Three-year outcomes of corneal crosslinking in progressive keratoconus

375 Corneal Imaging, Topography, and Keratoconus

Tuesday, May 09, 2017 3:45 PM–5:30 PM

Exhibit/Poster Hall  Poster Session

Program #/Board # Range: 3499–3558/A0352–A0411

Organizing Section: Ophthalmology

Program Number: 3499 Poster Board Number: A0352

Presentation Time: 3:45 PM–5:30 PM

Keratoconus and cross linking: Analysis of 90 cases over 3 years

Julie FRANCOIS1, 2, SOT Maxime3, Nadia ouamara2, Christophe Goetz1, Marie-Soline LUC1, 2, Jean-Marc Perone1,

1Ophthalmology, Metz-Thionville Regional Hospital Center, Mercy Hospital, Metz, France; 2Lorraine University, Nancy, France; 
3Clinical Research Support Unit, Metz-Thionville Regional Hospital Center, Mercy Hospital, Metz, France.

Purpose: The aim of our study is to analyze the correlation between higher order aberrations (HOA) of posterior corneal and visual acuity after crosslinking treatment (CXL) in advanced keratoconus

Methods: We performed an observational retrospective study including patients treated with CXL from March 2014 to April 2016. All patients received the same procedure: accelerated-CXL protocol (30 minutes of riboflavin impregnation and 10 minutes of UVA irradiation). The flow data were assessed preoperatively and at 6 months post CXL: best corrected visual acuity, Kmax, and HOA RMS at 3, 5 and 6 mm. Included patients were splitted into 2 groups; according to their delta visual acuity: in the first group patients with decreased or stabilized visual acuity at 6 months post CXL, in the second group patients with an improvement of visual acuity. The primary endpoint was the delta RMS at 5 mm pupillary diameter. The secondary endpoints were the delta RMS at 3 and 6 mm and the delta Kmax.

Results: Eighty patients (90 eyes) were included: 50 in the decreased or stabilized visual acuity group, 40 in the improved visual acuity group at 6 months. The mean age of the patients was 26 +/- 10 years. The mean visual acuity was 0.33 +/- 0.20 logMAR before CXL and 0.29 +/- 0.19 logMAR post-CXL. The mean RMS at 5 mm was 2.67 +/- 1.3 D eq and 2.71 +/- 1.24 D eq respectively before and after CXL. The delta RMS at 5 mm was 0.01 +/- 0.69 in the improvement visual acuity group and 0.05 +/- 0.64 in the decreased or stabilized visual acuity group. There was no correlation between the visual acuity difference and the HOA (student test p = 0.76). The mean Kmax was 57.08 D and 57.37 D respectively in pre and post-operative.

Conclusions: The analysis of the 2 groups: decrease or stabilization versus improvement of visual acuity does not therefore reveal any significant difference in the evolution of HOA RMS. It seems therefore that the improvement of the visual acuity post crosslinking is not related to a reduction of the HOA.

Commercial Relationships: Marie-Soline LUC, None;
SOT Maxime, None; Julie Francois, None; Christophe Goetz, None; Nadia ouamara, None; Jean-Marc Perone, None
accordance with the Dresden protocol. Briefly, the corneal epithelium was removed and riboflavin eye drops were applied for 30 min. After confirming riboflavin penetration through the entire thickness of the cornea, ultraviolet irradiation (3 mW/cm², Opto Xlink) was administered. Isotonic or hypotonic riboflavin ophthalmic solution (Opto Ribolin) was administered in accordance with intraoperative corneal thickness. Kaplan–Meier analysis was performed to assess the effect corneal crosslinking on arresting disease progression. Surgery was considered to have failed at average K values > 1 D.

**Results:** The mean uncorrected visual acuity at 3 years post-surgery (logMAR[decimal], 0.35 ± 0.27[0.45]) was significantly better compared to the preoperative value (0.56 ± 0.38 [0.24]). There were no significant differences in corrected visual acuity between baseline and 3 years post-surgery (p = 0.20). While the mean K value had significantly decreased between baseline and 3 years post-surgery (46.8 ± 3.9 D and 46.2 ± 4.4 D, respectively; p = 0.018), the mean corneal endothelial cell density did not exhibit a significant change (2836 ± 372/mm² and 2725 ± 380/mm²; p = 0.32). Two eyes exhibited an increase in average K value of > 1 D during the follow-up period. The total success rate was 92%. Three eyes exhibited mild corneal opacities in the deep corneal stroma 3 years post-surgery.

**Conclusions:** Corneal crosslinking is a safe and effective method for arresting the progression of keratoconus. Careful follow-up is required to assess disease progression.

**Commercial Relationships:** Takashi Kojima, None; Tomoaki Nakamura, None; AKENO TAMAOKI, None; Kazuo Ichikawa, None

**Clinical Trial:** UMIN-CTR, R000028778

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**Program Number:** 3502 **Poster Board Number:** A0355

**Presentation Time:** 3:45 PM–5:30 PM

**Ten years of Specular Microscopy performed in a Cornea Service**

**Fernando C. Abib**

**1** Anatomy, Federal University of Parana, Curitiba, Brazil; **2** Clinica de Olhos Prof. Dr. Fernando Abib, Curitiba, Brazil.

**Purpose:** The choice of the best Corneal Specular Microscope (CSMe) type is a difficult task and it is decisive to the objectivity of the examination. Non-Contact (NC) CSMe acquire endothelial mosaic (EM) images when the patient sets the target light, each model of this type has different fixed positions. Contact (C) CSM can acquire EM images of any area of the endothelial surface by sliding the objective lens on the cornea. The correct choice, NC or C CSMe, is decisive to the objectivity of the Specular Microscopy (SM). The purpose is to describe the profile of the patients, the used CSMe to perform the SM, the SM results considering an age match control, and the reliability indexes of the performed SM.

**Methods:** Cross-sectional study with database of patients submitted to SM at the Cornea Service of the Clinica de Olhos Prof. Dr. Fernando Abib, Curitiba, Brazil from 12/17/2007 to 11/22/2016. The choice of the used CSMe was as recommended according the autor, ARVO 2015 (Figure 1A). The used microscopes: 1. Non-Contact CSMe CSO, Italy; 2. Contact CSMe BioOptics Bambi 2500, USA. The Cells AnalyzerUSA Patent - Corneal Statistical Endothelial Lab software was used to guide the SM utilizing the Reliability Indexes described by the author in the Cornea Journal 2013, perform the analysis of the results through age control (Figure 1B). The results of SM (Endothelial cell density - ECD, average cellular area - Area, Ceficient of variation - CV and hexagonal percentage - Hex) are classified considering the normal values to the age: IN if inside of the normal range, and OUT if outside of the normal range. The results are presented by descriptive statistics.

**Results:** The total examined patients was 4,899. Male 2803 (57%) and female 2096 (43%). The range of the age was 7 to 98, the average

49±19 years old. The used CSMe: Non-Contact in 2,985 (61%) patients and Contact in 1,914 (39%) patients. The SM was performed in both eyes in 4,498 (98%) patients and in one eye only in 106 (2%). The total number of the performed SM 9,102, distribution per decade of life, the reliability indexes (sample error) and the analysis of the SM results considering the age match control is shown in the table.

**Conclusions:** The patients age were from 2nd to 9th decades of life, the most used was the NC CSM but the C CSM was frequently used. A large number of SM was considered not normal relatively to the age that demonstrate how important is this endothelial examination.

**Figure 1A - Methodology to choose the type of the Corneal Specular Microscope**

Using the ocular pathology knowledge, directed anamnesis and slit lamp examination its possible to classify the corneal endothelium mosaic pattern in three different patterns: 1. Regular Pattern; 2. Irregular Pattern; 3. Chameleon Pattern.

