

# Adapting CXL technology to veterinary ophthalmology

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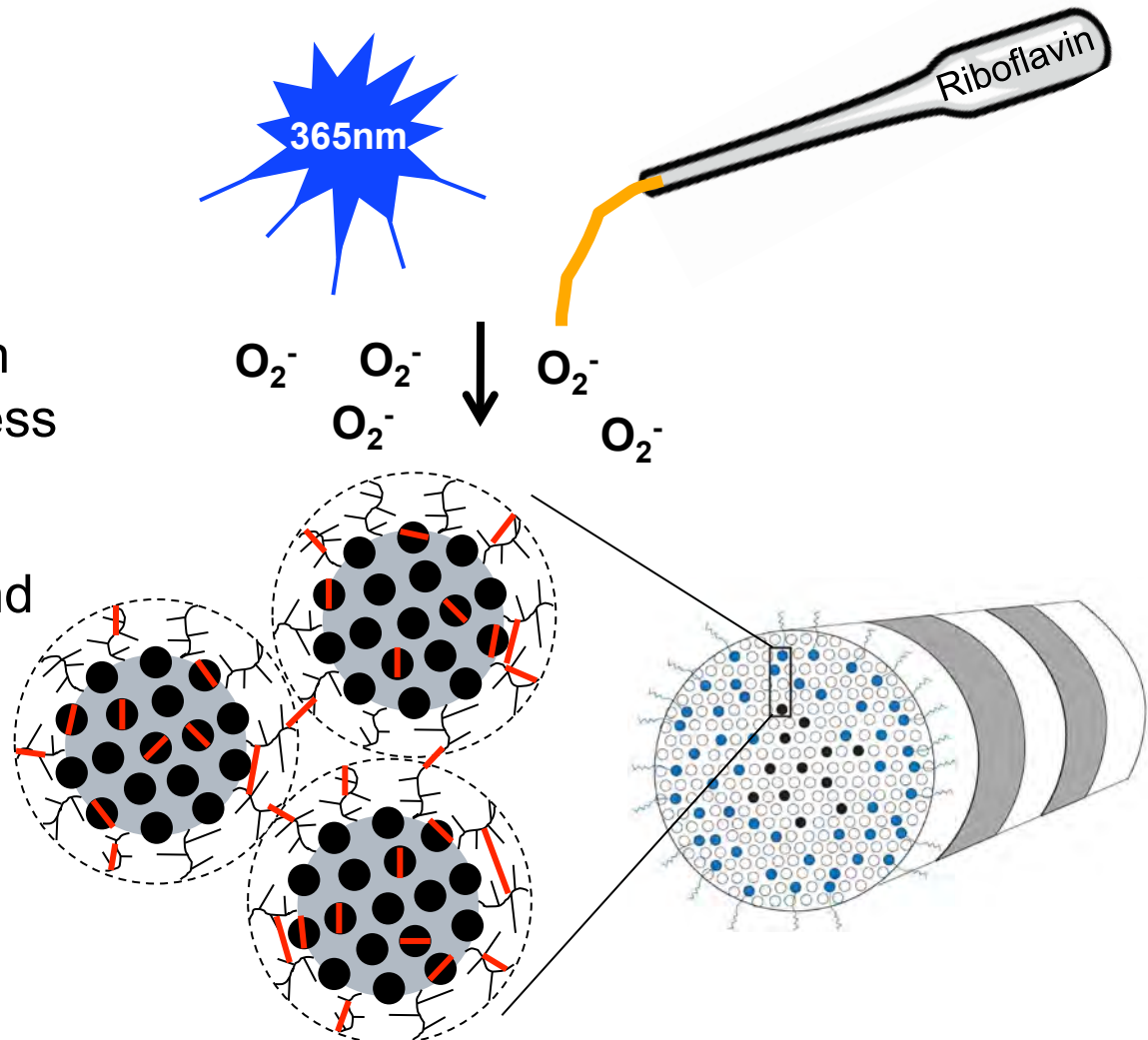
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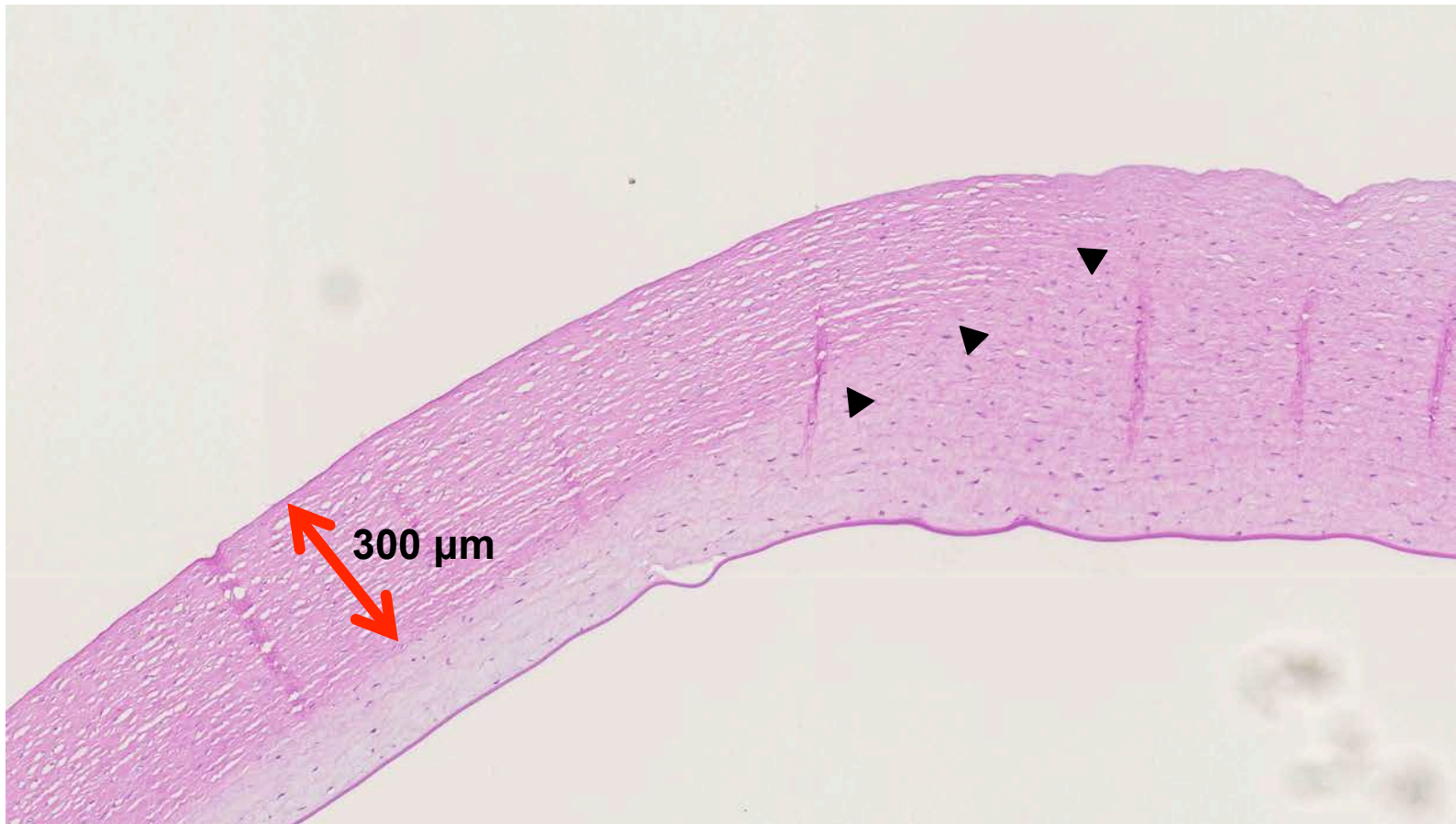
## UV-A/Riboflavin corneal collagen crosslinking (CXL)

- Combined application of riboflavin and UVA
- Free oxygen radical-driven photopolymerization process
- Covalent cross-links on and within collagen fibers and in proteoglycan-rich ground substance



