Corneal diagnostic techniques

Sara Thomasy DVM, PhD, DACVO

With appreciation for slides provided by Drs. Ellison Bentley
Corneal Innervation

• Critical to corneal health
• ↓ innervation
  – Poor wound healing
  – Poor endothelial cell function
Innervation of the canine cornea
Aesthesiometry

• Measurement of corneal sensation
• Quantitative vs qualitative
• No topical anesthesia
• Blink must be present
Low-tech aesthesiometry

• Cotton-tip applicator-use wisp, gently touch
• Look for: blink, withdrawal
• Qualitative only-detects large amount of change
High-tech Aesthesiometry

- **Cochet-Bonnet**
  - Most common
  - Quantitative measurement
  - 6.0 cm nylon filament
  - Touch cornea with filament
  - Response = blink

- **Larson-Millodot**
  - Platinum wire
  - Automated (advance/retract)
  - Defined pressure
C-B aesthesiometer

- Shorten filament 0.5 cm until response
- Filament length inversely related to pressure
C-B aesthesiometer

- Corneal sensitivity-reported in cm or mm
- Conversion table for each type of aesthesiometer
- Range: 6.0-0.5 cm = 0.96-17.68 g/mm²

<table>
<thead>
<tr>
<th>Filament length, cm</th>
<th>6.0</th>
<th>5.5</th>
<th>5.0</th>
<th>4.5</th>
<th>4.0</th>
<th>3.5</th>
<th>3.0</th>
<th>2.5</th>
<th>2.0</th>
<th>1.5</th>
<th>1.0</th>
<th>0.5</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean values of pressures, g/mm²</td>
<td>0.96</td>
<td>1.08</td>
<td>1.16</td>
<td>1.40</td>
<td>1.84</td>
<td>2.40</td>
<td>3.20</td>
<td>4.60</td>
<td>6.64</td>
<td>8.84</td>
<td>12.84</td>
<td>17.68</td>
</tr>
</tbody>
</table>
C-B aesthesiometer response

- Blink/percentage of blinks
- But how much blink?
- Repeatable?
C-B aesthesiometer

- Behavior
- Cats???
- Variability in data
C-B aesthesiometer

- Other sources of variability:
  - Filament changed between 1990-2010: New conversion table
    - Old 60=11mg
    - New 60=5mg
  - Filament doesn’t actually produce consistent pressure
    - Variability increases as the filament is shortened
    - 30 mm filament length 21+/- 8.7 with a range of 3-35

Wieser, 2013 VO
Clinical Application of Aesthesiometry

WAIT. HOLD ON. STOP TALKING.

SHOW ME THE DATA
A comparison of corneal sensitivity between brachycephalic and Domestic Short-haired cats

Tiffany Blocker and Alexandra van der Woerdt

Department of Medicine, The Bobst Hospital of the Animal Medical Center, 510 East 62nd Street, New York, New York 10021, USA

- Corneal sensitivity:
  - Significantly less in brachycephalic vs. DSH
  - Central and peripheral cornea
    - Central >> peripheral in both groups
  - Similar findings to dogs:

Corneal sensitivity in dogs with diabetes mellitus.

Good KL, Maggs DJ, Hollingsworth SR, Scagliotti RH, Nelson RW.

- Diabetic dogs significantly less sensitive vs. normal
  - 5 locations: central, dorsal, ventral, medial, lateral
- No correlation between CTT:
  - Duration of diabetes mellitus
  - Blood glycated protein concentrations
What about horses?
Cochet–Bonnet aesthesiometer-determined corneal sensitivity in neonatal foals and adult horses

D. E. Brooks,* C. K. Clark,† G. D. Lester*

*Departments of Large and Small Animal Clinical Sciences, University of Florida, PO Box 100126, Gainesville, FL 32601, USA, †Peterson and Smith Equine Hospital, 4747 SW 60 Ave, Ocala, FL 34474, USA

- 3 groups: Adult horses, sick and well neonatal foals
- Corneal sensitivity:
  - ↓ in sick neonates vs. well neonates & adults
  - Central > peripheral
- STT-1 ↓ in foals vs. adults
How does age and PPID affect corneal sensitivity in horses?

