Introduction to Corneal Cross-Linking (CXL)
Keratokonus

- Incidence approx. 1 / 2000
- > 150’000 affected in US
- Commonest cause for PK in Western countries
- ~ 20% of PK’s in US (EBAA 2008)
- ~ 50 million spent on KC healthcare in US
- Begins in puberty and can progress or arrest at any time, usually arrests at ~ age 40
- Progressive corneal thinning resulting in mixed myopic and irregular astigmatism
- Usually bilateral; though asymmetric
- May be unilateral or asymmetric at initial presentation
Agenda

1. History
2. Mechanism
3. Practical Application
4. Clinical Experience
1. History
In the 70’s polymerization compounds came up in dentistry. Compounds were fixed by cross-linking of polymer side chains.
Hornhaut
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Erhöhung der Festigkeit der Hornhaut durch Vernetzung *

Zusammenfassung
Um die Form der Hornhaut zu verändern, z.B. beim Astigmatismus, bei der Myopie und bei der Hyperopie, stehen dem Augenarzt zahlreiche Therapieverfahren zur Verfügung. Ändern sich aber die Hornhautform und -dicke, wie z.B. beim Keratoconus, so bleibt als einzige kurative Therapie die Keratoplastik. Das Krankheitsbild des Keratoconus ist klar umschrieben. Biochemische Untersuchungen zur Ursache des Keratokonus verstärkt beim Diabetes auf. Die Vernetzung wird auch gezielt zur Erhöhung der Stabilität von kollagenen Biomaterialien für Bioprothesen, wie z.B. Herzklappen, Blutgefäße und Durarserum, genutzt. In der Augenheilkunde wurde über die erste Anwendung der künstlichen Vernetzung bei der Herstellung von kollagenen Biomaterialien für die synthetische Epikeratoplastik berichtet.

**Induction of cross-links in corneal tissue.**

**Spoerl E, Huhle M, Seiler T.**

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**Abstract**

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 keratectasia. Collagenous biomaterials can be stabilized by chemical and physical agents. The epithelium of enucleated porcine eyes was removed. Eight test groups, 10 eyes each, were treated with UV-light (λ=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 cornea. The biomechanical behaviour of the cornea 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.

Number of CXL in Dresden since 1998
Cross-Linking and the Eye?

- Transforming soft tissue into a more rigid tissue
- Transforming progressive keratoconus into forme fruste keratoconus
History

- Cross-linking of human collagen is a physiological process
- Stiffening of connective tissue is well known in diabetes and aging
- Diabetes is a protection factor against keratoconus (smoking too!)
- During cross-linking new chemical bonds are induced
History

1. Combined application of UVA and riboflavin

Riboflavin (Vitamin B2)

2. Creation of oxygen radicals $O_2$

3. Induction of collagen cross-links

Speck 2006
2. Mechanism
Intra-/Interhelical and Interfibrillar XL

Speck 2006
Photochemically cross-linked collagen scaffolds gave fine microstructures with interconnected nano-sized fibers.

Photochemical cross-linking improves the physicochemical properties of collagen scaffolds.

Cross-linked vs. non-cross-linked Cornea

after

before
3. Practical Application
Indications

- Progressive keratoconus
- Iatrogenic keratoconus
- Pellucid marginal degeneration
- Bullous keratopathy
- Prevention of keratectasia
- Infectious keratitis
Contra-Indications

- Corneal thickness < 400 µ
- Pregnancy
- Prior herpetic infection
- Ruptured Descemet`s membrane
- Autoimmune disorders
Necessary Equipment for CXL

- CCL-corneal cross-linking system
- PESCHKE® riboflavin solution TE, M, D, L
- PESCHKE® H hypotonic solution in case of cornea < 400 µ
- BSS – balanced salt solution
- Topical anesthetic (i.e. oxybuprocaine, tetracaine)
- Ultrasound pachymeter
- Speculum (with blades)
- Hockey knife/Amoils brush for epi-off
- LASIK sponge for limbal protection
- Sponges, pouch
- Drapes
- Bandage contact lens
- Post-op medication: pain killer, antibiotics, lubricants
CCL Vario

