004). After adjusting for anterior chamber volume, however, there was still a significant difference between the groups in aqueous humor flow (P = 0.02, analysis of covariance). Therefore, the difference in anterior chamber volume (larger eyes) does not seem to explain the difference in aqueous humor flow. In addition, there was no significant correlation between anterior chamber volume and flow in either group.

Second, an elevated epithelial permeability in contact lens wearers could raise the clearance of fluorescein across the anterior surface of the cornea and increase the apparent flow rate. As mentioned earlier, however, epithelial permeability is not elevated in soft or rigid gas-permeable contact lens wearers.37 McNamara et al.42 recently measured a 40% elevation in epithelial permeability after 1 hour of contact lens wear intended to produce hypoxia.42 When we modeled mathematically the transfer of fluorescein across the epithelium for a past presentation,43 at least a 10-fold increase in epithelial permeability was necessary to increase the estimate of flow by 1%. Moreover, if the epithelial permeability were increased, more fluorescein should have entered the corneal stroma when fluorescein was instilled. The stromal concentration at 8 AM, however, was not higher in contact lens wearers than it was in control subjects (1072 ± 836 ng/ml versus 1035 ± 706 ng/ml; P = 0.88). For these reasons, even if epithelial permeability was increased in the contact lens wearers, it should not have produced the differences in flow that we measured.

Finally, the ciliary body may have been stimulated to produce aqueous humor at a higher rate by the decrease in pH44 or oxygen45 in the aqueous humor when contact lenses are worn. Unfortunately, the responses of the ciliary body to pH and oxygen levels in the aqueous humor are not known. Although there was prominent polymegethism (increased coefficient of variation of cell area) in the contact lens group, there was no significant difference in the skewness of cell area. This result suggests that the endothelia of contact lens wearers have higher proportions of both large and small cells.46

The 10 subjects who wore only soft contact lenses were younger than the remaining 10 subjects, and this difference may explain their greater endothelial cell densities and higher pump rates (Table 3). The soft lens wearers also had signifi-
cantly higher aqueous humor flow (Fig. 2) and permeability to fluorescein than did the remaining subjects. Thus, the 10 soft lens wearers accounted for the increased aqueous humor flow rate in the contact lens group compared with that in the control subjects (Table 2). Moreover, the 10 subjects who had worn lenses other than soft lenses accounted for the decreased AM permeability in the contact lens group compared with that in the control subjects (Table 2). We have no reasonable explanation for these findings. In our previous study, in which aqueous humor flow rate was higher in contact lens wearers than in control subjects, the increased flow was also present only in the soft lens wearers. Soft lenses have larger diameters, but we have no plausible hypothesis for why this difference would affect aqueous humor flow rate.

In the contact lens group, we found significant negative correlations between years of contact lens wear and both corneal deswelling rate (PRPH) and relative endothelial pump rate. We could not attribute this negative correlation to an increasingly depressive effect of prolonged contact lens wear on the deswelling and pump rates, however, because of a similar correlation between these rates and age and a stronger correlation between age and years of wear. Therefore, age could not be ruled out as the cause of the decrease in deswelling and pump rates that occurred with increasing years of contact lens wear. The same can be said of the significant negative correlation between PRPH and coefficient of variation of cell area, which also was more strongly correlated with age in the contact lens group. Although we found no significant correlations in the control group between age and PRPH, endothelial permeability, endothelial cell density, coefficient of variation, or percentage of hexagonal cells, such correlations have been found in other studies with more subjects and larger ranges of age.  

In summary, despite the known effects of long-term contact lens wear on corneal endothelial cell morphology, we could show no effects on endothelial cell function.

References

ANNOUNCEMENT

GRF Names Scientific Advisory Committee Chair
Paul Kaufman, M.D., to Lead Research Funding into 21st Century

The Glaucoma Research Foundation (GRF) has named Paul Kaufman, M.D., Director of Glaucoma Services at University of Wisconsin, Madison, as chairman of its Scientific Advisory Committee.

Kaufman will lead the committee in determining funding for promising research projects to protect sight from glaucoma. Last year, GRF's Scientific Advisory Committee approved funding of $1.5 million.

"I believe fervently in GRF," Kaufman said. "GRF programs develop the science and the scientists of the future, at a stage usually too preliminary to light up radar screens of larger organizations."

"At the same time, GRF resources are now substantial enough to make a real impact."

GRF is a national not-for-profit organization dedicated to protecting the sight and independence of people with glaucoma. The ultimate goal is a cure.