- Horses <10 years of age = 4.75 ± 0.45 cm
- Corneal sensitivity decreased with:
  - Age (> 15 years) = 2.81 ± 0.52 cm
  - PPID (> 15 years) = 2.15 ± 0.37 cm
- No change in STT for the 3 groups
- Effect of treatment for PPID?
Comparison of efficacy and duration of effect on corneal sensitivity among anesthetic agents following ocular administration in clinically normal horses

AJVR, Vol 74, No. 3, March 2013

Jonathan D. Puckett, MS, DVM; Rachel A. Allbaugh, MS, DVM; Amy J. Rankin, MS, DVM; Zhining Ou; Nora M. Bello, DVM, PhD

- 60 horses (1 agent), 8 horses (each agent 1 week apart)
- 0.5% proparacaine HCl, 0.5% bupivacaine HCl, 2% lidocaine HCl, 2% mepivacaine HCl
- Duration of anesthetic effect:
  - Mepivacaine = 35 min
  - Proparacaine = 35 min (max ↓)
  - Lidocaine = 45 min
  - Bupivacaine = 60 min (max ↓)
Effects of action of proparacaine and tetracaine topical ophthalmic formulations on corneal sensitivity in horses

8 horses (each eye assigned - 2/4 agents 48 h apart)

- 0.5% proparacaine HCl, 0.5% aqueous tetracaine HCl, 0.5% viscous tetracaine HCl, 0.9% NaCl (control)

- Max effect of 3 txs: 10 min

- Duration of max effect:
  - Proparacaine: 20 min
  - Aqueous tetracaine: 20 min
  - Viscous tetracaine: 30 min
Live cell imaging of the cornea

Confocal

Specular
Confocal microscopy

- Pinhole/screen aperture for viewing-eliminates scatter from rest of tissue-very sharp image
- Pinhole conjugate to focal point of lens = confocal pinhole = confocal microscope

http://www.physics.emory.edu/~weeks/confocal/
Confocal microscopy

- Laser - provide high intensity
- Mirrors - scan across sample
- Light focused on pinhole
  - measured by detector

http://www.physics.emory.edu/~weeks/confocal/
In Vivo Confocal Microscopy (IVCM)

- Acquire series of thin optical sections
- 3-D reconstruction
- All corneal layers
  - Better detail vs. specular
Anterior epithelium

(1000x Arndt, C., doct. thesis. LMU-München, 1999)

Pictures courtesy C Kafarnik
Stroma and endothelium

Pictures courtesy C Kafarnik
YEEEEAAAAAH

I'M GONNA NEED YOU TO BRING MORE DATA
In vivo confocal microscopy for the detection of canine fungal keratitis and monitoring of therapeutic response

Eric C. Ledbetter,* Mary L. Norman* and Jennifer K. Starr†

*College of Veterinary Medicine, Department of Clinical Sciences, Cornell University, Ithaca, NY 14853, USA; and †College of Veterinary Medicine, Department of Population Medicine and Diagnostic Sciences, Cornell University, Ithaca, NY 14853, USA

Fusarium incarnatum-equiseti

Candida albicans

Filamentous Fungi

Yeast
Corneal innervation in mesocephalic and brachycephalic dogs and cats: assessment using *in vivo* confocal microscopy

Christiane Kafarnik,* Jens Fritsche† and Sven Reese*

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<table>
<thead>
<tr>
<th>Animals</th>
<th>Mean NFD ± SD (mm/mm²)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>subepithelial</td>
</tr>
<tr>
<td>Domestic Short-haired cats (<em>n = 9</em>)</td>
<td>15.49 ± 2.7*</td>
</tr>
<tr>
<td>Mesocephalic dogs (<em>n = 9</em>)</td>
<td>12.39 ± 5.25</td>
</tr>
<tr>
<td>Brachycephalic dogs (<em>n = 7</em>)</td>
<td>10.34 ± 4.71</td>
</tr>
<tr>
<td>Persian cats (<em>n = 6</em>)</td>
<td>9.50 ± 2.3*</td>
</tr>
</tbody>
</table>

Nerves!

(a) Subepithelial – GSD
(b) Subbasal – GSD
(c) Subbasal – DSH
(d) Subbasal – Persian
Can you do IVCM in horses?