Technical Specifications:
Wavelength range: 365 nm
Illumination intensity: 3 – 9 – 18 mW/cm²
Working distance: 50 mm ± 5 mm
Light emission: Continuous wave
Spot sizes (continuously adjustable): 7.0 – 11.0 mm
Timer: 30 – 10 – 5 min
Electric power: 100 – 240 V
Dimensions Hard case (w,l,h in cm): 37 x 46 x 14
Weight (total): 7,5 kg
Peschke CXL

Technical Specifications:
Wavelength: 365 nm
Illumination intensity (continuously adjustable): 3 – 30 mW/cm²
Working distance: 50 mm ± 5 mm
Fixation light with adjustable brightness
Light emission: Continuous, Interval or Pulsed
Spot sizes (adjustable): 1.0 – 11.0 mm
Timer: Automatic time adjustment / audible alarm
Auxiliary timer for Riboflavin Installation
Electric power: 100 – 240 V, 50/60 Hz
Dimensions: 28 x 12.5 x 10 cm
Weight: 1.8 kg
PESCHKE Riboflavin Solutions

**Peschke® TE - Our new transepithelial Solution for epi-on procedure**
- No removal of the corneal epithelium necessary
- Significant reduction of pain and danger of postoperative infections
- Recommended instillation time: 20 minutes (1 drop every 2 minutes = 10 drops)
- Pre-loaded glass syringe containing 2.0 ml liquid
- Ingredients: 0.25 % Riboflavin (Vitamin B2), 1.2 % HPMC, 0.01 % Benzalkoniumchloride

**Peschke® M - Standard Riboflavin Solution without Dextran for epi-off procedure**
- Does not reduce corneal thickness
- Recommended instillation time: 20 minutes (1 drop every 2 minutes = 10 drops)
- Pre-loaded glass syringe containing 3.0 ml liquid
- Ingredients: 0.1 % Riboflavin (Vitamin B2), 1.1 % HPMC

**Peschke® D - Standard Riboflavin Solution with Dextran for epi-off procedure**
- The Dresden Original: Time proven Riboflavin solution with dextran
- Recommended instillation time: 20 minutes (1 drop every 2 minutes = 10 drops)
- Pre-loaded glass syringe containing 3.0 ml liquid
- Ingredients: 0.1 % Riboflavin (Vitamin B2), 20 % dextran 500

**Peschke® H - Hypotonic Riboflavin Solution for corneal swelling**
- To swell thin corneas (< 400 μ) by means of osmotic effect
- Recommended instillation time: 1 drop every 5 seconds until corneal thickness has reached 400 μ
- Pre-loaded glass syringe containing 1.5 ml liquid
- Ingredients: 0.1 % Riboflavin (Vitamin B2)

**Peschke® L - Riboflavin Solution for use with LASIK procedures**
- For use in connection with LASIK procedures on thin corneas
- Recommended usage: after flap preparation and excimer treatment: put 3 – 5 drops on stroma, put flap back, wait 3 – 4 minutes, open flap and rinse off Riboflavin, put flap back and radiate with 1/2 of the recommended energy (1/2 of the time)
- Pre-loaded glass syringe containing 1.95 ml liquid
- Ingredients: > 0.23 % Riboflavin (Vitamin B2)
Peschke D
Riboflavin
20 %Dextran (Peschke)

Riboflavin
BAC, 0.44% NaCl
no dextran

Peschke M
no dextran
no NaCl

Peschke-TE
BAC, no dextran
no NaCl, methylcellulose

VibeX
(Avedro)

ParaCel

Ricrolin TE
ETDA
15 %dextran

Ricrolin
15% dextran
(SOOF)

ring application
iontophoresis

perfect transepithelial riboflavin
Cross-Linking Options

Peschke® D
'Dresden' Method (+ Peschke® H)

Peschke® M
(+ Peschke® H)

Peschke® D
(+ Peschke® H)

Peschke® M
(+ Peschke® H)