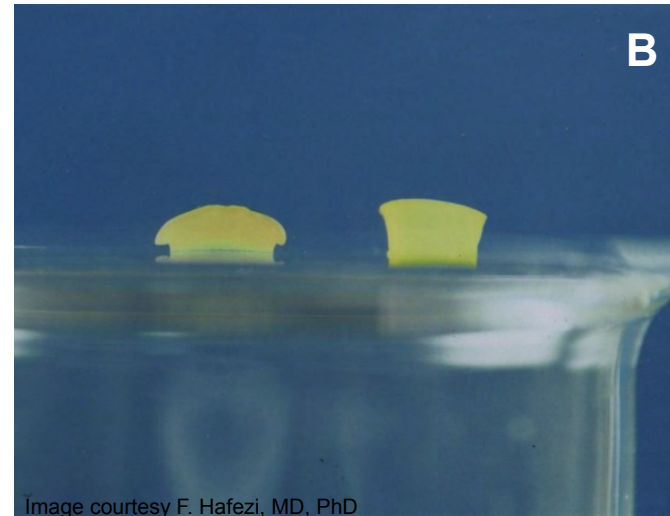
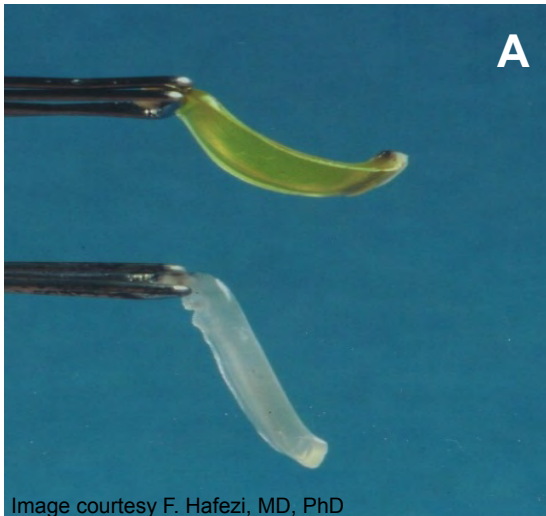


## CXL: effect



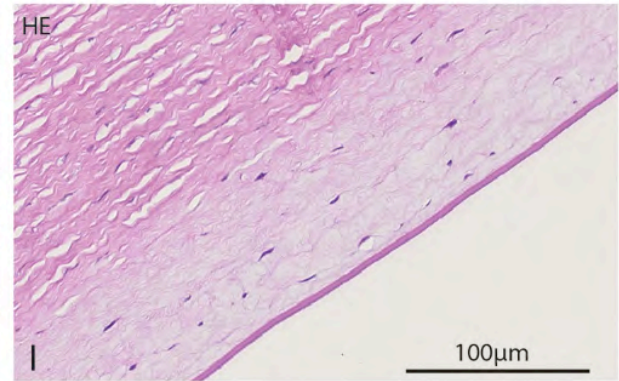
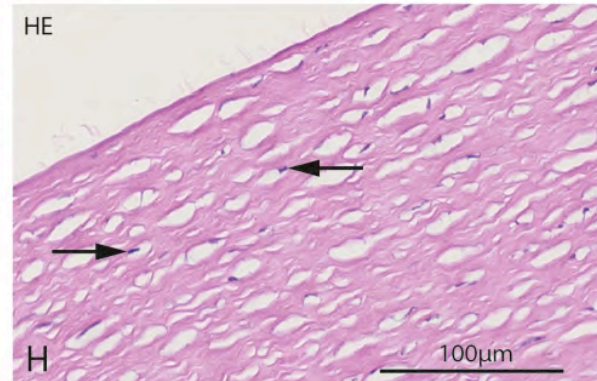
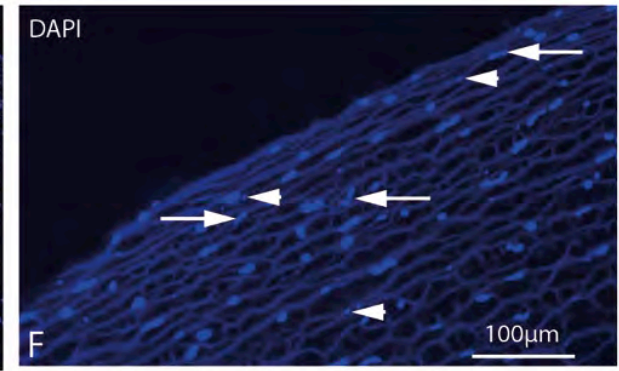
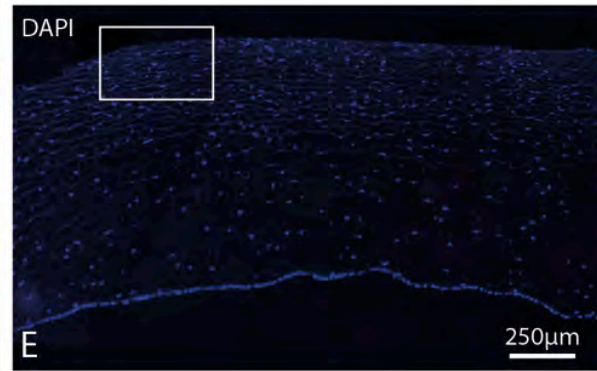
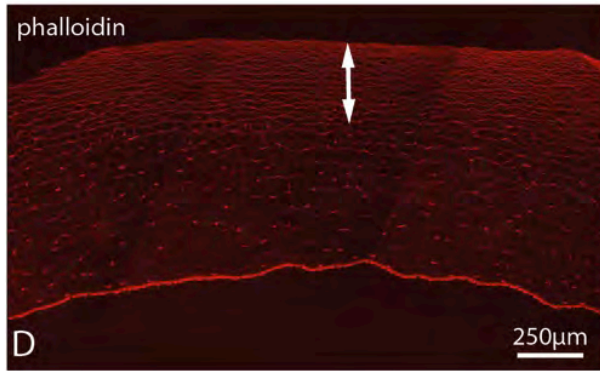
## CXL: basic concept

- Additional molecular bonds within collagen fibers and PG-GAG ground substance of the corneal stroma
  - Increased biomechanical stability (A)
  - Increased biochemical stability (B)



- Intercalation with microorganismal RNA/DNA
- Direct free radical-induced elimination of microorganisms

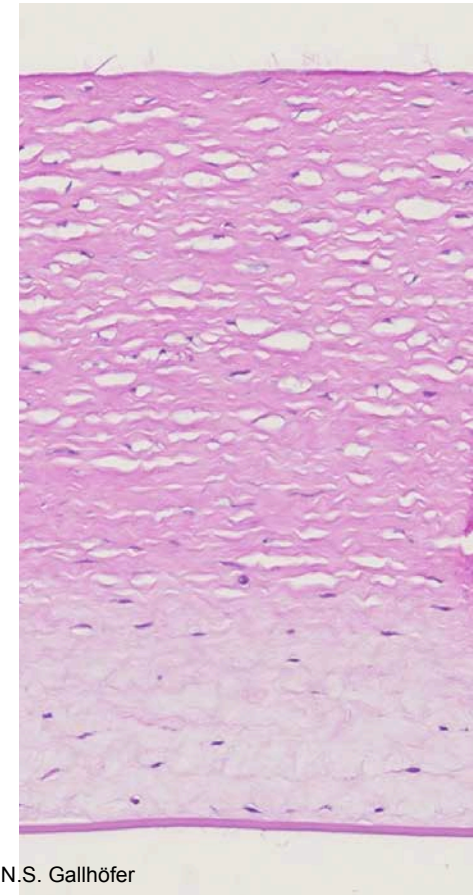
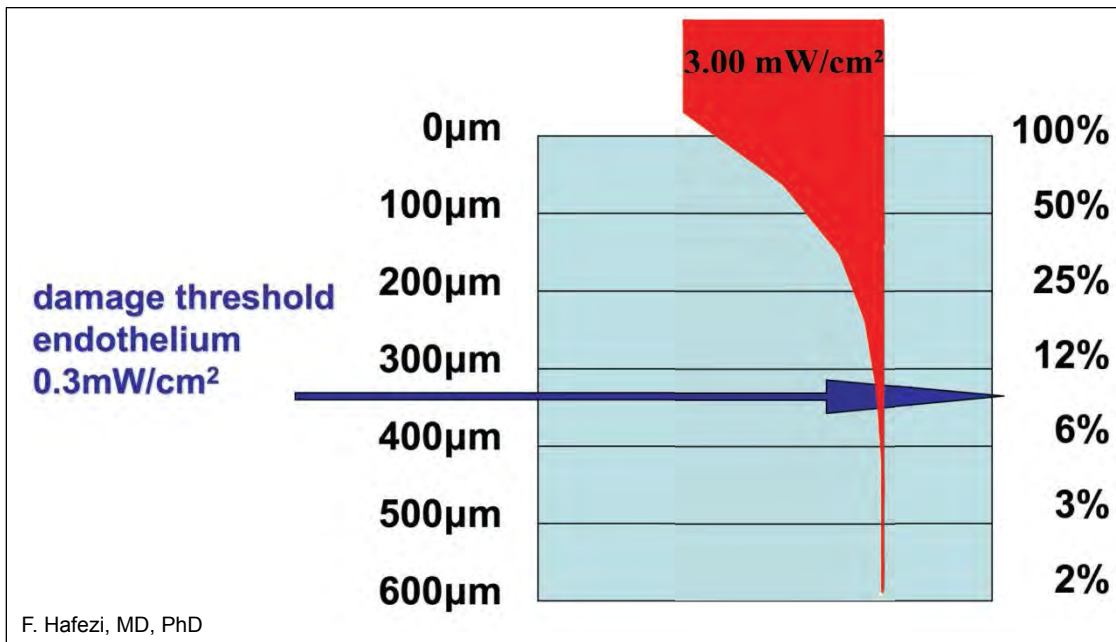
## The trade-off