Heidelberg HRT Powercheck Cornea with Rostock Corneal Module
In vivo confocal microscopy of the normal equine cornea and limbus

Eric C. Ledbetter* and Janet M. Scarlett†

*Department of Clinical Sciences, College of Veterinary Medicine, Cornell University, Ithaca, NY 14853, USA; †Department of Population Medicine and Diagnostic Sciences, College of Veterinary Medicine, Cornell University, Ithaca, NY 14853, USA
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In vivo confocal microscopy of equine fungal keratitis

Eric C. Ledbetter,* Nita L. Irby* and Sung G. Kim†

*Department of Clinical Sciences, College of Veterinary Medicine, Cornell University, Ithaca, NY 14853, USA; and †Department of Animal Science, College of Agriculture and Life Sciences, Cornell University, Ithaca, NY 14853, USA
In vivo confocal microscopy of brachycephalic dogs with and without superficial corneal pigment

Lucien V. Vallone,* Andrew M. Enders,* Hussni O. Mohammed† and Eric C. Ledbetter*†

*Department of Clinical Sciences, College of Veterinary Medicine, Cornell University, Ithaca, NY 14853, USA; and †Department of Population Medicine and Diagnostic Sciences, College of Veterinary Medicine, Cornell University, Ithaca, NY 14853, USA

Figure 2. In vivo confocal microscopic photomicrographs of brachycephalic dogs with superficial corneal pigment: (a) pigment migration through the superficial–intermediate epithelium; (b) leukocytes within the superficial–intermediate epithelium; (c) pigment migration through basal epithelium; (d) leukocytes within basal epithelium; (e) Langerhans cells within basal epithelium; (f) epithelial disorganization; (g) anterior stromal dendritic cells; (h) vascularization; (i) blended pathology including pigment and leukocytes within the basal epithelium; (j) blended pathology showing pigment and Langerhans cells within basal epithelium. Bars = 50 μm.
In vivo confocal microscopy of brachycephalic dogs with and without superficial corneal pigment

Lucien V. Vallone,* Andrew M. Enders,* Hussni O. Mohammed† and Eric C. Ledbetter*†

*Department of Clinical Sciences, College of Veterinary Medicine, Cornell University, Ithaca, NY 14853, USA; and †Department of Population Medicine and Diagnostic Sciences, College of Veterinary Medicine, Cornell University, Ithaca, NY 14853, USA
What about corneal endothelial dystrophy?
In Vivo Imaging of Corneal Endothelial Dystrophy in Boston Terriers: A Spontaneous, Canine Model for Fuchs’ Endothelial Corneal Dystrophy


Sara M. Thomasy,¹ Dennis E. Cortes,¹,* Alyssa L. Hoehn,¹ Allison C. Calderon,¹ Jennifer Y. Li,² and Christopher J. Murphy¹,²

Control (n=15)  
Affected (n=16)
Phenotypic Characterization of Corneal Endothelial Dystrophy in German Shorthaired and Wirehaired Pointers Using In Vivo Advanced Corneal Imaging and Histopathology


Olivia R. Shull, DVM,* Christopher M. Reilly, DVM, MAS, DACVP,† Lola B. Davis,* Christopher J. Murphy, DVM, PhD, DACVO,* † and Sara M. Thomasy, DVM, PhD, DACVO*†
ROCK inhibitors & corneal endothelium

- ↑ Proliferation
- ↑ Adhesion
- ↓ Apoptosis

Enhancement of corneal endothelium wound healing by Rho-associated kinase (ROCK) inhibitor eye drops

Naoki Okumura, Noriko Koizumi, Morio Ueno, Yuji Sakamoto, Hiroaki Takahashi, Kana Hirata, Ryuzo Torii, Junji Hamuro, Shigeru Kinoshita
IVCM can be used to assess the efficacy of ROCK inhibitors in dogs.

Figure 1.

