Corneal Crosslinking: Keratoconus and Beyond

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Acknowledgements

• The presenter does not have a financial interest in the subject matter of this presentation
• Keratoconus: non-inflammatory progressive ectatic disorder
  – Glasses
  – Contact lenses
  – Transplant surgery
Induction of Cross-links in Corneal Tissue

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(Received Lund 5 May 1997 and accepted in revised form 15 September 1997)

The aim of this study was to investigate the possibility of induction of cross-links in corneal tissue in order to increase the stiffness as a basis for a future conservative treatment of keratoconus. Collagenous biomaterials can be stabilized by chemical and physical agents. The epithelium of excised porcine eyes was removed. Eight test groups, 10 eyes each, were treated with UV-light ($\lambda = 254$ nm), 0.5% riboflavin, 0.5% riboflavin and UV-light (365 nm) blue light (436 nm) and sunlight, and the chemical agents glutaraldehyde (1% and 0.1%, 10 min) and Karnovsky’s solution (0.1%, 10 min). Strips of 5 mm in width and 9 mm in length were cut from each cornea and the stress-strain behaviour of the strips was measured to assess the cross-linking process. For comparison, ten untreated corneas were measured by the same method. Compared to untreated corneas treatment with riboflavin and UV-irradiation as well as weak glutaraldehyde or Karnovsky’s solutions resulted in an increased stiffness of the corneas. The biomechanical behaviour of the corneas can be altered by glutaraldehyde, Karnovsky’s solution, and with riboflavin and UV-irradiation which offers the potential of a conservative treatment of keratoconus. To optimize this effect further investigation is necessary regarding the dose-response and in-vivo application.

Key words: biomechanics; biomaterials; collagen; cornea; cross-linking; keratoconus; proteoglycans.

Cross-linking

- Standard approach in polymer chemistry to increase elastic modulus
- Had entered use to increase stability and reduce biodegradation of porcine heart valves etc.
- Considered for stabilizing synthetic collagens for epikeratoplasty
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Corneal Cross-linking: Mechanism of Action

- Combines the use of ultra-violet (UV) light and riboflavin (vitamin B2) drops
- The absorption of UVA by riboflavin generates radical riboflavin and singlet oxygen to form cross-links
- Cross-linking:
  - Creates new corneal collagen cross-links
  - Results in a shortening and thickening of the collagen fibrils
  - Leads to the stiffening of the cornea

Where do cross-links occur?

- Collagen fibrils within lamellae are regulated by an interconnecting network of proteoglycans.¹,²
- Cross-linking with UVA/riboflavin has no obvious effect on any collagen structural parameter measured by x-ray scattering except uniformity of nearest neighbor interfibrillar spacing.³
- Suggests that cross-links are formed predominantly at fibril surfaces and within the protein network surrounding the collagen.³


Riboflavin/Ultraviolet-A–induced Collagen Crosslinking for the Treatment of Keratoconus

GREGOR WOLLENSAK, MD, EBERHARD SPOERI, PhD, AND THEO SEILER, PhD, MD

• Pilot study of 23 eyes of 22 patients
  – Moderate to advanced keratoconus (Kmax 48-72 D)
  – 6 month interval follow-up, 3 months to 4 years
  – No progression noted
  – In 16 eyes, mean reduction in Kmax of 2D, mean reduction in SE of 1D
Avedro FDA Study

- Study eye randomized into one of two groups
  - CXL treatment
  - Sham Control
- At Month 3 or later:
  - Non-randomized fellow eyes could receive CXL treatment
  - Control eye could receive CXL treatment
- The primary efficacy parameter evaluated over time was corneal curvature, as measured by maximum keratometry ($K_{max}$)
- Study success was defined as a difference of at least 1 diopter in the mean change in $K_{max}$ from baseline comparing the CXL treatment group and the control group at 12 months