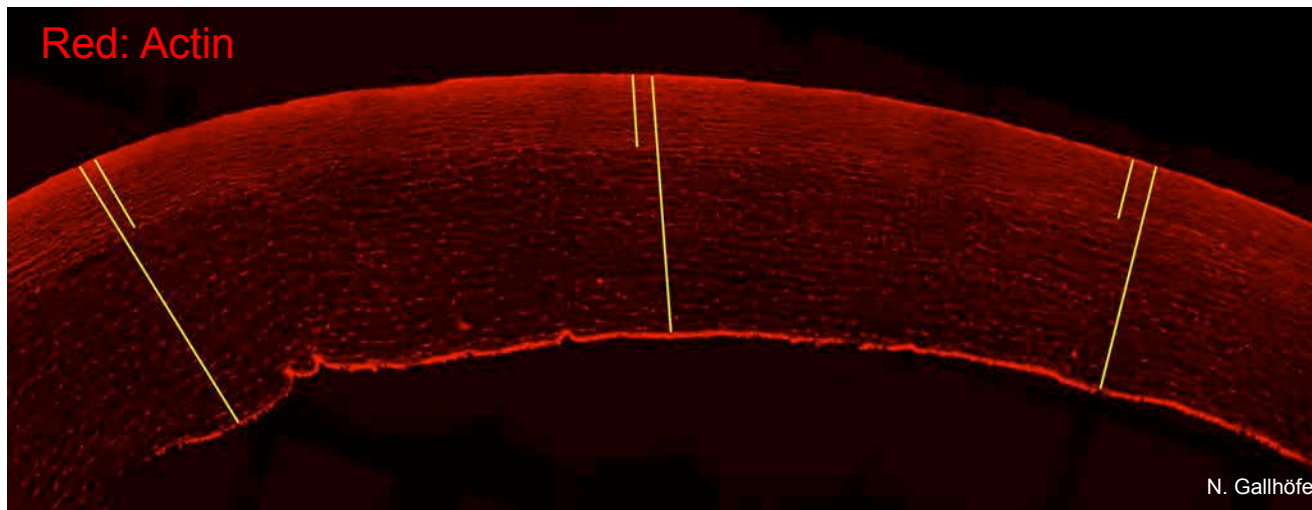
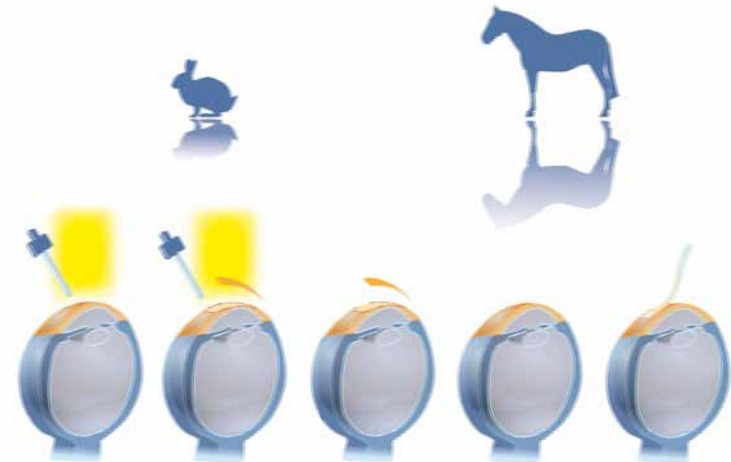
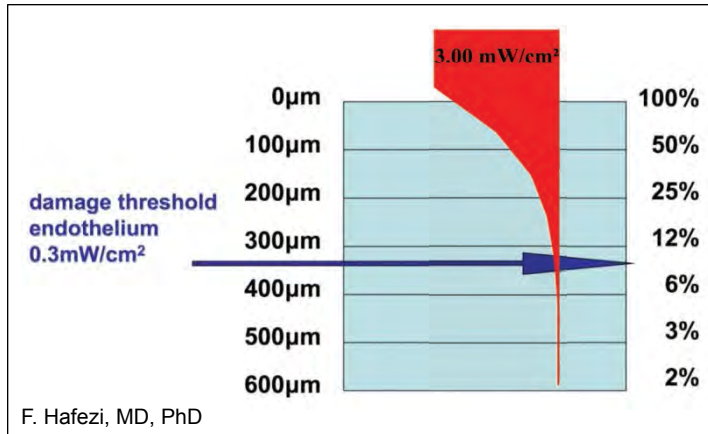
- Gallhoefer, N.S., et al., Penetration depth of corneal cross-linking with riboflavin and UV-A (CXL) in horses and rabbits. *Vet Ophthalmol*, 2016. 19(4): p. 275-84.

## CXL: Riboflavin shielding

- Riboflavin enhances absorption of 365nm UV light in cornea
  - Efficacy UVA crosslinking ↑↑
  - Energy absorption: shielding of deeper structures
- Cell death depending on rate of UVA absorption
  - Keratocytes, corneal endothelial and lens epithelial cells