**REGULAR PATTERN:** This endothelium mosaic has only cells with similar morphology without other attached structures. This pattern is represented by the normal corneal endothelium. It's possible to use Contact or Non-Contact CSM.

**IRREGULAR PATTERN:** This endothelium mosaic always has attached structures, they need to be searched and they always to be found. They do the diagnosis: Cornea guttata, Fuchs Endothelial Dystrophy, Polimorphous Posterior Oedhym, ICE Syndrome, and others. You must to use the Contact CSM.

**CHAMELEON PATTERN:** The endothelium mosaic may or not has a specific characteristic or attached structure, its presence depends of stage of the disease. Some disease stages presents the specific characteristic or attached structure, other stages not. They need to be searched thoroughly: Corneal endothelium of contact lenses wewers, after corneal transplantation, ocular trauma, and others. You must to use the Contact CSM.

**Figure 2B - Algorithm of the CELLS ANALYZER method to use the Reliability Indexes of the Corneal Specular Microscopy and the analysis of the results using the Statistical-Analytical Rulers:**

The not normal values are between the blue bar and the red gradient.
The results of the Specular Microscopy during 10 years per degree of life

<table>
<thead>
<tr>
<th>Decade</th>
<th>Patient Number</th>
<th>CXL Number</th>
<th>Raster of Image Number</th>
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<th>Area B</th>
<th>Area C</th>
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Program Number: 3503 Poster Board Number: A0356

Presentation Time: 3:45 PM–5:30 PM

To assess the 1-year corneal topography and aberrometry changes one-year after transepithelial corneal crosslinking using iontophoresis versus standard corneal crosslinking (T-ionto CL) versus standard crosslinking (standard CL) for the treatment of progressive keratoconus.

Methods: Thirty-four eyes of 25 participants were randomized into T-ionto CL (22 eyes) or standard CL (12 eyes) treatment. T-ionto CL was performed by using an iontophoresis device with dexamethan-free 0.1% riboflavin solution and by irradiating the cornea with 10 mW/cm² UV-A device for 9 minutes. Standard CL was performed according to the Dresden protocol. Participants underwent ophthalmic examinations preoperatively and 3-days, 1-week, 1-, 3-, 6- and 12-months after treatment. Changes of corneal shape and optical performance were assessed by analysis of the maximum simulated keratometry (K_max), central corneal thickness (CCT) and corneal high-order wavefront aberration (from 3rd to 7th orders). Other outcome measures included corrected distance visual acuity (CDVA) and manifest refraction.

Results: Twelve months after T-ionto CL and standard CL, K_max averagely flattened by 0.52±1.30 D (P=0.06) and 0.82±1.20 D (P=0.04) respectively. No significant average changes were found in corneal high-order wavefront aberration after T-ionto (from 1.94±0.91 μm to 2.00±0.79 μm; P=0.56) and Standard CL (from 2.24±0.77 μm to 2.12±0.85 μm; P=0.16) respectively. The average CCT did not significantly change at 12 months from preoperatively in either group (T-ionto CL: +9 μm; P=0.07; Standard CL: -1 μm; P=0.80). The mean change in CDVA was -0.10±0.12 logMAR (P=0.003) and -0.02±0.06 logMAR (P=0.10) after T-ionto CL and standard CL respectively. The manifest spherical equivalent refraction changed averagely by +0.71±1.44 D (P=0.03) and +0.21±0.76 D (P=0.38) respectively. No statistically significant differences were found in the outcome measures between treatments.

Conclusions: At 1 year, there was an average improvement in corneal shape, visual and refractive outcomes after T-ionto CL, though average K_max flattening was lower than standard CL. No changes were found in the optical performance of the cornea after the operation.

Commercial Relationships: sebastiano serrao, None; Giuseppe Lombardo, None; Daniela Giannini, None; Marco Lombardo, None

Support: National and Innovation PON (grant n. 01_00110)

Clinical Trial: NCT02117999

Program Number: 3503 Poster Board Number: A0358

Presentation Time: 3:45 PM–5:30 PM

Long-term follow-up of Corneal Collagen Crosslinking for Pellucid Marginal Degeneration


Purpose: Pellucid marginal degeneration (PMD) is a rare ectatic disorder with steepening and thinning in the peripheral cornea which may also, as keratoconus, benefit from corneal collagen crosslinking (CXL). This entity has been evaluated much less than keratoconus especially with regard to CXL treatment. We performed a retrospective, observational clinical study to learn about the clinical and topographic changes after CXL in PMD, and to determine safety and effectiveness of this treatment option to prevent disease progression.

Methods: Clinical charts from year 2008 to 2016 of subjects diagnosed with PMD who were treated with CXL were reviewed. Nineteen eyes of 13 patients (8 women) were included, mean age was 33.78 years (range 14 to 46). Visual acuity (VA), mean keratometies (KM), corneal astigmatism, intraocular pressure (IOP), spherical equivalent (SE) and pachymetry were compared preoperatively and

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postoperatively with a mean follow-up of 13.89 months (range 3 to 24 months). Paired t test and STATA 8.0 were used for statistical analysis.

**Results:** No statistical significant differences were found in mean keratometries from $47.71 \pm 3.72$ to $47.51 \pm 4.07$ diopters ($p=0.544$), corneal astigmatism from $4.2 \pm 2.49$ to $4.23 \pm 2.03$ diopters ($p=0.995$), IOP from $13.66 \pm 1.58$ to $12.77 \pm 1.85$ mmHg ($p=0.175$), SE from $-9.53 \pm 7.67$ to $-4.82 \pm 3.64$ diopters ($p=0.141$) and pachymetry from $425.68 \pm 24.51$ to $434.43 \pm 27.36$ microns ($p=0.107$).

Statistical significant differences were found in visual acuity from $0.44 \pm 0.40$ to $0.25 \pm 0.26$ logMAR ($p=0.007$).

None of the subjects presented any complication related or not to the procedure.

**Conclusions:** This is a large study with long-term follow-up of subjects with PMD treated with crosslinking. Corneal collagen crosslinking in progressive PMD is a safe and effective long-term treatment for avoiding ectasia progression and also improves visual acuity, hence avoiding other invasive surgical treatments such as corneal transplantation when treated early.

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**Curvature map of a right eye of a 37-year-old-woman before crosslinking.**

**Curvature map of a right eye of the same woman 12 months after crosslinking treatment.**

**Commercial Relationships:** CHRISTIAN DENISSE PINKUS HERRERA, None; ENRIQUE O. GRAU-HERNANDEZ, None; ARTURO J. RAMIREZ-MIRANDA, None; MARIANA LUCILA URDAPILLETA, None; ALEJANDRO NAVAS, None

**Program Number:** 3506  
**Poster Board Number:** A0359  
**Presentation Time:** 3:45 PM–5:30 PM  
**Twelve-months functional results and confocal microscopic corneal features in pediatric and adult progressive keratoconus treated with corneal collagen cross-linking**


**Purpose:** To evaluate the functional and anatomical corneal changes after corneal collagen crosslinking (CXL) for progressive keratoconus (KC) in pediatric and adult population.

**Methods:** In this prospective study, patients with progressive KC underwent epithelium-off CXL. Pre- and 12 months postoperative examinations included: slit lamp examination, corneal epithelial fluorescein staining, best spectacle-corrected visual acuity (BSCVA), anterior segment optical coherence tomography, corneal topography and, in vivo confocal microscopy (IVCM).

**Results:** Sixty-five eyes of 57 patients were included in this study. Twenty (31%) procedures were performed in pediatric patients (age <18); 29 (44%) in women. Preoperatively, pediatric and adult populations did not differ significantly in BSCVA and topographic parameters. At 12-months, a slight but significant improvement in BSCVA was noted in mild cases (stages 1-2) but not in severe cases (stages 3-4). Central corneal thickness remained unchanged. Anterior corneal astigmatism reduction was -0.44 D (95% CI -0.76 to -0.11; $p=0.01$) and -0.60 D (95% CI -1.93 to 0.72 $p>0.05$) respectively in the adult and pediatric populations.