### Progressive Keratoconus

#### Demographics

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Statistic</th>
<th>CXL Group</th>
<th>Control Group</th>
<th>Total</th>
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</thead>
<tbody>
<tr>
<td>Received Randomized Treatment</td>
<td>N</td>
<td>102</td>
<td>103</td>
<td>205</td>
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<tr>
<td>Completed</td>
<td>n (%)</td>
<td>92 (90.2)</td>
<td>85 (82.5)</td>
<td>177 (86.3)</td>
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<tr>
<td>Age (yrs)</td>
<td>Mean</td>
<td>31.1</td>
<td>35.0</td>
<td>33.0</td>
</tr>
<tr>
<td>Gender</td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>Female - n (%)</td>
<td></td>
<td>27 (26.5)</td>
<td>35 (34.0)</td>
<td>62 (30.2%)</td>
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<tr>
<td>Male - n (%)</td>
<td></td>
<td>75 (73.5)</td>
<td>68 (66.0)</td>
<td>143 (69.8%)</td>
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<tr>
<td>Kmax</td>
<td>Mean (SD)</td>
<td>60.9 D (+/- 9.14)</td>
<td>60.4 D (+/- 8.94)</td>
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</tbody>
</table>
### Corneal Ectasia

#### Demographics

<table>
<thead>
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<th>Parameter</th>
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<th>Pooled Studies</th>
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<tr>
<td></td>
<td></td>
<td>CXL Group</td>
<td>Control Group</td>
<td>Total</td>
</tr>
<tr>
<td>Received Randomized Treatment</td>
<td>N</td>
<td>91</td>
<td>88</td>
<td>179</td>
</tr>
<tr>
<td>Completed</td>
<td>n (%)</td>
<td>78 (85.7)</td>
<td>72 (81.8)</td>
<td>150 (83.8)</td>
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<tr>
<td>Age (yrs)</td>
<td>Mean</td>
<td>43.5</td>
<td>41.8</td>
<td>42.7</td>
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<tr>
<td>Gender</td>
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<td></td>
<td></td>
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</tr>
<tr>
<td>Female - n (%)</td>
<td>33 (36.3)</td>
<td>24 (27.3)</td>
<td>57 (31.8)</td>
<td></td>
</tr>
<tr>
<td>Male - n (%)</td>
<td>58 (63.7)</td>
<td>64 (72.7)</td>
<td>122 (68.2)</td>
<td></td>
</tr>
<tr>
<td>Kmax</td>
<td>Mean (SD)</td>
<td>55.4 D (+/- 6.86)</td>
<td>54.8 D (+/- 6.40)</td>
<td></td>
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</tbody>
</table>

#### Mean Change $K_{\text{max}}$ (D) – Month 12

<table>
<thead>
<tr>
<th>Condition</th>
<th>n</th>
<th>Mean Difference $K_{\text{max}}$ Change</th>
<th>p-value</th>
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<tbody>
<tr>
<td>Progressive Keratoconus</td>
<td>103</td>
<td>2.6 D</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Corneal Ectasia</td>
<td>88</td>
<td>1.4 D</td>
<td>&lt;0.0001</td>
</tr>
</tbody>
</table>

Mean Difference $K_{\text{max}}$ Change = 2.6 D<br>Mean Difference $K_{\text{max}}$ Change = 1.4 D

*Includes LOCF
After topical anesthesia, debride the epithelium to a diameter of approximately 9 mm using standard aseptic technique.

Post epithelial debridement, instill 1 drop of viscous riboflavin topically on the eye every 2 minutes for 30 minutes.

At the end of the 30 minute soaking period, examine the eye under the slit lamp for the presence of a yellow flare in the anterior chamber.

If the yellow flare is not detected, instill 1 drop of Photrex Viscous every 2 minutes for an additional 2 to 3 drops and recheck for the presence of a yellow flare. This process can be repeated as necessary.

Once the yellow flare is observed, perform ultrasound pachymetry. If corneal thickness is less than 400 microns, instill 2 drops of hypotonic riboflavin every 5 to 10 seconds until the corneal thickness increases to at least 400 microns.

Irradiation should not be performed unless this 400 micron threshold is met and the yellow flare is seen.
Irradiate the eye for 30 continuous minutes at 3mW/cm² at a wavelength of 365 nm, centered over the cornea.

During irradiation, continue topical instillation of viscous riboflavin onto the eye every 2 minutes for the 30 minute irradiation period.

Long term results

- Limited
- “long-term” study from Germany
- 480 eyes of 272 patients treated
- Only 241 eyes available, follow up ranging from 12 months to 5 years
- Progressive reduction in Kmax seen up to 3 years, stable at 6

Frederik Raiskup-Wolf, Anne Hoyer, Eberhard Spoerl, Lutz E. Pillunat

Collagen crosslinking with riboflavin and ultraviolet-A light in keratoconus: Long-term results

Figure 2. A: Change in maximum keratometry.