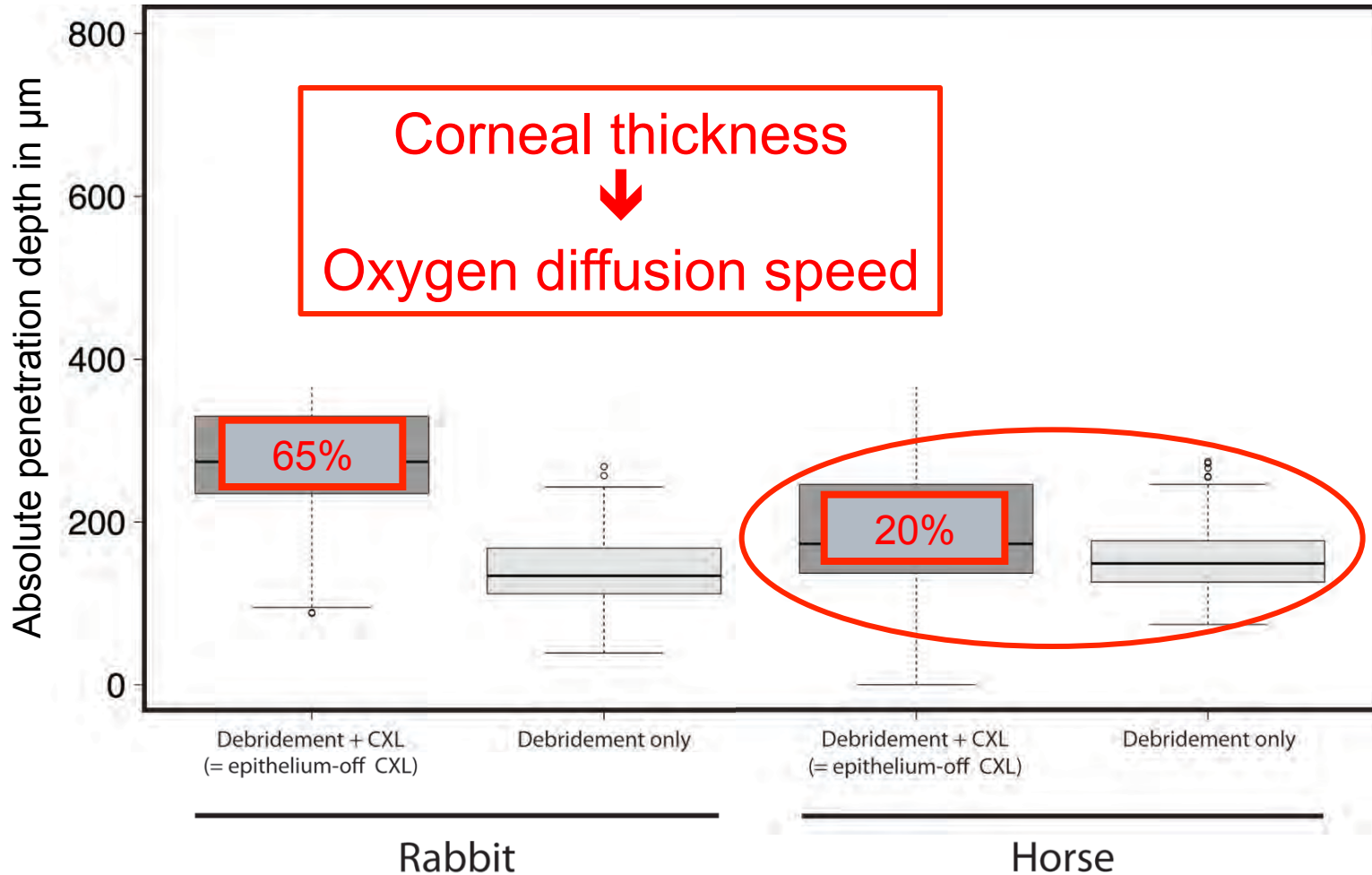
## CXL: interspecies variations



- Gallhoefer, N.S., et al., Penetration depth of corneal cross-linking with riboflavin and UV-A (CXL) in horses and rabbits. *Vet Ophthalmol*, 2016. 19(4): p. 275-84.



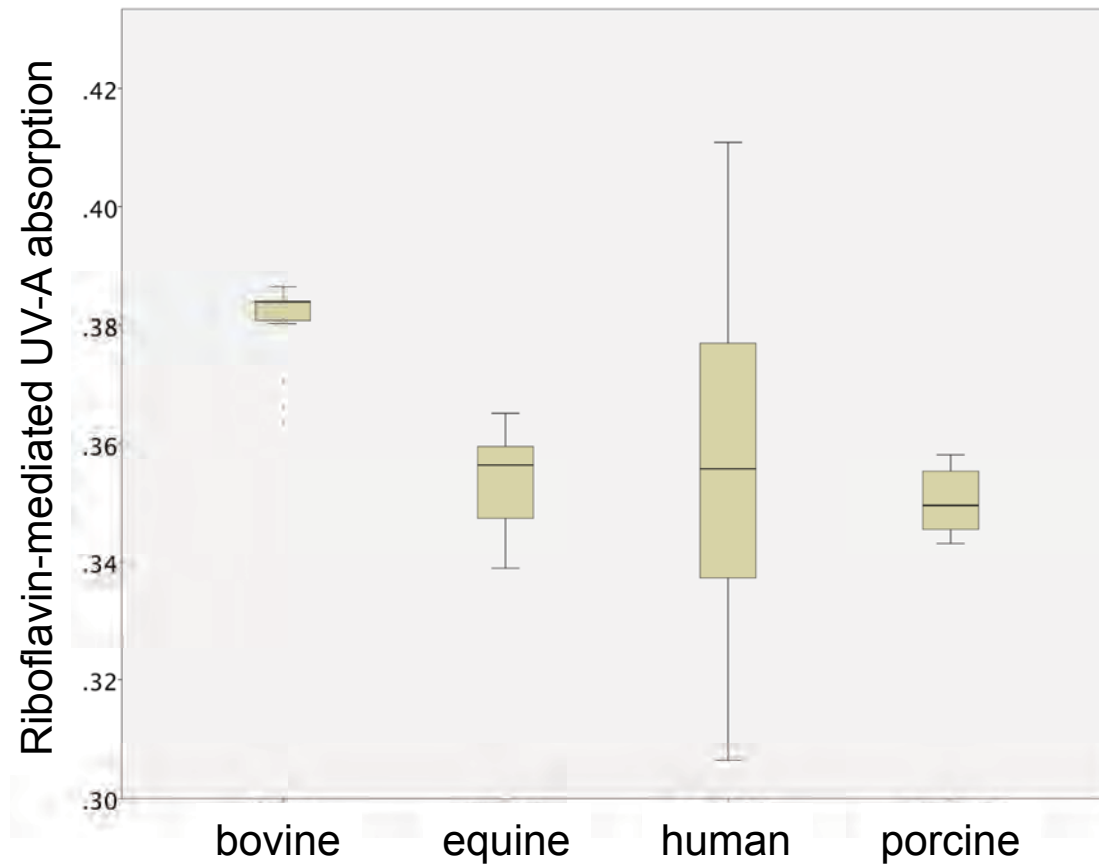
## CXL: interspecies variations







## UVA absorption: interspecies variations



- Wuarin R, Richo O, Kling S, Pot SA, Tabibian D, Salmon B, Hafezi F. Riboflavin-mediated UV-A absorption in corneal cross-linking is species-dependent. JRS