In the severe stages, both corneal apex and surface asymmetry index decreased ($p<0.05$). At 12-months, IVCM showed stromal changes from baseline.

**Conclusions:** CXL for progressive KC is proven to be an effective and safe procedure. Our data show that both the stage of the disease...
Program Number: 3508 Poster Board Number: A0361
Presentation Time: 3:45 PM–5:30 PM
Improving OCT Reproducibility with an Automated Corneal Scan Quality Report

Purpose: The quality of OCT corneal scans affects clinical interpretation and the performance of algorithms that generate corneal maps and other measurements. A scan quality report would help operators of all skill levels quickly determine whether a scan should be repeated.

Methods: The study was performed on data from a prospective single-site study where three Pachymetry scans on each of three CIRRUS™ HD-OCT 5000 (ZEISS, Dublin, CA) devices were acquired on subjects in three groups: Normal Cornea (n=45), Post-LASIK (n=40), and Corneal Pathology (n=37).

After each scan, a quality metric algorithm automatically evaluates the scan and displays an acceptability report on the screen. All scans were saved, but in post-analysis, scans were excluded where the quality report indicated the scan was not acceptable for reasons such as poor scan quality, scan too high/low, vertex off center, or large motion. Poor quality may be caused by blinks, partial blinks, eyelid/eyelash interference, or low contrast. The algorithm uses the dewarped anterior surface segmentation and its confidence value at each segmentation point to detect issues outside an acceptable range. To assess the impact of the algorithm recommendations, the reproducibility of epithelial mapping of all scans was compared with a subset of non-excluded scans.

Results: Table 1 reports the number of total number of scans in each group (Normal, Post-LASIK, and Corneal Pathology), the number of scans in each group recommended for exclusion based on the scan quality algorithm, and the reasons for exclusion. Table 2 demonstrates improvement in reproducibility of epithelial mapping once the scans deemed unacceptable by the algorithm are removed from analysis. The improvement is most significant in corneal pathology subjects and in the inferior subfield for normal corneas.

Conclusions: An automated scan quality report is helpful in improving reproducibility. It could be used to immediately alert the operator to repeat poor scans that would affect clinical interpretation or algorithm performance. Currently, when doctors receive poor scans for review, they must request for the patient to be scanned again, which is inconvenient, time-consuming, and disruptive to all parties. With this aid, workflow efficiency may be improved by increasing the consistent quality of scans by independent operators of all skill levels and decreasing the need to bring patients back for repeat scanning.

Table 1: Number of eyes scanned, scans acquired, and reasons for scan exclusions.

<table>
<thead>
<tr>
<th></th>
<th>Normal Corneas</th>
<th>Post-LASIK</th>
<th>Corneal Pathology</th>
</tr>
</thead>
<tbody>
<tr>
<td>Eyes Scanned</td>
<td>45</td>
<td>40</td>
<td>37</td>
</tr>
<tr>
<td>Scans Acquired</td>
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<td>360</td>
<td>333</td>
</tr>
<tr>
<td>Scans Included</td>
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<td>19</td>
<td>120</td>
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<td>- Poor Scan Quality</td>
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<td>9</td>
</tr>
<tr>
<td>- Scan Too Low</td>
<td>51</td>
<td>10</td>
<td>11</td>
</tr>
<tr>
<td>- Scan Too High</td>
<td>3</td>
<td>8</td>
<td>6</td>
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<tr>
<td>- Corneal Implants</td>
<td>0</td>
<td>0</td>
<td>94</td>
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</tbody>
</table>

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Commercial Relationships: Yingjian Wang, Carl Zeiss Meditec, Inc. (E); Homayoun Bagherinia, Carl Zeiss Meditec, Inc. (E); Patricia Sha, Carl Zeiss Meditec, Inc. (C); Michael H. Chen, Carl Zeiss Meditec, Inc. (C); Mary K. Durbin, Carl Zeiss Meditec, Inc. (E)

Purpose: To study the repeatability of corneal power measured by spectral domain optical coherence tomography (SD-OCT). Corneal power measurements are important for planning intra-ocular lens placements.

Methods: A CIRRUS™ HD-OCT (Carl Zeiss Meditec, Inc., Dublin, CA) system with Anterior Segment Premier Module was used to scan normal eyes, post-LASIK eyes and eyes with corneal pathology, using a scan pattern with 24 radial scans over 9 mm diameter with 1024 samples per B-scan centered at the vertex with an A-scan acquisition rate of 27 kHz. Three repeated scans were acquired. Scans with poor quality were detected by a scan quality algorithm and excluded from the repeatability calculations. The corneal power measurement software calculates the anterior and posterior corneal powers by segmenting the anterior and posterior surfaces followed by robust parabolic fitting in the 3mm diameter central zone. The anterior \((k_a = (n_1-n_0)/R_a)\), posterior \((k_p = (n_2-n_1)/R_p)\), and net \((k = k_a + k_p - D_x k_a x k_p / n_1)\) corneal powers are calculated for each meridian [1]. Overall anterior, posterior and net powers are determined by averaging over selected meridians.

The repeatability standard deviation of the corneal power measurements was evaluated by calculating the variance of all measurements from a single visit and taking the square root of the average of all variances. The coefficient of repeatability was also calculated as the ratio of the repeatability standard deviation to the mean.

Results: The following table shows the repeatability standard deviation (SD) of corneal power measured for the normal, post-LASIK, and pathology (including Keratoconus).

<table>
<thead>
<tr>
<th>Corneal Power</th>
<th>Normal Corneas</th>
<th>Post-LASIK</th>
<th>Corneal Pathology</th>
</tr>
</thead>
<tbody>
<tr>
<td>Reproducibility in central subfield (all scans)</td>
<td>3.64 (2.7%)</td>
<td>1.41 (2.8%)</td>
<td>12.80 (9.7%)</td>
</tr>
<tr>
<td>Reproducibility in central subfield (included scans)</td>
<td>3.48 (2.6%)</td>
<td>1.40 (2.8%)</td>
<td>6.16 (4.7%)</td>
</tr>
<tr>
<td>Reproducibility in inferior subfield (all scans)</td>
<td>7.52 (5.9%)</td>
<td>1.49 (3.3%)</td>
<td>13.28 (10.05%)</td>
</tr>
<tr>
<td>Reproducibility in inferior subfield (included scans)</td>
<td>3.72 (2.9%)</td>
<td>1.50 (3.3%)</td>
<td>7.17 (5.7%)</td>
</tr>
<tr>
<td>Worst case reproducibility (all scans)</td>
<td>Superior subfield</td>
<td>11.79 (9.9%)</td>
<td>Inner nasal subfield</td>
</tr>
<tr>
<td>Worst case reproducibility (included scans)</td>
<td>6.50 (5.6%)</td>
<td>3.22 (6.3%)</td>
<td>6.54 (5.0%)</td>
</tr>
</tbody>
</table>

Conclusions: Our study indicates that a corneal power repeatability SD of less than 0.25 diopter can be achieved for normal and post-LASIK cases. Because SD-OCT directly measures the anterior and posterior surfaces simultaneously when the eye is in the natural and relaxed condition without any external intervention, it may yield more consistent results than standard keratometry in post-LASIK and keratoconic eyes in which the anterior-posterior corneal curvature ratios are altered by surgery or disease.

References:
Assessment of opacities in donor corneas using optical coherence tomography
Narae Ko1, Perry Isaac2, Divakar Gupta1, Michael Tramber2, Anthony N. Kuo1,3. 1.Ophthalmology, Duke University, Durham, NC; 2.Miracles In Sight, Winston-Salem, NC; 3.Biomedical Engineering, Duke University, Durham, NC.