CXL

epi-off
3 mW
30 min

epi-on
3 mW
30 min

Peschke® TE
(+ Peschke® H)

epi-off
18 mW
5 min

epi-on
18 mW
5 min

Peschke® TE
(+ Peschke® H)
• Corneal thickness at least 400 µm.
• Oxybuprocaine or tetracaine (as needed).
• If not transepithelial ⇒ abrasio corneae Ø 8 – 9 mm.
• Peschke® riboflavin solution, 1 drop every 2 minutes for 20 minutes (AC must be yellow under blue light)
• If cornea thinner than 400 µ ⇒ swell with hypotonic PESCHKE® H solution.
• Radiate only clear cornea (protect limbal stem cells!); 1 drop BSS every 5 seconds
## Parameters and Time

<table>
<thead>
<tr>
<th>Power (mW)</th>
<th>Time (min)</th>
<th>Calculation</th>
</tr>
</thead>
<tbody>
<tr>
<td>3</td>
<td>30</td>
<td>3 mW x 1800 sec = 5400 mJ</td>
</tr>
<tr>
<td>9</td>
<td>10</td>
<td>9 mW x 600 sec = 5400 mJ</td>
</tr>
<tr>
<td>18</td>
<td>5</td>
<td>18 mW x 300 sec = 5400 mJ</td>
</tr>
<tr>
<td>30</td>
<td>3</td>
<td>30 mW x 180 sec = 5400 mJ</td>
</tr>
</tbody>
</table>
Post-operative Treatment

- Antibiotic ointment and bandage contact lens
- Antibiotic drops 3 times a day until epithelium is closed
- Pain killer (as needed)
- Lubricants as needed
- Remove contact lens when epithelium is closed (day 3)
- Post-op visits: day 1, 3, 5, 1 m, 3 m, 6 m, yearly

Steroids ➔ slow down epithelial healing!
NSAIDS ➔ can cause corneal melting!
Post-operative Observations

6 weeks:
• Slight clouding of the cornea
• Movement of cone to center $\Rightarrow$ eye gets more myopic!

3 months:
• All eyes back to at least original K-values.

6 months:
• Average flattening $\sim$ 2 diopters
Safety Aspects - Riboflavin Shielding

**Damage threshold - endothelium**

- 0.3 mW/cm²

**Graph:**

- 3 mW/cm²
- 100% damage
- 100μm to 600μm

**Values:**

- 0μm: 3.00 mW/cm²
- 100μm: 1.49 mW/cm²
- 200μm: 0.74 mW/cm²
- 300μm: 0.36 mW/cm²
- 400μm: 0.18 mW/cm²
- 500μm: 0.09 mW/cm²
- 600μm: 0.06 mW/cm²

*Speck, 2006*
Safety Aspects - Keratocites

300 μ
Safety Aspects - Corneal demarcation line
Safety Aspects - Confocal Microscopy

- Cross-linking kills keratocytes 300 μm deep
- Re-population takes 6 months

A. Caporossi, MD
4. Clinical Experience
Clinical Experience

1. Biomechanical Stability

2. Biochemical Stability
Degree of Cross-Linking in Relation to Corneal Thickness

Biomechanics after CXL

- Increase of tangential solidity
- Restructuration and reshaping of the cornea
- Move of corneal apex to center
- Regularization of corneal surface
- Increased contact lens tolerance
- Reduction of corneal sensitivity
Biochemical Effects
First report 2004

Case 2

- filamentous fungus (Acremonium) 10 weeks after LASIK
- 4 days after CCL

M.A. Thiel, Lucerne
Patient no. 2

Biochemical Stability

- 84 year old woman
- Symptoms since one week
- Large ulcer, hypopyon
- BCVA Hand Movements

Mortensen/Makdoumi, 2008
Patient no. 2

Biochemical Stability

- Treatment with CXL day of diagnosis
- Greatly reduced symptoms day after treatment
- Disappearance of hypopyon
- Slow epithelialization
- Picture after one week