## CXL: indications in veterinary ophthalmology

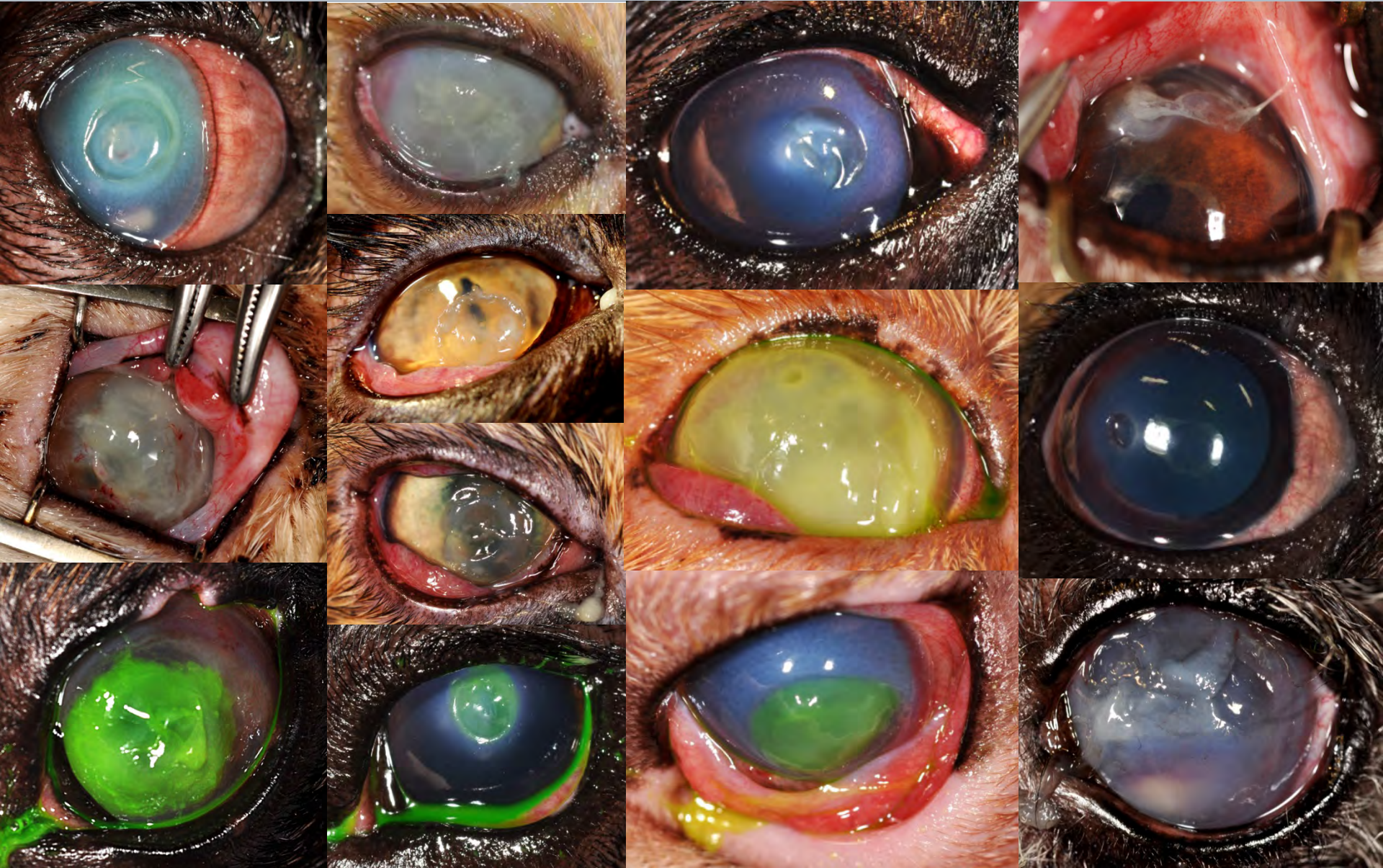
- Bullous keratopathy
  - Treatment of painful recurrent erosions



- Pot SA, Gallhofer NS, Walser-Reinhardt L, et al. Treatment of bullous keratopathy with corneal collagen cross-linking in two dogs. *Vet Ophthalmol.* 2013.
- Famose F. Evaluation of accelerated corneal collagen cross-linking for the treatment of canine bullous keratopathy (10 eyes - 8 dogs), Proceedings ECVO conference London 2014.

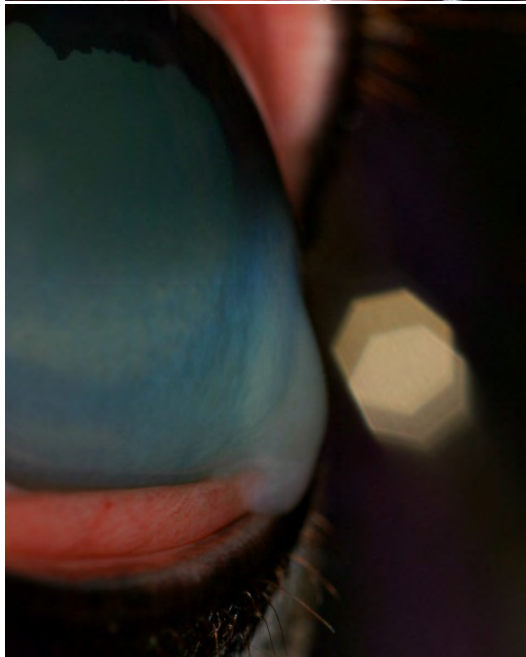


Veterinary Ophthalmology





## Keratomalacia - melting keratitis



- Collagenase enzyme activity
- Typically bacterial infections in companion animals
- Fight structural degradation:
  - Medical Tx
  - Surgical Tx
- In some cases **no timely control** of stromal degradation.



## The many faces of globe perforation





University of  
Zurich<sup>UZH</sup>

Veterinary Ophthalmology



University of Bern | University of Zurich

vetsuisse-faculty

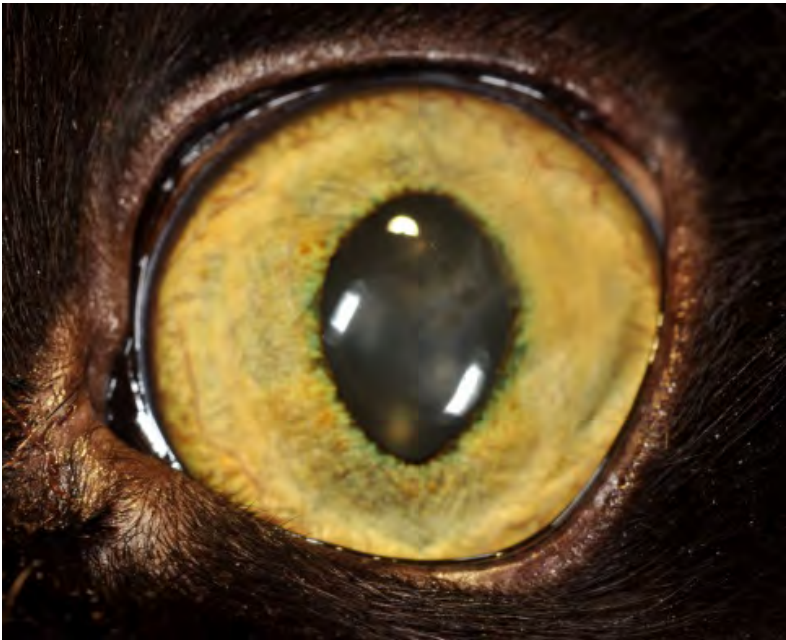
## Melting ulcer therapy

UW-Madison, Veterinary Ophthalmology





## Melting ulcer therapy: outcome





## Melting ulcer therapy

- E-collar
- Broad spectrum antibiotics every 1-2 hours (Neomycin/Polymyxin/Gramicidin, Ofloxacin)
- Collagenase-inhibitors (6-12 times daily):
  - Autologous serum
  - EDTA
- Atropine to effect
- Systemic non-steroidal anti-inflammatories
- Systemic antibiotics (Globe perforation)







# Antibiotic resistance: an emerging problem?

## WHO report on antibiotic resistance 2014

**ANTIMICROBIAL RESISTANCE**  
Global Report on surveillance 2014

**What you need to know**  
WHO's first global report on antimicrobial resistance, with a focus on antibiotic resistance, reveals that it is no longer a prediction for the future. Antibiotic resistance - when bacteria change and antibiotics fail - is happening **right now**, across the world.

**The report is the most comprehensive picture to date**, with data provided by 114 countries.

**Looking at 7 common bacteria** that cause serious diseases from bloodstream infections to gonorrhoea.

**High levels of resistance** found in all regions of the world.

**Significant gaps** exist in tracking of antibiotic resistance.

**Over the last 30 years, no major new types of antibiotics have been developed**

Timeline: 1910 (Penicillin), 1930 (Cephalosporin), 1970 (Carbapenem), 1980 (Fluroquinolones), 1990-2010 (Discovery void)

**What does this mean?**  
Without urgent action we are heading for a post-antibiotic era, in which common infections and minor injuries can once again kill.

**How can infections be prevented in the first place to reduce the need for antibiotics?**

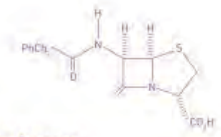
- Better hygiene
- Access to clean water and sanitation
- Infection control in healthcare facilities
- Vaccination

**What you can do**

- Use antibiotics only when prescribed by a health professional
- Complete the full prescription, even if you feel better
- Never share antibiotics with others or use leftover prescriptions

World Health Organization

More information at [www.who.int/drugresistance](http://www.who.int/drugresistance)



- Antibacterial resistance is on the rise
  - Post-antibiotic era coming?



The report is the most comprehensive picture to date, with data provided by 114 countries



Looking at 7 common bacteria that cause serious diseases from bloodstream infections to gonorrhoea



High levels of resistance found in all regions of the world



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### What does this mean?

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How can infections be prevented in the first place to reduce the need for antibiotics?