Purpose: A severe imbalance exists in the supply and demand of corneas world-wide. Given the imbalance, it is critical to maximize the utilization of procured corneas. Current standard of care screening of donor corneas is with the slit lamp, which provides limited information to distinguish infectious infiltrates from sterile opacities. Optical coherence tomography (OCT) may be useful as it has better depth resolution and could potentially better characterize corneal opacities. In this pilot study, we sought to characterize donor tissues with opacities and determine relative changes in their thickness.

Methods: 22 donor corneas at Miracles In Sight eye bank were found to have an opacity on slit lamp exam and diverted for research. Using OCT (Leica EnVisu R4300), the donor corneas were scanned within their sealed sterile containers immersed in Optisol GS. The scan protocol consisted of 1000 A scan x 100 radial B scans spanning 10mm diameter. Opacities and other pathologic features were identified in the OCT images, and the relative thickness of opacities in the corneas were assessed using ImageJ.

Results: All 22 corneas had opacities near the inferior limbus on slit lamp exam (Figure A). On the OCT images, all opacities were easily identified by increased reflectivity (Figure B). On OCT, twenty (90.9%) corneas had increased thickness in the area of the opacity (range 2.4-36.8%, mean 14.9%) when compared to adjacent clear cornea, and 2 corneas showed no change in thickness. Seventeen (77.3%) corneas had epithelial defect over the area of opacities. Seventeen (77.3%) corneas had opacities confined to the anterior 50% of the cornea.

Conclusions: Donor corneal thickness is increased optically in the presence of opacities. OCT can be used to assess small changes in corneal thickness that may be difficult to evaluate using slit lamp exam alone. OCT was able to localize the opacity in depth within the cornea. Additional work, such as histopathology studies, are needed to determine the underlying etiology of the opacities and understand their correlation with the observed changes.

Figure A: Slit-lamp photo of the inferior corneal opacity (arrow) B: Corresponding opacity on OCT (arrow) shown as a hyper-reflective lesion sparing posterior lamella

Commercial Relationships: Patricia Sha, Carl Zeiss Meditec, Inc. (C); Homayoun Bagherinia, Carl Zeiss Meditec, Inc. (E); Mary K. Durbin, Carl Zeiss Meditec, Inc. (E)

Program Number: 3511 Poster Board Number: A0364
Presentation Time: 3:45 PM–5:30 PM

Comparison of Ultrasound Pachymetry Versus Optical Coherence Tomography in Precut Descemet’s Stripping Automated Endothelial Keratoplasty
Abigail Gordon, Abhinav Golla, Mitchell Weikert, Li Wang, Sumitra Khandelwal. Lion’s Eye Bank of Texas, Cullen Eye Institute, Department of Ophthalmology, Baylor College of Medicine, Houston, TX.

Purpose: To compare corneal thickness measurements from ultrasound pachymetry technology with optical coherence tomography (OCT) in tissues precut for endothelial keratoplasty
**Methods:** In this prospective study, donor corneas that were suitable for Descemet stripping automated endothelial keratoplasty (DSAEK) were enrolled. Precut corneas were prepared for Descemet stripping automated endothelial keratoplasty by using standard eye bank protocol. Prior to cutting, OCT (RTVue, Optovue, Inc.) was used to measure central and midperipheral (3 mm from center) thickness of donor corneas in the storage chamber. Prior to cutting and epithelial scraping, central and four midperipheral measurements were taken of the cornea via ultrasound pachymetry (PalmScan Pachymeter, MicroMedical Devices). After dissection, the anterior lamellar cap was removed and the measurements were taken again via ultrasound and parallel measurements without the cap were taken using OCT. Differences between measurements were studied using a pairwise t-test and correlation coefficients.

**Results:** The study will enroll 50 donor corneas. Preliminary baseline data of 38 corneas measured with ultrasound pachymetry showed an average central corneal thickness of 465 μm with a standard deviation of 68 μm before DSEK preparation. Preliminary results from 4 corneas showed a difference in mean central corneal thickness of 2.25 μm between ultrasound pachymetry and OCT before cutting and a difference of 8.25 μm in cut tissue. Midperipheral corneal thickness showed a difference in average thickness of 38.56 μm between OCT and ultrasound pachymetry. Central corneal measurements of DSEK tissue thickness taken via OCT were positively correlated with ultrasound measurements (r=.88), and mid-peripheral corneal measurements of DSEK tissue were also positively correlated with OCT and ultrasound (r=.19).

**Conclusions:** Preliminary results suggest that OCT and ultrasound measurements of central corneal thickness are highly correlated, while measurements taken at the mid-periphery of the tissue are not well correlated. Additional donor corneas are currently being enrolled and final results and conclusions will be presented. Comparison of OCT anterior segment precut DSEK tissue analysis will be discussed.

**Commercial Relationships:** Abigail Gordon, None; Abhinav Golla, None; Mitchell Weikert, Ziemer (C); Li Wang, Ziemer (R); Sumitra Khandelwal, Allergan (C), Shire (C)

**Support:** Research to Prevent Blindness

**Program Number:** 3513 **Poster Board Number:** A0366

**Presentation Time:** 3:45 PM–5:30 PM

**Agreement between Automated and Manual Corneal Thickness Mapping for Pachymetry, Epithelium, and Stroma from iVue SD-OCT and Avanti SD-OCT**

Xiwei Wang, Kelly A. Soules, Yulia Wolfson, Susan Luh, Yi-Sing Hsiao, Ben K. Jang, Qiennuan Zhou. Optovue, Fremont, CO.

**Purpose:** To evaluate the accuracy of automatically produced corneal thickness maps from iVue and Avanti SD-OCT corneal B-scans (Optovue, Inc., Fremont, CA) and their agreement with manual B-scan corneal thickness measurements in normal eyes and eyes with corneal pathologies.

**Methods:** A total of 88 eyes from 88 subjects (17 Normal, 16 contact-lens, 19 dry-eye, 20 post-laser refractive surgery, and 16 keratoconus) were enrolled in IRB approved studies across three study sites. Four eyes’ data were excluded from analysis due to insufficient OCT scan quality: one from iVue and three from Avanti data sets. Manual grading was performed on the 87 qualified iVue and 85 Avanti scans. Corneal pachymetry scans from the same eye were acquired on iVue and Avanti (Fig. 1A and 1B) systems. Thickness maps including corneal pachymetry, epithelial, and stroma measurements were automatically generated in the central 6 mm and 9 mm diameter regions of the cornea on iVue and Avanti, accordingly. The accuracy of the software-generated thickness maps computed from the automatic segmented boundaries (Fig. 1C) was assessed by the agreement with averaged manual measurements (Fig. 1D) from three independent human graders for 17 and 25 zonal thickness parameters on iVue and Avanti, respectively (Fig. 2A and 2B).

**Results:** For all map zones on both systems, the mean of differences between the automatic and manual measurements was less than 0.7 and 1.4 μm for corneal epithelial mapping, less than 1.9 and 5.9 μm for corneal stroma mapping, and less than 2.3 and 4.7 μm for pachymetry mapping on iVue and Avanti systems respectively (Fig. 2, Tables 1 and 2).

**Conclusions:** This study demonstrated the automatically generated corneal maps by iVue and Avanti are accurate, as shown by good agreement with manual measurements.

Figure 1. Pachymetry B-scans with (A) iVue SD-OCT (6 mm diameter) and (B) Avanti SD-OCT (9 mm diameter), (C) automatically segmented anterior epithelial, posterior epithelial and endothelial boundaries, and (D) manual thickness measurements.
Figure 2. The differences between manual measurements and software-generated thickness maps for all zones on iVue 6 mm and Avanti 9 mm cornea scans.