Frederik Raiskup-Wolf, Anne Hoyer, Eberhard Spoerl, Lutz E. Pillunat
Collagen crosslinking with riboflavin and ultraviolet-A light in keratoconus: Long-term results

Figure 2. B: Change in astigmatism in the central 3.0 mm zone over the 6-year follow-up (mean ± standard deviation) (Kmax = maximum keratometry).

Frederik Raiskup-Wolf, Anne Hoyer, Eberhard Spoerl, Lutz E. Pillunat
Collagen crosslinking with riboflavin and ultraviolet-A light in keratoconus: Long-term results
• Long term results

  - “10 year” study from Germany
  - 34 eyes of 24 patients
  - Followed for 11 years (+/-1.7SD)
  - Pre and post indices compared

Figure 1. Mean K values before and 10 years after CXL (apex = the apex of the keratoconus; max = maximum; min = minimum).

Frederik Raiskup, Anja Theuring, Lutz E. Pillunat, Eberhard Spoerl

Corneal collagen crosslinking with riboflavin and ultraviolet-A light in progressive keratoconus: Ten-year results

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Redistribution of Corneal Stress

- Finite element analysis modeling suggests the potential for application of cross-linking to produce shape change through focal stiffening of the cornea.¹
- Preferentially stiffening the weakest zone of the cornea leads to redistribution of corneal stress and shape change.

Photorefractive Intrastromal Cross-Linking (PiXL)

- PiXL is the application of zonal CXL using spatial application of UVA
- The goal of PiXL is to introduce controlled corneal shape change using patterned UVA CXL, without ablation
- Regularize keratoconic corneas
- Suggested for refractive corrections

Establish treatment transitions
- Eye tracking to compensate for patient eye movement

Complex patterning
- Digital Micromirror Device (DMD) to project specific UVA pattern

Spatial differentiation of energy intensity
- Pulsed illumination
- Variable irradiance and time
• Greatest energy dose applied over area of steepest anterior curvature, encompassing area of posterior elevation

• Pentacam mapping of cornea
• 2D steps relative to Kmax point to establish shape
  – 43-47D: 7.2 J/cm for 8 minutes
  – 48-52D: 10 J/cm for 11 minutes
  – >52D: 15J/cm for 17 minutes
Refactive improvements and safety with topography-guided corneal crosslinking for keratoconus: 1-year results
Maria Nordström, Maria Schiller, Anneli Fredriksson, Anders Behndig

- 37 patients, 50 eyes
- Randomized to PiXL vs CXL
- Assessed at 1,3,6,12 months
- Improvement in SE and VA in PiXL but not in CXL group (between group comparisons inconsistent)

PIXL treatment of Keratoconus: Case Example
Anders Behndig, MD
Umeå University Hospital, Sweden
Differential topography of the right and left eyes of the same individual at 12 months post treatment.

Anders Behndig, MD
Umeå University Hospital, Sweden
Specific UVA patterning is applied to the riboflavin-soaked cornea. Focal stiffening is achieved in the treatment zone, resulting in redistribution of biomechanical stresses. “Bulging” of the cornea in untreated regions results in central flattening: reduction of myopia.

Photo-Activated Chromophore for Keratitis – Corneal Cross Linking (PACK-CXL)

- Antiseptic property of riboflavin and ultraviolet light due to:
  - UV light microbial DNA and RNA damage
  - Riboflavin ROS release damaging microbial nucleic acids and cell membranes
- Corneal stromal stabilization
Photo-Activated Chromophore for Keratitis – Corneal Cross Linking (PACK-CXL)

- Bacterial keratitis
- Fungal keratitis
- Acanthamoeba keratitis

- Administered along with conventional treatment
- Adversely affect antibiotic penetration?

UCSF Cross-Linking Assisted Infection Reduction (CLAIR I and II) Trial

- Prospective randomized clinical trial
- Aravind Eye Hospital

- Do patients with culture-positive bacterial keratitis achieve microbiological cure on repeat cultures more quickly with CXL compared to antibiotics alone (day 1 repeat culture)
- Does CXL reduce perforation rate or pachymetry at 3 months controlling for baseline pachy
- Differences in acuity, astigmatism, infiltrate/scar size at 3 months?