Better hygiene



Access to clean water and sanitation



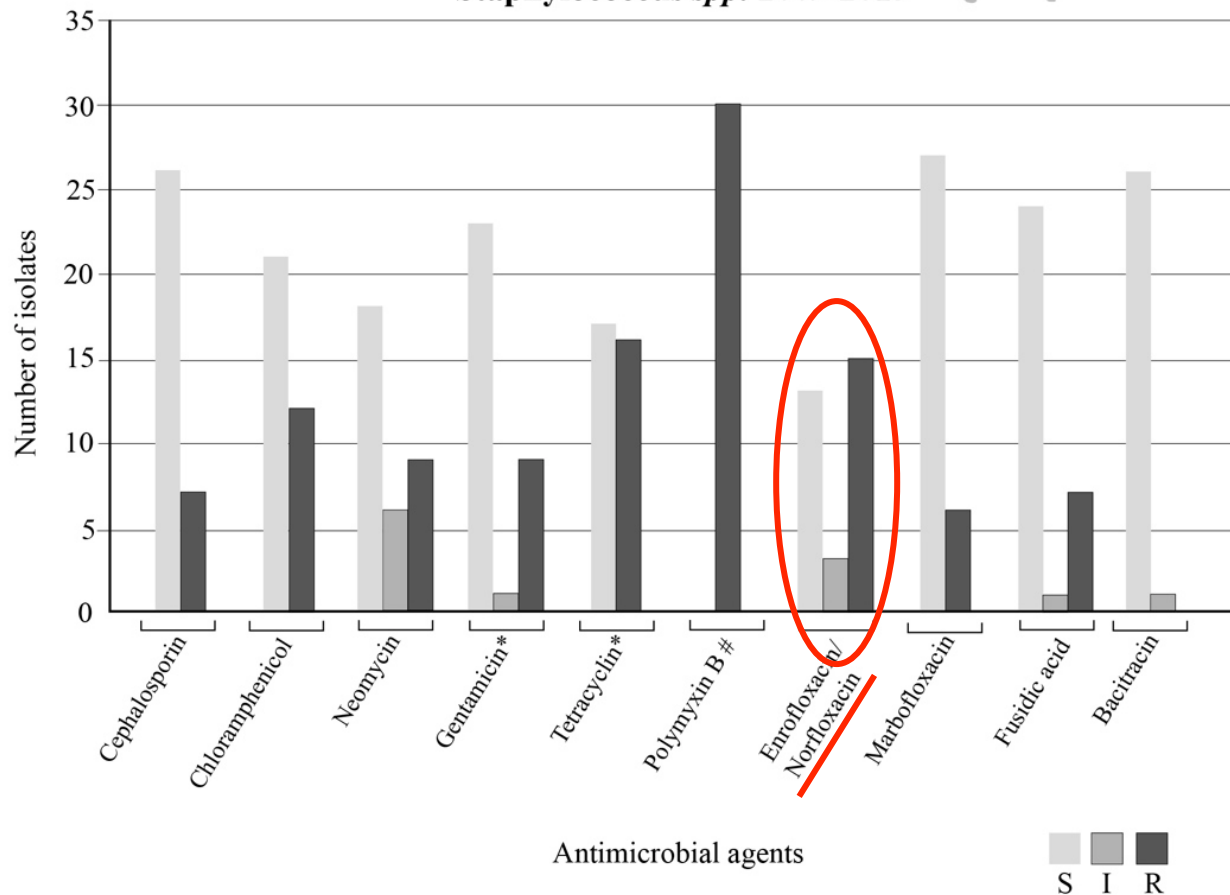
Infection control in healthcare facilities



Vaccination

# Antibiotic resistance

Canine isolates  
*Staphylococcus spp.* 2009-2013

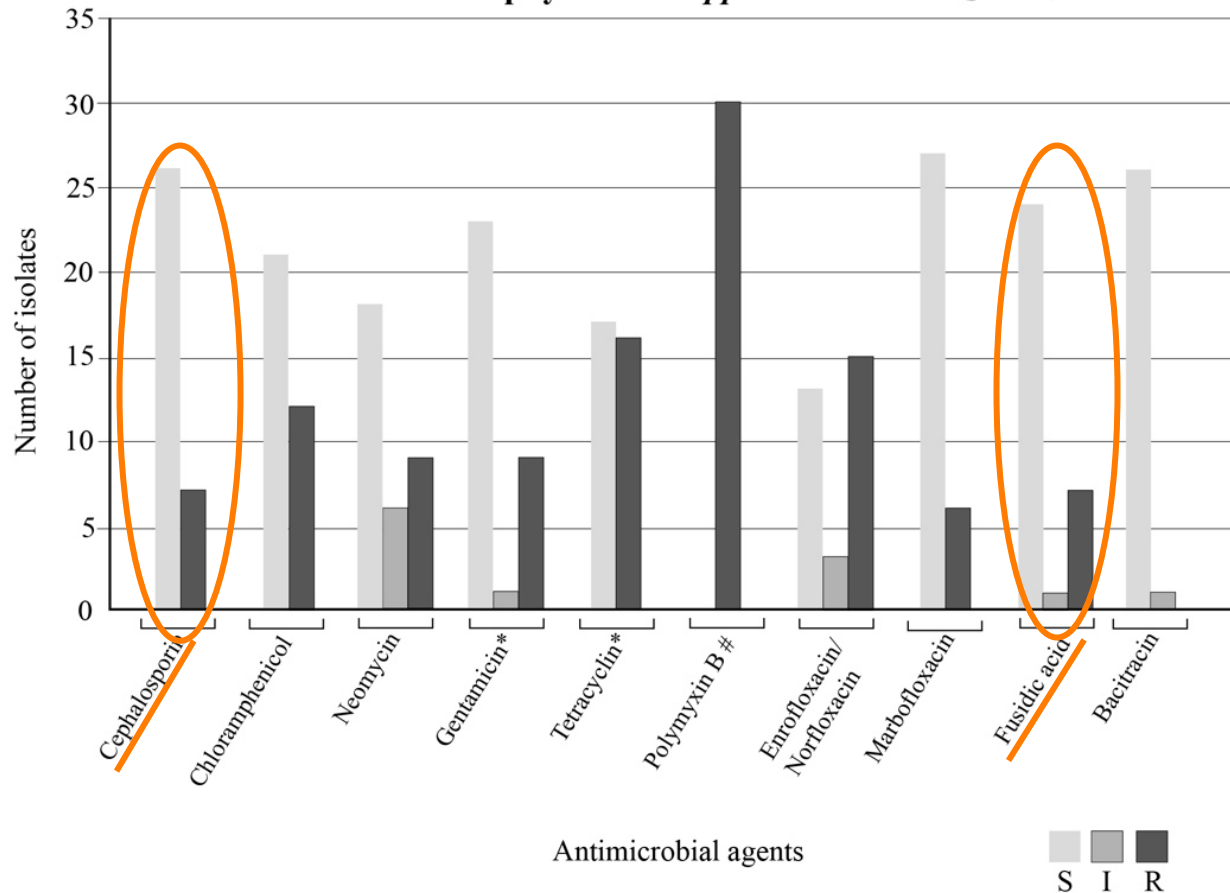


Resistance to 1<sup>st</sup> and 2<sup>nd</sup> generation Fluoroquinolones

# Antibiotic resistance



Canine isolates  
Staphylococcus spp. 2009-2013



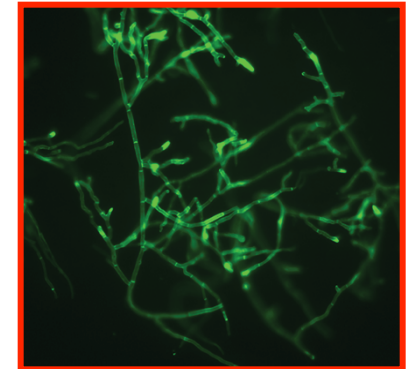
- Suter A, Voelter K, Hartnack S, Spiess BM, Pot SA. Septic keratitis in dogs, cats and horses in Switzerland; associated bacteria and antibiotic susceptibility. Submitted: Vet Ophthalmology

Resistance to cefalexin or fusidic acid in +/- 20% of isolates



## Equine Keratomycosis in Switzerland

- Healthy and diseased equine eyes sampled
- **Aspergillus fumigatus**
- No Fusarium spp. (but prevalent in other regions)
- Hospitalization  $\approx$  15 days, Total Tx time  $\approx$  6 weeks
- 64% of globes saved



- Voelter Ratson K, Pot SA, Florin M, Spiess BM. Equine Keratomycosis in Switzerland: a retrospective evaluation of 35 horses (Jan 2000 - Aug 2011). Equine Vet J. 45(5), 2013: 608-612.
- Voelter Ratson K, Monod M, Unger L, Spiess BM, Pot SA. Evaluation of the conjunctival fungal flora and its susceptibility to antifungal agents in healthy horses in Switzerland. Vet Ophthalmol. 17(s1), 2014: 31-36.
- Voelter-Ratson K, Monod M, McMullen Jr. RJ, Spiess BM, Pot SA. Evaluation of fungal flora isolates from equine keratomycosis patients: species and susceptibility patterns. Vet Ophthalmol. In preparation.