Commercial Relationships: Xingwei Wang, Optovue (E); Kelly A. Soules, Optovue (E); Yulia Wolfson, Optovue (E); Susan Luh, Optovue (E); Yi-Sing Hsiao, Optovue (E); Ben K. Jang, Optovue (E); Qienyuan Zhou, Optovue (E)

Program Number: 3514 Poster Board Number: A0367

Repeatability and Reproducibility of Corneal Epithelial Thickness Mapping with iVue SD-OCT

Kelly A. Soules¹, Linda M. Zangwill², Natalie A. Afshari¹, Robert N. Weinreb², Yulia Wolfson¹, Xingwei Wang¹, Ben K. Jang¹, Qienyuan Zhou¹. ¹Clinical Affairs, Optovue, Fremont, CA; ²Shiley Eye Institute, Department of Ophthalmology, UCSD, La Jolla, CA.

Purpose: Corneal epithelium can be affected by conditions such as contact lens wear, dry eye, laser refractive surgery (LRS) and keratoconus (KCN). iVue SD-OCT provides high-resolution cross-sectional imaging of the cornea and automated segmentation of the pachymetry scan for corneal epithelial boundary. The purpose of this study is to evaluate the performance of the epithelial thickness mapping (ETM) in normal eyes and eyes of patients with corneal conditions.

Methods: Three repeated pachymetry scans were acquired on 3 different iVue SD-OCT devices (Optovue, Fremont, CA) with device-designated operators from consented subjects enrolled in an IRB-approved study. Manual editing for segmentation artifacts was permitted at operator discretion. Epithelial measurements were derived automatically from the ETM software. Repeatability (based on the random error of repeated scans) and reproducibility (including the random error of repeated scans and the instrument/operator variability) were assessed based on 17 sector parameters (see Map below) derived from a 6 mm diameter corneal epithelial thickness map centered on the pupil.

Results: A total of 59 eyes of 59 subjects (12 Normal, 12 Contact Lens, 11 Dry Eye, 12 Post-LRS, and 12 KCN) qualified for final analysis. Seventy one out of 598 total acquired scans (11.9%) were excluded from R&R analysis due to the following scan quality issues: decenteration of the scan, eyelid artifacts, cropped OCT image, low signal and motion artifacts. The percentage of disqualified scans was similar across Normal eyes (10.7%) and Corneal Patients eyes (12.1%). Out of 527 scans qualified for final analysis, 40 (7.6%) scans required manual edits of the segmentation lines. Repeatability and Reproducibility results are similar (see Table below) indicating minimal device/operator variability for both Normal and Corneal Patient groups.

Conclusions: The study showed good corneal epithelial thickness mapping repeatability and reproducibility in normal eyes and eyes of patients with corneal conditions in all map zones.
Comparison of Corneal Topography Performed by Orbscan II and CASIA Cornea/Anterior Segment Swept Source Optical Coherence Tomography

Omar Shakir1, 2, Sandeep Grover1. 1Ophthalmology, Yale University, New Haven, CT; 2Ophthalmology, University of Florida - Jacksonville, Jacksonville, FL.

Purpose: Orbscan II (Bausch & Lomb, Rochester, NY) has been one of the gold standard methods of measuring corneal topography. The advancement of Optical Coherence Tomography (OCT) has led to its use in all parts of the eye, including the anterior segment. Recently introduced CASIA Swept Source OCT (Tomey, Tokyo, Japan) has the ability to evaluate corneal curvature and obtain corneal topography. A comparison of SS-OCT against the gold standard measurement in normal patients for corneal topography is necessary and is the purpose of our study.

Methods: 45 eyes of 25 patients were included in this study. Patients were examined and found to have no clinical evidence of corneal disease other than refractive error between -6.00 diopters and +4.00 diopters. Astigmatism corneas were included. Screening examinations were performed by a single ophthalmologist. Topography was obtained on the same day. The first scan was obtained on the Tomey SS-1000 CASIA and the second scan was performed on the Orbscan II Corneal Topographer. The corneal topography 4 map mode was selected for analysis on the Tomey SS-1000 CASIA because it most closely parallels the 4 map presentation from Orbscan II. Values for steep keratometric value, flat keratometric value, cylinder value, and steep axis of cylinder were compared. Correlation coefficients and two-tailed T-test p-values were compared.

Results: The difference between steep axis on the two machines was found to be 0.87 diopters (p-value 0.27). The difference between flat axis was found to be 0.77 diopters (p-value 0.59). The difference in cylinder was 0.4 diopters (p-value 0.64). And the difference in axis was found to be 14.7 degrees (p-value 0.86). All p-values were insignificant. Correlation coefficient was 0.95, 0.97, 0.93, and 0.86 respectively.

Conclusions: The Tomey CASIA SS-1000 Swept Source OCT can be used for corneal topography analysis in normal patients with similar results as the Orbscan II. Our results indicate that the CASIA SS-1000 can be used reliably in normal patients to obtain corneal topography. Further studies could include the use of Swept Source OCT in pathologic patients such as keratoconus. Also further comparison of normal patients with Oculus Pentacam (Wetzlar, Germany) would be of use since it is considered another gold standard of corneal topography.

Commercial Relationships: Omar Shakir, None; Sandeep Grover, None

Quantitative Analysis of Epithelial and Total Corneal Thickness in Keratoconus using Sub-Micrometer Axial Resolution Optical Coherence Tomography

Kirsten Carter1, Lacey Haines1, Benjamin MacLellan2, Olivia Krafl1, Ahmed Gawish1, Paul Fieguth1, Luigina Sorbara1, Kostadinka K. Bizheva1. 1School of Optometry and Vision Science, University of Waterloo, Milverton, ON, Canada; 2Department of Physics and Astronomy, University of Waterloo, Waterloo, ON, Canada; 3Systems Design Engineering Department, University of Waterloo, Waterloo, ON, Canada.

Purpose: An important application of ultra-high resolution optical coherence tomography (UHR-OCT) is quantifying the thickness of the human cornea in corneal diseases such as keratoconus. The purpose of this prospective cross-sectional research project was to measure with high precision the epithelial and total corneal thickness in keratoconic and healthy subjects by using a sub-micrometer axial resolution OCT system.

Methods: Keratoconic (KC) subjects and healthy controls were recruited from the University of Waterloo School of Optometry and Vision Science Contact Lens Clinic. A total of 38 eyes were imaged from 10 male and 10 female KC subjects aged 41.6 ± 10.9 years (range: 24-55 years) and controls aged 41.4 ± 9.6 years (range:

Commercial Relationships: None; Francesco Paribello, None; Ernesto d’Alloja, None; Maurizio Fossarello, None

Program Number: 3517 Poster Board Number: A0370
Presentation Time: 3:45 PM–5:30 PM

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27-52 years). The subjects were imaged with a research grade OCT system that provided 0.95 μm axial and < 3 μm lateral resolution in corneal tissue and an image acquisition rate of 34,000 A-scans/s. Volumetric OCT images were acquired from a 4mm x 1mm area in the cornea centered at the location of the minimum total corneal thickness as determined by Pentacam® topography. The OCT images were processed and analysed with custom image processing and segmentation algorithms developed by our research group. Epithelial thickness at the thinnest corneal location was also measured. Data was analysed using the Student’s t-test, with Statistica 12®.

**Results:** The mean total corneal thickness was 438 μm ± 38 μm, ranging from 356 μm to 489μm for the KC group, and 503 μm ± 29 μm, ranging from 442 μm to 556 μm for the healthy control group. The difference in the total corneal thickness between KC and control subjects was not significant (p=0.10). The epithelial thickness measured at the thinnest location of the cornea was 48 μm ± 31 μm, ranging from 26 μm to 75μm for the KC subjects, and 54 μm ± 3 μm, ranging from 50 μm to 60 μm for the control group. This difference was significant, p<0.0001.