Mortensen/Makdoumi, 2008
Patient no. 2

Biochemical Stability

• Follow-up after six months
• BCVA 20/200
• No epithelial defect
• Large macula nasally
• Complete epithelialization

Mortensen/Makdoumi, 2008
Mechanism of Action
Effects and Reasons

• Gram positive + gram negative strains
• Healing one week to several months
• Quick improvement: pain, light sensitivity, inflammation, hypopyon
• Reduced corneal sensation
• Effect on Langerhans and dendritic cells
• Inhibitory effect on corneal immune system (?)
• Increased resistance against collagenasis
• Induction of apoptosis of corneal keratocites modify environment for pathogens
Other Applications
Results for Keratoconus
Results in Keratoconic Eyes

maximal K-reading / D

9-03  3-04  9-04  3-05  9-05  5-06

52  54  56  58

cross-linking
Results in Keratoconic Eyes
Results in Keratoconic Eyes

- No adverse reactions except haze
- According to corneal topography, progression halted in every case
- Corneal thickness decreases after surgery
Results in Keratoconic Eyes

max. K-readings decrease!

Cross-Linking

Prof. Dr. E. Spoerl, 2006
Results in Keratoconic Eyes

BSCVA increases!

Prof. Dr. E. Spoerl, 2006
Results in Keratoconic Eyes

- BSCVA increases
- Corneal surface regularizes, keratoconus indices decrease > 60 % of all patients!

Prof. Dr. E. Spoerl, 2006
Safety:
Lines lost / gained – 1 year

[Bar chart showing lines lost/gained for UCVA and BSCVA over a range of values from -4 to 12, with values ranging from 0 to 16.]

Reiter, 2010
Clinical Observations

- No permanent side effects or adverse reactions (haze, myopia)
- According to corneal topography, progression halted in every case
- Maximal K-readings were significantly reduced in 65% of the cases
- Duration of treatment 30 – 60 min
Iatrogenic Keratectasia
Results in Iatrogenic Ectasia

- LASIK for -4 D
- Pre-op pachymetry 500 μm, no signs of FFKC
- Bilateral iatrogenic keratectasia starting 12 months after surgery
- Cross-linking right eye
12 Months CXL follow-up

Cross-Linking, $K_{\text{max}}$: 57,27

3.5 months po, $K_{\text{max}}$: 54,87

5 months po, $K_{\text{max}}$: 54,55

12 months po, $K_{\text{max}}$: 53,96
Corneal thickness should be at least 400 μm.

What shall we do with thin corneas (<400 μm)?

- The human cornea can swell up to 2 - 3-fold in thickness
- Let the cornea swell by means of hypotonic riboflavin solution:

  PESCHKE® H hypotonic solution

Important: 1 drop every 5 seconds!!!
Special cases

Instillation of PESCHKE® hypotonic solution

pre  post  difference
Pachymetry < 400μm

- 22 yr, ♂
- BCVA: 0.8
- ORA: CH 6.9mmHg
- Pachymetry: 360μm
- w/o epithelium: 280μm
- After application of hypotonic riboflavin: 495μm
Future

- **CXL in bullous keratopathy and cornea edema**
  - Kanellopoulos, CXL Congress 2007
  - Ehlers N. et al, CXL Congress 2008

- **CXL for the treatment of infectious keratitis**

- **CXL in combination with intracorneal ring-segments**
  - Fabiani L., Las Palmas 2007, SEO
  - Coscunseven E. et al, CXL Congress 2008

- **CXL and PRK / Lasik**
  - Kanellopoulos, CXL Congress 2008

- **Scleral cross-linking – Stabilisation of Myopia**
  - Iseli H. P. et al, CXL Congress 2008
Conclusion

- Corneal cross-linking is a minimally invasive procedure to improve biomechanical and biochemical stability of the cornea.
- Progression of keratoconus and corneal melting can be stopped with corneal cross-linking.
- Treatment is inexpensive and easy to handle.
- Safety issues: 400 µm corneal thickness required to prevent endothelial damage. In case of thinner corneas use hypotonic riboflavin solution to swell cornea.
Thank you!