## PACK-CXL

**P** hoto

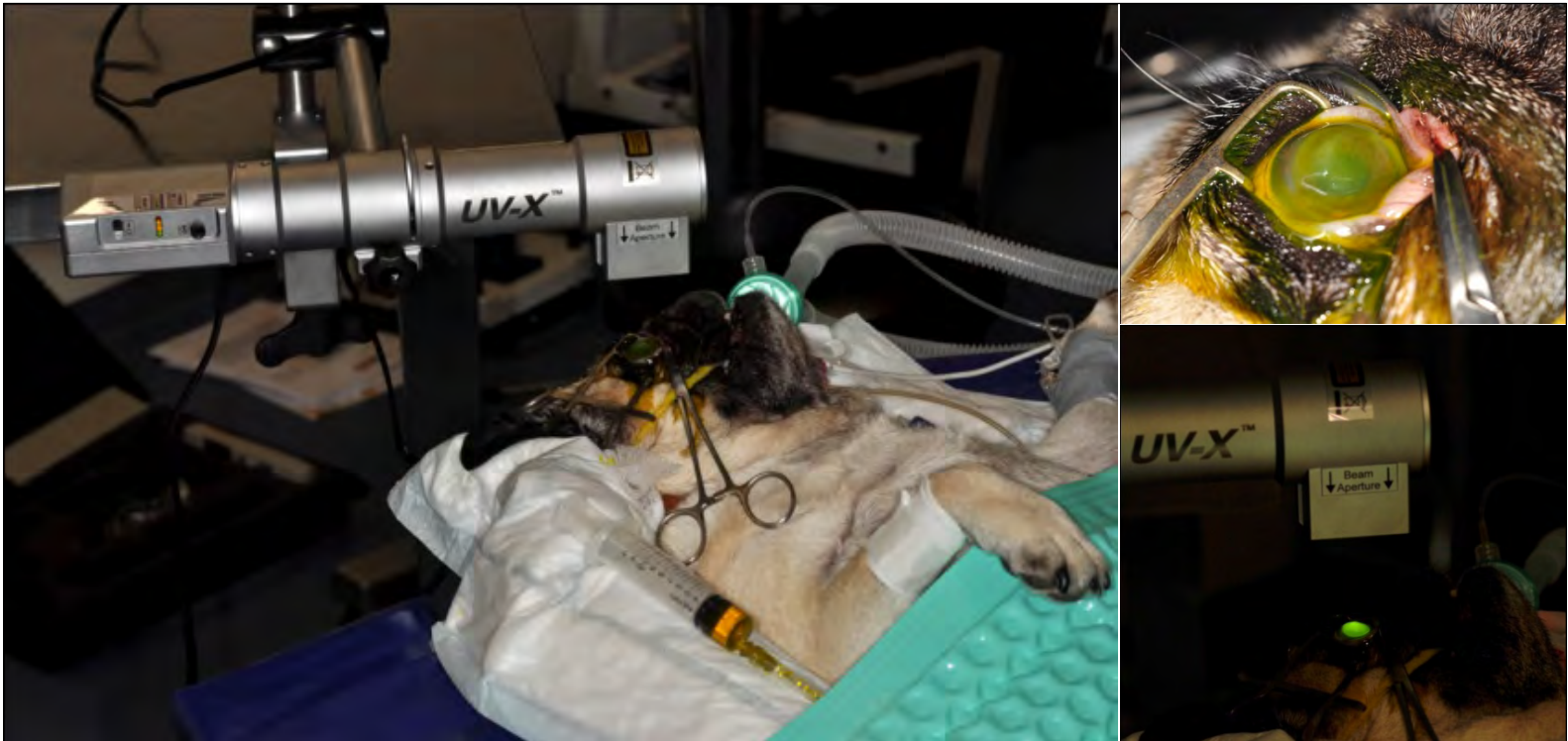
**A** ctivated

**C** hromophore for

**K** eratitis



## PACK-CXL: current clinical set-up Zürich



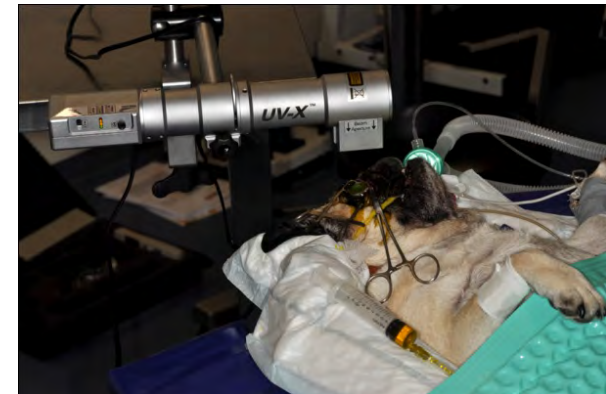
- General inhalation anesthesia





## PACK-CXL: current clinical set-up Zürich

- General inhalation anesthesia
- 30 min. riboflavin saturation, 30 min. UVA at 3mW
  - No fluorescein staining prior to PACK-CXL
    - Careful wound bed debridement
- Modulation of corneal (wound bed) thickness:  
hypo/hypertonic solutions