**Conclusions:** The sub-micrometer resolution OCT system was capable of generating higher resolution images of healthy and KC corneas compared to the most advanced clinically available OCT technology. The higher axial resolution combined with a custom automatic segmentation algorithm allowed for more precise measurement of the epithelial and total corneal thickness, as well as more detailed visualization of the morphological changes in the KC cornea.

**Commercial Relationships:** Kirsten Carter, None; Lacey Haines, None; Benjamin MacLellan, None; Olvera Kralj, None; Ahmed Gawish, None; Paul Fieghuth, None; Luigina Sorbara, None; Kostadinka K. Bizarre, None

**Support:** NSERC 312037, CIHR-CHRP 446387, CIHR-CPG 127791

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Purpose: Current protocols do not quantify riboflavin concentration or cross-linking and typically require removal of the corneal epithelium. This study measures the concentration of stromal riboflavin in vacuum-assisted transepithelial corneal cross-linking using a novel UV-A theranostics device.

**Methods:** Nine human donor corneoscleral tissues with intact epithelium underwent transepithelial corneal cross-linking using a vacuum-assisted device for delivering dextran-free, hypotonic, 0.1% riboflavin with enhancers, and irradiating the cornea at 12 mW/cm² for 9 minutes. The drug delivery device was composed of a small chamber with a perforated contoured surface in contact with the epithelium and a vacuum pump to control chamber air pressure. Seven additional control human donor corneoscleral tissues were de-epithelialized and soaked with 20% dextran-enriched 0.1% riboflavin solution for 30 minutes and then irradiated with 10 mW/cm² for 9 minutes and used as controls. In both cases, the UV-A device (Chromo4Vis) used was based on theranostics technology, which provided real time measurement of the intrastromal concentration of riboflavin during treatment and assisted the user to best tailor corneal cross-linking to each corneal tissue.

**Results:** Successful transepithelial riboflavin deposition with the vacuum-mediated device was achieved in all corneas with an average stromal concentration of 0.009%±0.003% and a maximum of 0.017%. After transepithelial UV-A irradiation, the intrastromal concentration of riboflavin decreased to 0.002%±0.0006%. In the epithelium-off control tissues, the average riboflavin concentration was 0.016% ± 0.003% after soaking; it decreased to 0.004% ± 0.002% after UV-A irradiation of the corneal stroma. In all cases, the stromal riboflavin concentration decreased non-linearly during UV-A irradiation of the cornea.

**Conclusions:** The novel UV-A theranostics device successfully quantified stromal riboflavin concentration and monitored the corneal cross-linking process in real time. The vacuum-assisted device demonstrated transepithelial riboflavin deposition at concentration levels comparable to the control tissues with epithelium removed.

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crosslinked regions can be imaged with collagen autofluorescence (CAF), which is indicative of local stiffening. While this method achieved rapid, localized, and spatially controllable CXL, the required laser power of 800 mW is 20 fold higher than the ANSI limits (46.1 mW) for use in humans. The purpose of this study was to test whether regeneratively amplified 760nm FS laser pulses (5 kHz, ~2 μJ pulse energy) can be used to photoactivate riboflavin within the cornea to induce precise collagen CXL and CAF with a single pulse while remaining under ANSI limits.

**Methods:** The same variable numerical aperture (NA), custom laser scanning delivery system with adjustable focal depth was used as in our previous studies. 800 nm FS pulses from a regenerative amplifier (5 kHz) were tuned to 1520 nm in an optical parametric amplifier (Coherent Inc, Santa Clara, Ca). The 1520 nm laser pulses were then frequency doubled in a custom bismuth triborate (BiB O₃) nonlinear crystal (Newlight Photonics, Ontario Canada) to 760 nm and then aligned into our delivery system. As a proof of concept, rabbit corneas soaked in 0.5% Riboflavin/PBS with dextran (20%) were raster scanned with 0.1 and 0.2 NA, 5 mm/s scan velocity, and 12 mW of average power. CAF was used to detect corneal collagen CXL.

**Results:** CAF was detected for both numerical apertures. Representative CAF images are presented in Figure 1. A single pulse through the 0.1 NA and 0.2 NA delivery produced CXL regions 173 ± 14 μm and 126 ± 14 μm in axial length, respectively. Both NA’s resulted in a lateral width of roughly 3 μm.

**Conclusions:** Corneal collagen CXL is possible with a single amplified 760nm FS laser pulse while remaining well under the 46.1 mW ANSI limit for our system. Given the 5 mm/s scan velocity, the current system deposits 1 high energy pulse per micron per 200 μs, compared to roughly 15,000 low energy pulses using a pulse repetition rate of 76 MHz. This innovation allows for adequate energy density per unit time to induce CXL without exceeding ANSI limits. Future optimization of the optical system will enable nearly 4x the current pulse energy while still respecting ANSI limits.

![Figure 1. CAF image of single FS pulses spaced 50μm apart. A) NA=0.1 B) NA=0.2.](image)

Program Number: 3520 Poster Board Number: A0373
**Presentation Time:** 3:45 PM–5:30 PM
**Assessing the Effects of Pulsed, High Intensity UVA Crosslinking on Corneal Elasticity and Viscosity Using Atomic Force Microscopy**

Wyndham M. Batchelor¹, Vasilios Diakonis³, Nikita Likht¹, Sarah Sutnick¹, Noel Ziebarth¹. ¹Department of Biomedical Engineering, University of Miami, Coral Gables, FL; ²Florida Lions Eye Bank, Bascom Palmer Eye Institute, University of Miami Miller School of Medicine, Miami, FL.

**Purpose:** To determine the effects of pulsed versus non-pulsed, high intensity ultraviolet light (UVA) crosslinking on corneal elasticity and viscosity.

**Methods:** Three pairs of donor human eyes kept in damp gauze storage containers were obtained from the Florida Lions’ Eye Bank for this study. The corneas were excised from the globes beyond the limbus, the epithelium was removed from each cornea with a cotton swab, and the corneas were placed into a vial containing 20% Dextran and stored overnight to restore the corneas to a near physiological hydration state. The corneas were then pre-treated with 20% riboflavin every 5 minutes for a total of 30 minutes. The left corneas (OS) underwent high intensity UVA crosslinking with an intensity of 9mW/cm² for 10 minutes using the Peschke cornea crosslinking system (total surface dose of 5.4J/cm²). The right corneas (OD) underwent the same intensity UVA, but the light was pulsed with a pulse on/pulse off time of 10 seconds and 10 seconds, respectively (same surface dose of 5.4J/cm²). Corneal hydration was maintained throughout the crosslinking by applying riboflavin every 5 minutes. After crosslinking, the corneas were allowed to equilibrate in 15% Dextran for 30 minutes. A microkeratome (Moria Evolution 2) with a 50μm head was then used to expose the anterior stroma in each cornea. An atomic force microscope, custom designed for biomechanical measurements, was used to assess the elasticity and viscosity at the anterior stromal layer of each cornea.

**Results:** For two out of the three pairs of corneas investigated, the corneas that underwent pulsed UVA crosslinking were significantly stiffer than their non-pulsed counterparts (2.33 and 2.86 times greater Young’s modulus). The viscosity was also significantly greater in these pairs (3.85 and 2.69 times greater). The third pair of corneas had comparable stiffness and viscosity for both pulsed and non-pulsed protocols.

**Conclusions:** These preliminary results suggest that high intensity, pulsed crosslinking is more effective than the same UVA intensity applied continuously at increasing the elastic modulus of the cornea; this suggests that pulsing the ultraviolet light could increase the efficacy of accelerated, non-pulsed procedures.