A

B

C

D

Baseline  Day 0  Day 28

Cryo + PBS

Cryo + Y-27632

Endothelial cell density (cells/mm²)

PBS

Y-27632

Endothelial cell density (cells/mm²)

Cryo+PBS

Cryo+Y-27632
Specular microscopy

• Specular reflection = mirror like
  – Angle of refraction = angle of incidence
  – Microscope captures reflected light

• Interfaces-2 media of different refractive indices
  – Reflection of some light
    • Difference in refractive indices related to amount of reflection
Specular microscopy

- Optical interface between corneal endothelium and aqueous humor
  - Also: corneal epithelium, stroma, lens
- Equipment: contact or non-contact
- Stationary slit, moving slit, moving spot

from Krachmer, Cornea 2005
Specular microscopy

- Four zones of reflection:
  - Zone 1-interface lens, coupling fluid, epithelium
  - Zone 2-light reflected from stroma
  - Zone 3-endothelial reflection
  - Zone 4-aqueous (very little reflection-dark)
Specular microscopy

- Dark boundary - between zone 3 & 4
- Bright boundary - between zone 2 & 3
- Particularly noticeable - slit images

http://www.djo.harvard.edu
Specular microscopy

• Increasing angle of incidence - wider slit, larger image area
  – ↓ image quality - increased illumination and increased scatter (stroma and epithelium)
  – Shortening of endothelial cells

• Modern solution:
  – Small slits/spot-scan over tissue - ↑ field of view
Specular microscopes

- Difficult in awake veterinary patients
- Patients typically fixate (no eye movement!)
- Many units are automated
  - Difficult to align and prevent movement

Figure 1 - Photographic images of non-contact specular microscopy (SP-3000P® specular microscope – Topcon, Japan). (A) Canine patient positioned for image capture. (B) Equipment screen showing the frontal image of patient’s eye immediately before image capture. Ophthalmology Unit, FCAV/UNESP, Jaboticabal, SP, Brazil, 2012.
Specular microscopes

- Konan NonCon Robot - very automated
  - Chin rest
  - Fixate
  - Machine aligns by Purkinje images
  - Focuses on endothelium
  - Picture acquired
Specular microscopy

- Density
- Polymegathism

Cornea Endothelium

- Normal Endothelium
  - High Cell Density

- Very Low Density
  - High Surgical Risk

- Polymegethism
  - EW Contact Lenses

- Stage 3 Guttata
  - Normal Cell Count

Konan CellChek™ Specular Microscope Imaging
Specular microscopy

• Analysis:
  – Endothelial cell density (cells/mm$^2$)
  – Pleomorphism (% of 3, 4, 5, 6, 7, 8 sided cells)
  – Cell area ($\mu$m$^2$)
  – Often automated counts

Figure 1: Specular photomicrograph from Topcon SP-3000P specular microscope. N: number of cells; T: central corneal thickness; MIN, MAX, AVG: minimum, maximum, and average size of cell area; CD: cell density; SD: standard deviation, CV: coefficient of value; HEX: hexagonal cell ratio.
Test:
1. When did the Pilgrims land at Plymouth Rock?

1620.

As you can see, I’ve memorized this utterly useless fact long enough to pass a test question. I now intend to forget it forever. You’ve taught me nothing except how to cynically manipulate the system. Congratulations.

They say the satisfaction of teaching makes up for the lousy pay.

- Contact specular – central & peripheral cornea
  - 59 normal dogs (6 weeks to 132 months)
  - Cell # did not differ - central vs peripheral
  - Cell # decreased with age (cells/mm$^2$)
    - < 1 year -> 2600
    - 1-9 years - 2300-2500
    - > 10 years - 1900-2100

Fig. 2. Relationship of endothelial cell density to age in the normal dog. Each point represents a single reading from either the central or peripheral cornea of each cornea examined.
• Dogs with mature cataracts – contact specular
  – Pre- and post-surgery measured ECD
  – Phaco (n = 14) - ECD ↓ 22% centrally, 13% peripherally
  – ECCE (n = 7) - ECD ↓ 34% centrally, 31% peripherally
  – Both surgeries: Polymegathism & pleomorphism
Effects of intracameral injection of tissue plasminogen activator on corneal endothelium and intraocular pressure in dogs

Gerding PA Jr, Essex-Sorlie D, Yack R, Vasaune S


- 16 healthy dogs – contact wide-field specular
  - 1 eye received TPA at 25 or 50 µg/100 µl (n = 8/group)
  - Contralateral eye = untreated control
  - High dose group - 18% decrease in % of hexagonal cells
  - No other Δ in cell density, morphology or thickness
How does cataract status affect endothelial density?