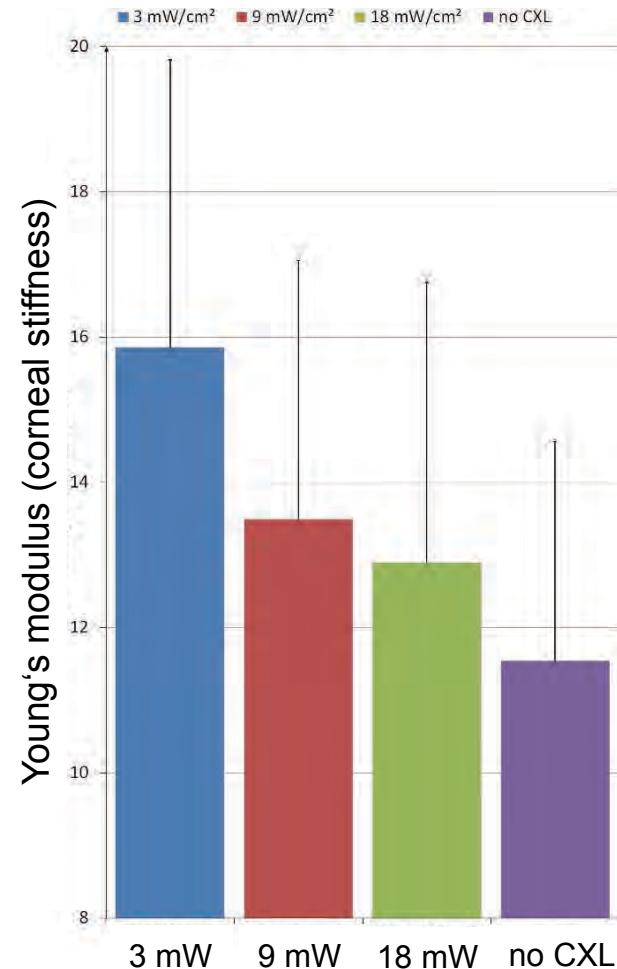


## PACK-CXL: faster

- 5.4 J/cm<sup>2</sup>...
  - 30 minutes at 3mW
  - 10 minutes at 9mW

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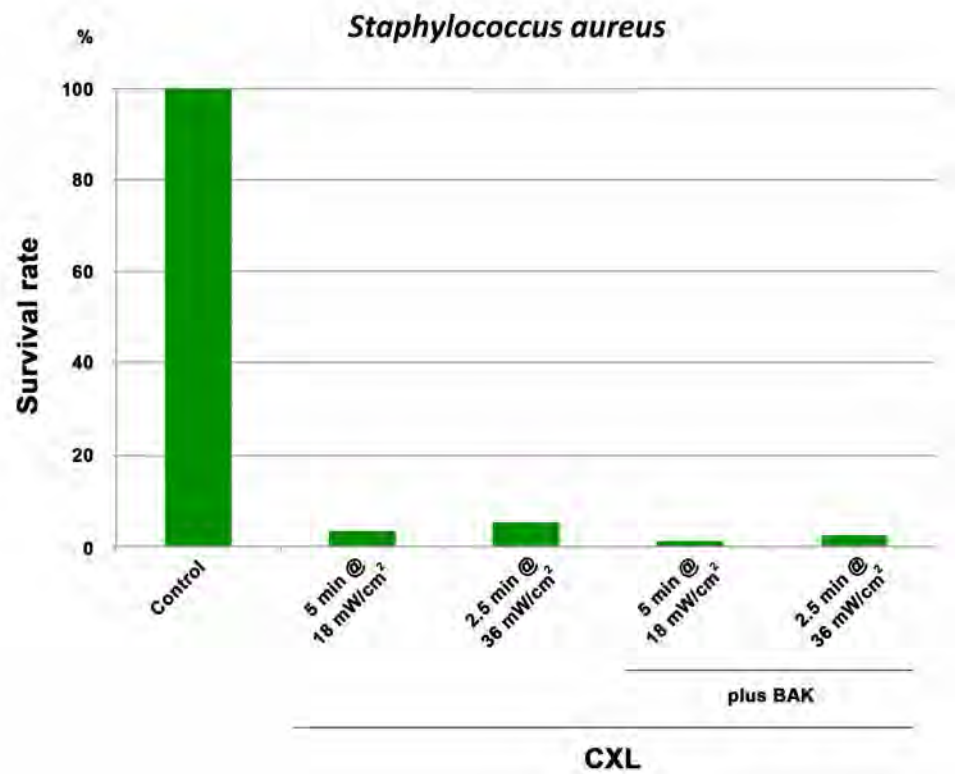
  - 5 minutes at 18mW
  - 3 minutes at 30mW
  - 2.5 minutes at 37.5mW
  - 2 minutes at 45mW





## PACK-CXL: faster

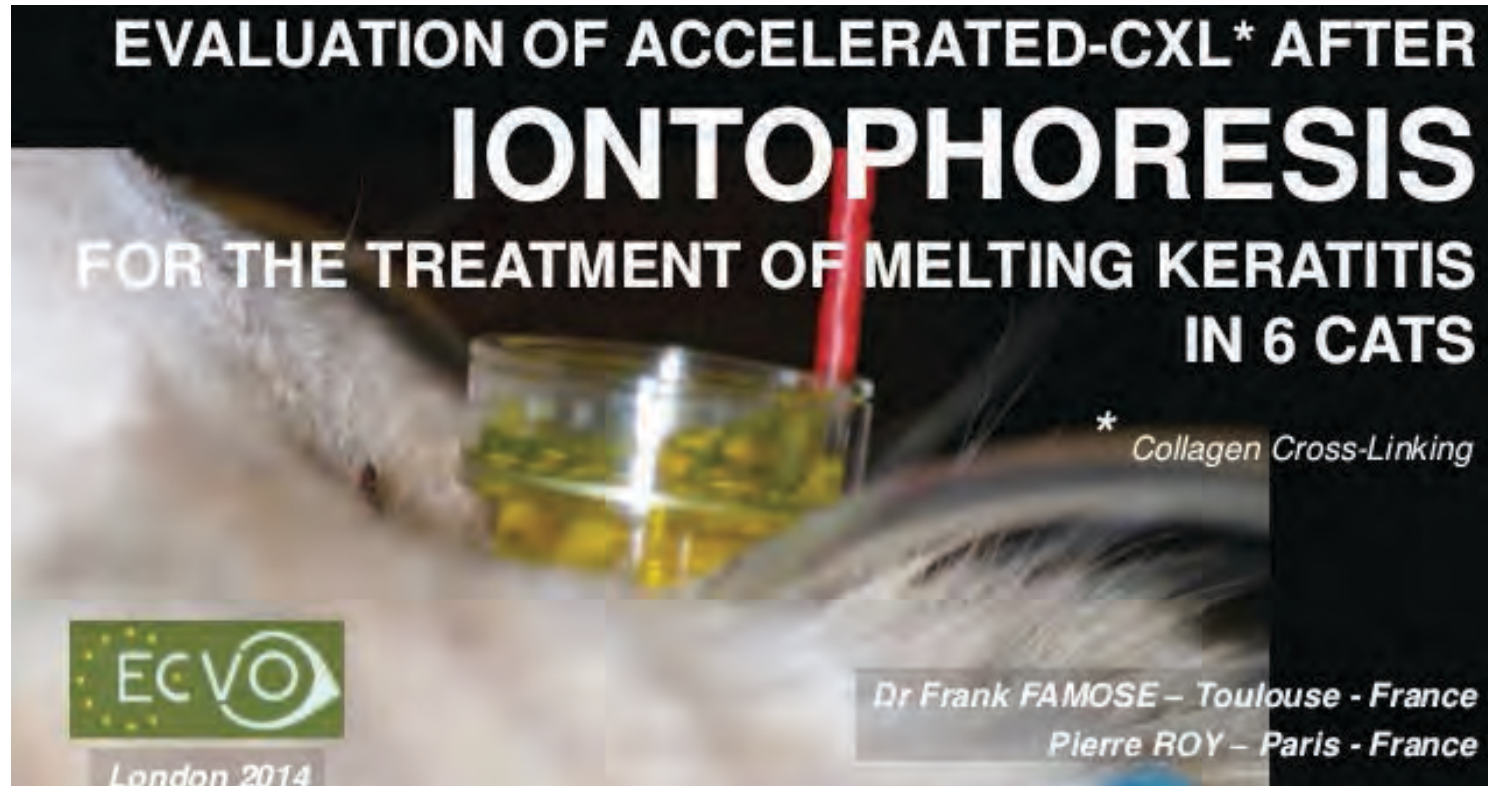
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  - 2 minutes at 45mW



• Richo, O., et al., Antibacterial efficacy of accelerated photoactivated chromophore for keratitis-corneal collagen cross-linking (PACK-CXL). J Refract Surg, 2014. 30(12): p. 850-4.



## PACK-CXL: faster

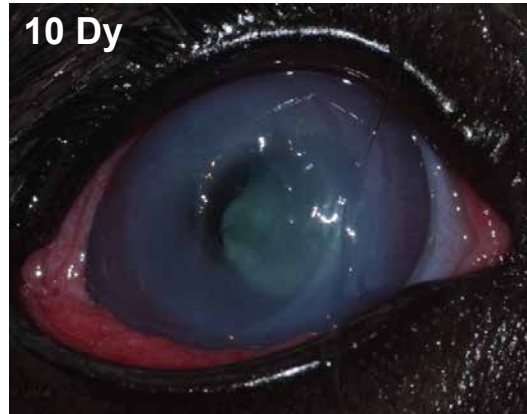


30 → 5 minute riboflavin saturation

- Cassagne, M., et al., Iontophoresis Transcorneal Delivery Technique for Transepithelial Corneal Collagen Crosslinking With Riboflavin in a Rabbit Model. Invest Ophthalmol Vis Sci, 2016. 57(2): p. 594-603.



## PACK-CXL: adjunct to surgery





**Questions ?**