**Commercial Relationships:** Wyndham M. Batchelor, None; Vasilios Diakonis, None; Nikita Likht, None; Sarah Sutnick, None; Noel Ziebarth, None

**Support:** University of Miami Dean’s Fellowship

Program Number: 3521 Poster Board Number: A0374
**Presentation Time:** 3:45 PM–5:30 PM
**Evaluation of corneal symmetry after UV corneal crosslinking for keratoconus**

hahan mofy¹, alzahrani khaled², Fiona Carley³, sophie harper¹, Arun Brahma¹, Debbie Morley¹, M C. Hillary². ¹optometry, King Saud university, Riyadh, Saudi Arabia; ²Division of Pharmacy and Optometry, university of manchester, Manchester, United Kingdom; ³Manchester Royal Eye Hospital, Manchester, United Kingdom.

**Purpose:** To assess UV corneal crosslinking (CXL) treatment for keratoconus by evaluating the corneal regularity through patients’ follow-up using Oculus Pentacam. This could provide relevant
transient corneal haze is associated with the spherical, Retinal, None; Royal Victorian Eye and Ear Hospital, Melbourne, VIC, 3522, None; The Keratoconus module of the Save Sight Registries, None; To report the 12 month outcomes of 194 eyes undergoing corneal cross-linking for keratoconus in an referral center. Ricardo Blas Medina, Arturo J. Ramirez-Miranda, Enrique O. Graue-Hernandez, Jesus Cabral, Adrian Escudero-Rodriguez. Instituto de Oftalmología Conde de Valenciana, Mexico city, Mexico. Purpose: To determine the postoperative visual, keratometric and topographic outcomes of epi-on vs epi-off corneal crosslinking. Methods: Prospective controlled longitudinal non-randomized study. We included 29 eyes of 15 patients with keratoconus diagnosis 73% males, mean age of 18.7 years-old. Twenty eyes underwent epi-off corneal crosslinking (≥400μm at the thinnest point) and 9 patients underwent epi-on corneal crosslinking (≤400μm at the thinnest point). Inclusion criteria were: progressive keratoconus diagnosed at least six months before the study (Progressive keratoconus was defined as an increase of ≥1D in manifest cylinder, an increase of ≥0.50 D in spherical equivalent, an increase of ≥1D in maximum keratometry and/or decreased in central corneal thickness of >5% in the previous six months). Amsler-Krumeich stage of keratoconus, uncorrected distance visual acuity (UDVA), corrected distance visual acuity (CDVA), keratometries, topographical data and pachymetry were considered to evaluate efficacy. Endothelial density (ED) Coefficient of Variation (CV), Hexagonality and IOP were considered to evaluate safety. Student T-test compared the preop measure vs the third month post-op visit. Results: Preoperative mean CDVA was 0.34 ± 0.18 logMAR compared with 0.25 ±0.13 at 3 months postoperatively (p=0.03). There was a significant decrease of mean K-values (p=0.05). No significant changes regarding pachymetry, ECD, VC and percentage of hexagonality were observed. No differences were observed between epi-on vs epi-off crosslinking. Conclusions: Corneal crosslinking is an effective procedure for the treatment of incipient or moderate progressive keratoconus. CDVA, K-values, and pachymetry improved or stabilized at 3 months of treatment compared to preoperative values, despite the used technique. No side effects were detected in the corneal endothelium study at 3 months after treatment.

Commercial Relationships: Ricardo Blas Medina, None; Arturo J. Ramirez-Miranda, None; Enrique O. Graue-Hernandez, None; Jesus Cabral, None; Adrian Escudero-Rodriguez, None

Program Number: 3522 Poster Board Number: A0375
Presentation Time: 3:45 PM–5:30 PM

Save Sight Registries Keratoconus; Tracking the outcomes of corneal cross-linking for Keratoconus from routine clinical practice across Australia and New Zealand
Stephanie L. Watson1,2, Elsie Chan3,4, Mark Daniell5, Yves Kerdraon6,7, John Males8,2, Nigel Morlet2,2, Richard A. Mills9, Daniel Barthelmes, Martina Boesch, Amparo Herrera Bond, Amanda Dinh, Vuong Nguyen, Marco Garcia, Mark C. Gillies.
1Ophthalmology, Save Sight Institute, University of Sydney, Bondi Junction, NSW, Australia; 2Sydney Eye Hospital, Sydney, NSW, Australia; 3Centre for Eye Research Australia, Melbourne, VIC, Australia; 4Royal Victorian Eye and Ear Hospital, Melbourne, VIC, Australia; 5Flinders University, Adelaide, SA, Australia; 6Ophthalmology, University Hospital Zurich, Zurich, Switzerland; 7The University of Western Australia, Perth, WA, Australia; 8Private Practise, Zurich, Switzerland.

Purpose: To report the 12 month outcomes of 194 eyes undergoing corneal cross-linking for keratoconus performed in routine clinical practice across Australia and New Zealand. Methods: Index visit characteristics, such as visual acuity (VA, in Logarithm of the Minimal Angle of Resolution [logMAR] letters), maximum keratometry [Kmax], pachymetry, as well as treatment parameters (epithelial status, riboflavin type, UV duration), outcomes (VA, Kmax, pachymetry) and ocular adverse events were recorded in a prospectively designed electronic database. Index visit characteristics associated with the 12-month VA outcome were identified using mixed effects linear regression. Results: Mean change in VA in the cohort after 12 months was +3.1 logMAR letters (95% confidence interval [CI]: -0.9, 5.3); Kmax +0.1 D (95% CI: -0.3, 0.5) and pachymetry -14.9 microns (95% CI: -21.0, -8.6). There was no significant difference in change in Kmax (p = 0.649); a significant improvement in VA (p = 0.006) and reduction in pachymetry was found at 12 months (p < 0.001). VA at the index visit was the strongest predictor of the 12-month VA outcomes (p < 0.001). Treatments were epithelial-off (n=178) with UV exposure that was accelerated (n= 118) or according to the Dresden protocol (n=60). Adverse events episodes occurred in 15 eyes and included clinically significant haze (n=22), microbial keratitis (n=1), persistent epithelial defect (n=2), scarring (n=6) and sterile infiltrates (n=2) from a total of 194 eyes within the first 12 months of follow-up.

Conclusions: The Keratoconus module of the Save Sight Registries can facilitate the collection of large amounts of data on corneal cross-linking. The finalised 12 month outcome data has shown that corneal cross-linking can stabilize visual acuity and corneal parameters when used in routine clinical practice.

Commercial Relationships: Stephanie L. Watson, None; Elsie Chan, None; Mark Daniell, None; Yves Kerdraon, None; John Males, None; Nigel Morlet, None; Richard A. Mills, None; Daniel Barthelmes, None; Martina Boesch, None;
SMG solution exhibits a dose-dependent bactericidal effect. School of Optometry, NSERC 312037; CIHR-CHRP 446387; CIHR-CPG 127791.

Comparison of corneal epithelial thickness before and after corneal cross-linking surgery with and without intracorneal ring segments using optical coherence tomography

Lacey Haines1, Olivia Kralj1, Sebastian Marschall1, Ahmed Gawish4, Paul Fieguth2, Neera Singal2, Hall Chew3, David Rootman4, Allan Slomovic5, Wendy Hatch4, Kostadinka K. Bizheva1,4, Luigina Sorbara1,2, School of Optometry and Vision Science, University of Waterloo, Waterloo, ON, Canada; 1Dept. of Physics and Astronomy, University of Waterloo, Waterloo, Ontario, Waterloo, ON, Canada; 2Oculis, Wetzlars, Germany; 3Systems Design Engineering Dept., University of Waterloo, Waterloo, ON, Canada; 4Dept. of Ophthalmology and Vision Sciences, University of Toronto, Waterloo, ON, Canada.