- Endothelial cell density (ECD), hexagonality, IOP & [PGE₂]
- 3 groups (n = 8/group)
  - Mature cataract
  - Hypermature cataract
  - Control
- No difference in ECD & IOP between groups
- ↓ Hexagonality & ↑ [PGE₂] in cataractous groups vs control
- No difference in any parameter studied in mature vs. hypermature
What about endothelial density in chinchillas?

- Endothelial cell density, average cell area & pleomorphism
- 3 age groups (n = 10/group)
  - 2-4 months: 3423 cells/mm², 351 mm², 70%
  - 48 months: 2650 cells/mm², 442 mm², 65%
  - 10 years: 2124 cells/mm², 584 mm², 63%
Specular microscopy in geriatric primates

<table>
<thead>
<tr>
<th>Age (yrs)</th>
<th>Density (cells/mm²)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Rhesus Macaque</td>
<td>$2328 \pm 1.5$</td>
</tr>
<tr>
<td>Human¹</td>
<td>$2690 \pm 220$</td>
</tr>
</tbody>
</table>

¹Moller-Pedersen, *Cornea* 1987
Pachymetry

• Why measure corneal thickness?
  – Indicator of corneal health
    • ↑ with edema or fibrosis
    • ↓ with stromal loss or refractive sx
  – Diagnosis and monitoring of disease
  – Surgical planning
Methods of measurement

• Historically:
  – Calipers on fresh or fixed tissue
    • Poor reflection of in vivo thickness
    • Tissue swelling after death
    • Fixation contracts corneal tissue
Ultrasonic pachymetry

• 1980’s
• Preferred method:
  – Independent of patient fixation
  – Ease of use
  – Peripheral & central measurement
  – Improved reproducibility
  – Improved accuracy
  – Portability
Ultrasonic pachymetry (USP)

• Modern machines
  – Excellent alignment detection
  – Fast information retrieval and storage
  – Better sampling algorithms

• Smaller probe tips
  – Hard plastic
  – Minimal maintenance
  – Easy to sterilize
  – Better for wider variety of animals
  – More precise measurement
Ultrasonic pachymetry

- Measures anterior PTF to posterior endothelium
- Automatic gain
- Angle sensitivity
  - within 5 degrees of axis
- 20-65 MHz
- Electronic pulse
  - Vibrates piezoelectric crystal
  - Reflects at DM
  - Returns to crystal
  - Electric signal goes to receiver & amplifies signal
Ultrasonic pachymetry

• Distance = velocity x time
• Measures time from end of transducer to DM & back
• Velocity - speed of sound in cornea
  – Human = 1640 m/s
  – Canine = 1577 ± 10 or 1590 m/s at physiologic temps \(^1\)
    • Most machines overestimate corneal thickness in dogs

\(^1\)Tang 2012 OSU dissertation thesis
Ultrasonic Pachymeters

- Other considerations:
  - LCD displays
  - Printing capabilities
  - Audible read-out
  - A-scan US combo
  - Probe size
# Ultrasonic Pachymetry

<table>
<thead>
<tr>
<th>Ultrasonic Pachymeter</th>
<th>Probe (mHz)</th>
<th>Corneal Velocity (m/sec)</th>
<th>Number of Measurements</th>
<th>Range (μm)</th>
<th>Accuracy (μm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Accupach IV</td>
<td>65</td>
<td>1640</td>
<td>Up to 9</td>
<td>300-999</td>
<td>±5</td>
</tr>
<tr>
<td>Pachette 4</td>
<td>20</td>
<td>1640</td>
<td>Up to 25</td>
<td>200-2000</td>
<td>±5</td>
</tr>
</tbody>
</table>
"You think that's bad, I've got test prep anxiety."
Feline CCT = 569 ± 36 μm
- USP with velocity = 1550 m/s
Diurnal variation = 49 ± 15 μm
- ↑ thickness after sleeping
- Corneal swelling following lid closure
Central, superior & temporal peripheral thickness
- 75 normal dogs (150 eyes)
- USP with velocity = 1630 m/s

Mean corneal thickness = 562 ± 6 μm
- Increased with age & weight
- Peripheral > central by ~50 μm
  - Difference increased with age
- Females < males (after adjusting for age & weight)
Central, mid-peripheral & peripheral thickness (4 quadrants)
- 20 normal horses (40 eyes)
- USP with velocity = 1640 m/s

Mean CCT = 793 ± 44 μm
- Peripheral > central
  - Range: 831-924 μm
- No effect: Auriculopalpebral block, xylazine, age, IOP
Corneal thickness, intraocular pressure and optical corneal diameter in Rocky Mountain Horses with cornea globosa or clinically normal corneas
Ramsey DT, Hauptman JG, Petersen-Jones SM.

- Corneal thickness in multiple locations
  - 129 Rocky Mountain horses
  - USP with velocity = 1640 m/s
- ↑ central and temporal (affected vs normal)
- Age & thickness positively correlated
- IOP and optical corneal diameter
  - No difference

Classic Article!
Optical pachymetry

- Attachment to table mounted slit lamp
- Measure oblique section of cornea with split prism
  - Split epithelial/endothelial images aligned by user
- Equations used to determine thickness
- Variables:
  - Refractive index
  - Anterior radius of curvature
- Sources of imprecision
  - Equation variables
  - Inter-observer variability
Optical pachymetry

• Automated systems:
  – Significantly decreases inter-observer variability
  – Increases reproducibility and reliability

Oculus Pentacam
Scheimflug Analysis

- **Oculus Pentacam**
  - Slit illumination system & Scheimpflug camera rotates around eye
  - Sectional image created and photographed in a side view by the camera
  - 3-D model formed from all images in **2 seconds**!
  - ~25,000 data points acquired
Scheimflug Analysis

- Gaililei dual Scheimpflug analyzer
  - Placido disc technology
    - Curvature of central cornea
  - 3D map similar to Pentacam
Anterior Segment
Optical Coherence Tomography (AS-OCT)

• Optical
  – Interference from 2 light waves
• Non-contact
• Assumes same refractive index throughout
• Cross-sectional images

Normal canine cornea
AS-OCT

• Practical Considerations:
  – Does not tolerate ocular movement
    • Sedation/anesthesia may be required
  – Some corneal opacities confound
    • Eg. Sequestra, epibulbar melanoma, dense melanosis
  – $$$
## AS-OCT

<table>
<thead>
<tr>
<th>AS-OCT</th>
<th>AKA</th>
<th>Example</th>
<th>Acquisition Time (A scan/sec)</th>
<th>Axial resolution (µm)</th>
<th>Superluminescent diode (nm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Time-domain</td>
<td>Time of flight</td>
<td>Visante</td>
<td>2048</td>
<td>18</td>
<td>1310</td>
</tr>
<tr>
<td>Fourier-domain</td>
<td>Spectral-domain</td>
<td>RTvue (Optovue)</td>
<td>26000</td>
<td>5</td>
<td>840</td>
</tr>
</tbody>
</table>
How do OCT images differ between TD & FD?

Visante (TD)

RTVue (FD – short lens)
THE IMPOSSIBLE QUIZ!
In vivo evaluation of the cornea and conjunctiva of the normal laboratory beagle using time- and Fourier-domain optical coherence tomography and ultrasound pachymetry

Ann R. Strom,* Dennis E. Cortés,†,‡ Carol A. Rasmussen,§ Sara M. Thomasy,* Kim McIntyre,¶ Shwu-Fei Lee,** Philip H. Kass,†† Mark J. Mannis† and Christopher J. Murphy*,†

• CCT measurements
  – 16 eyes of 8 young intact Beagles - USP & FD-OCT
  – 152 eyes of 125 young intact Beagles (35 F, 90 M) – TD-OCT
  – USP & TD-OCT > FD-OCT
  – CCT ↑ with age and body weight
  – Using TD-OCT, M > F
What about the snake spectacle & OCT?
Spectral domain optical coherence tomography imaging of spectacular ecdysis in the royal python (Python regius)

Charlotte A. Tusler,* David J. Maggs,* Philip H. Kass,† Joanne R. Paul-Murphy,‡ Ivan R. Schwab§ and Christopher J. Murphy*§
Spectral domain optical coherence tomography imaging of spectacular ecdysis in the royal python (*Python regius*)

Charlotte A. Tusler,* David J. Maggs,* Philip H. Kass,† Joanne R. Paul-Murphy,‡ Ivan R. Schwab§ and Christopher J. Murphy*§

(a) Cuticular zone overlies hyperreflective stromal layer
(b) Small, reflective foci in subcuticular space
(c) Spectacular cuticle separated from hyperreflective stroma
(d) Spectacle & cornea have uniform reflectivity
Clinical utility of OCT?

Epithelial/subepithelial Lesions

Famose Vet Ophthalmol
2014;17:12-22
Clinical utility of OCT?

Infectious and stromal keratitis

Famose Vet Ophthalmol 2014;17:12-22
Clinical utility of OCT?

Corneal foreign bodies

Famose Vet Ophthalmol 2014;17:12-22
What about surgical planning?

Superficial keratectomy with conjunctival advancement hood flap (SKCAHF)
Superficial Keratectomy and Conjunctival Advancement Hood Flap (SKCAHF) for the Management of Bullous Keratopathy: Validation in Dogs With Spontaneous Disease

Taemi Horikawa, DVM, DACVO,* Sara M. Thomasy, DVM, PhD, DACVO,† Amelia A. Stanley,† Allison S. Calderon, BS,† Jennifer Li, MD,† Lana L. Linton, DVM, DACVO,* and Christopher J. Murphy, DVM, PhD, DACVO††
Specular microscopy

- Optical pachymetry
- Use electromechanical device
- Central corneal thickness
  - Newer models: mid-peripheral
Specular microscopy

• Measure from posterior surface of tear film to posterior surface of Descemet’s membrane
  – May have error of 20-30 μm

• Contact
  – Corneal touch → compressive → thinner measurement

• Non-contact
  – Lower readings vs USP
Does transcorneal iridal photocoagulation affect corneal thickness?

- Corneal thickness & endothelial density - post diode TCIP
  - 16 young mixed breed dogs prior to & 3 weeks later
  - Non-contact specular microscope
- Central and temporal cornea
  - No significant $\Delta$ in corneal thickness/endothelial density
Confocal microscopy

- Recently: good repeatability
- Measure corneal epithelial thickness
- Bowman’s layer (in humans)
High frequency ultrasound

- Typically 50 MHz or higher
- Intraobserver reproducibility - high
- Interobserver reproducibility - lower
  - Observer perception of landmarks

Much more from Dr. Bentley soon!

Normal Beagle

Corneal sequestrum in a cat
So which pachymetric method is best?
Pachymetry in the human literature

• USP vs optical pachymetry
  – Less reproducible & reliable
    • Between and within observers
  – USP
    • High intraobserver reproducibility
    • Lower interobserver reproducibility (better than optical)
  – Optical pachymeters – Systemic left eye bias
    • Central left eye thicker readings
Pachymetry in the human literature

- USP vs UBM vs Specular
  - USP & UBM less variable than optical (specular)
  - USP
    - Will not read without proper probe placement
    - Average of multiple readings (assess error)
  - UBM
    - Centrality and perpendicularity can be assessed
  - Some optical devices (eg. specular)
    - These criteria cannot be assessed
Central corneal thickness measurements in normal dogs: a comparison between ultrasound pachymetry and optical coherence tomography

Anthony F. Alario and Christopher G. Pirie

- **CCT**
  - 15 healthy dogs (mean age = 2.3 yrs)
  - USP (PacScan 300 AP) vs FD-OCT (Optovue)
    - 6 mm automated pachymetry scan used
    - Mean USP = 599 ± 32 μm vs FD-OCT = 588 ± 32 μm
      - USP consistently overestimates by ~11 μm due to velocity (1630 m/s)
      - High intraclass correlation (0.92)
    - No difference due to age or gender
Reliability of manual measurements of corneal thickness obtained from healthy canine eyes using spectral-domain optical coherence tomography (SD-OCT)

Anthony F. Alario, Christopher G. Pirie


• 3 measurements: CCT, ET & NET
  – 20 healthy dogs (mean age = 4.7 yrs)
  – Manual caliper measurements by 2 observers
  – No difference between eyes or due to age or gender
  – No difference in replicate measurements (same observer)
  – Small significant difference between observers (ET)
Sources of error

• Instrument itself
• Repeated measurements
  – < 1.5% variability
  – Blink between measurements to ↓ variability
• Drying of cornea
• Patient positioning
  – Supine ↓ corneal thickness
• Marking of cornea