Purpose: Corneal cross-linking (CXL) is a surgical treatment option that can decrease the progression of keratoconus (KC) and can be combined with intracorneal ring segment (ICRS) insertion in an attempt to improve refractive outcomes. In vivo imaging of KC patients using optical coherence tomography (OCT) was performed to compare total corneal epithelial thickness before and after CXL and CXL+ICRS surgeries.

Methods: There were 11 KC eyes that underwent CXL and 10 KC eyes that had CXL+ICRS surgery. All eyes were imaged before surgery and 6 and 12 months after surgery. Images were acquired using a research grade swept source OCT system which provided 5um axial, ~15um lateral resolution, and ~4mm scanning depth in corneal tissue. Volumetric images of the KC cornea were acquired at the speed of 100,000 A-scans/sec. The images were analyzed with a novel image processing algorithm that segmented the anterior and posterior surfaces of the cornea, and the posterior epithelium. The epithelial and total thicknesses at the point of minimum total thickness and four adjacent locations were obtained for comparison.

Results: The CXL+ICRS group showed a greater epithelial thickness than the CXL only group (p=0.01) and both groups demonstrated increased epithelial thickness after surgery, though there was only a significant difference between visits 1 and 4 (p=0.004) and not for visits 1 and 3 (p=0.22) or 3 and 4 (p=0.27). There were no significant differences in epithelial thickness between the five adjacent zones used in this study. The CXL+ICRS group also showed greater total corneal thickness measurements than the CXL group (p<0.05). The total corneal thickness increased after surgery for both groups and the difference was only significant for visits 1 and 4 (p<0.05) and not for visits 1 and 3 (p=0.05) or 3 and 4 (p=0.07). There were significant differences in total thickness according to location with the central zone found to be thinner than the superior (p=0.05), inferior (p=0.02), and nasal (p=0.04) zones, but not the temporal zone (p=0.16).

Conclusions: Epithelial and total corneal pachymetry measurements before and after CXL and CXL+ICRS surgery for KC showed an increase in thickness after surgery with the CXL+ICRS group having thicker total and epithelial measurements compared to the CXL only group.

Commercial Relationships: Lacey Haines, None; Olivia Kralj, None; Sebastian Marschall, Oculus (E); Ahmed Gawish, None; Paul Fieguth, None; Neera Singal, None; Hall Chew, None; David Rootman, None; Allan Slomovic, None; Wendy Hatch, None; Kostadinka K. Bizheva, None; Luigina Sorbara, None.

Support: NSERC 312037; CIHR-CHRP 446387; CIHR-CPG 127791.
Sub epithelium curvature: A new method to compare efficacy of surface ablation between manual removal and epi-clear removal of epithelium

Rachana C S1, Mathew Francis1, Natasha Pahuja2, Rushad Shroff2, Rohit Shetty2, Abhijit Sinha Roy1. 1Narayana Nethralaya Foundation, Bangalore, India; 2Refractive and Cornea, Narayana Nethralaya, Bangalore, India.

Purpose: To compare post operative change in topography of sub epithelium surface between manual and Epi-clear removal of epithelium during photorefractive keratectomy(PRK)

Methods: The study included 10 subjects who underwent corneal epithelium removal manually in one eye and using Epi-clear(Orca, Israel) in the other eye before PRK. Subjects were screened for refractive error, topography from Pentacam and optical coherence tomography(OCT) before(pre) and after(post) surgery(between 6-12 weeks). 25 radial scan mode of Pentacam(OCULUS Optikgerate GmbH, Germany) was used. Corneal imaging was performed by OCT(RTVue-100, Optovue Inc., CA) which acquired 8 radial scans from 0° to 180° at an A-scan rate of 26,000 lines per second. Since OCT acquired 6 mm diameter scans only, the Pentacam data was also truncated to 6 mm diameter. The anterior corneal surface(ACS) and sub-epithelium surface(SES) edge were detected in radial scans of OCT and axial curvature was computed. Further, keratometry and wavefront aberrations(Zernike analyses upto order 6) were calculated for Pentacam, ACS OCT and SES OCT with refractive index of 1.3375

Results: In manually scraped eyes, changes(pre-to-post change) in keratometry variables were similar to Epi-clear, e.g., flat axis(K1;p=0.6,0.6,0.8), steep axis(K2;p=0.8,0.8,0.8) and maximum keratometry(p=0.7,0.6,0.4) for Pentacam, ACS OCT and SES OCT, respectively. Also, changes in mean keratometry in Pentacam, ACS OCT and SES OCT was similar (p=0.6,0.9,0.5) in Epi-clear and manually scraped eyes. The astigmatism(K1-K2) was similar between Epi-clear and manually scraped eyes [2.2±0.3D and 1.8±0.2D from Pentacam(p=0.7); 1.9±0.5D and 1.7±0.4D from ACS OCT(p=0.1); 2.2±0.5D and 1.7±0.4D from SES OCT(p=0.9)]. Changes in wavefront aberrations of Epi-clear were similar to manually scraped eyes(p>0.05). Figure 1 shows an example of Pentacam topography, ACS OCT and SES OCT topography in two eyes(one with epi-clear and other with manual scraping) pre and post surgery

Conclusions: The topographic changes after Epi-clear and manual removal of epithelium were similar. However, longitudinal follow-up studies are needed to assess the differential effect of Epi-clear surgery on wound healing and haze relative to manual removal of epithelium. This is the first study to quantify changes in surface topography underneath the epithelium non-invasively before and after surgery

Figure 1 shows the axial curvature of two eyes (one with epi-clear and other with manual scraping) pre and post surgery

Commercial Relationships: Rachana C S, None; Mathew Francis, None; Natasha Pahuja, None; Rushad Shroff, None; Rohit Shetty, Narayana Nethralaya (P); Abhijit Sinha Roy, SIBAC Grant (F), Indo-German Science and Technology centre, Germany (F), Narayana Nethralaya (P)

Support: SIBAC grant, Indo-German Science and Technology centre, Germany

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Automatic grading of corneal sub-basal nerves tortuosities

Fabio Scarpa, Pedro Guimarães, Alfredo Ruggeri. University of Padova, Padova, Italy.

Purpose: The clinical perception of corneal subbasal nerve tortuosity (CNT) was recently shown to have two distinct forms, namely a short-range tortuosity (SRT) and a long-range tortuosity (LRT). We developed a method for the automatic grading of confocal corneal nerve images according to these two forms of CNT.

Methods: CNT as estimated from confocal microscopy is becoming a widely used clinical measure. The clinical perception of CNT was however recently shown to have two distinct forms, characterized by either short-range (high frequency, low amplitude) or long-range (low frequency, high amplitude) directional changes (Lagali et al., IOVS, 2015).

Seven expert graders from different institutions visually assessed 30 confocal microscopy images and ordered them by increasing nerve tortuosity, according to either tortuosity definition (Fig.1). Their average ordered rankings were assumed as GT.

Using the same 30 images, we traced corneal nerves with a custom computerized procedure and computed for each image both SRT and LRT. SRT was obtained by a custom combination of angle-, curve-, and twist-based tortuosity measurements at different scales. LRT was obtained by a custom combination of twist- and curve-based tortuosity measurements only, still at different scales. Measurements and scales used for SRT were in general different from the ones used for LRT, since the former have to detect high frequency and low amplitude directional changes, while the latter have to detect low frequency and high amplitude directional changes.

The correlation with respect to GT of the ordered ranking estimated using either SRT or LRT was then assessed using the Spearman Rank Correlation (SRC) coefficient.

Results: The proposed method for the automatic grading of CNT using either SRT or LRT was able to achieve SRC coefficients of 0.98 and 0.96 with GT, respectively.

Conclusions: The proposed SRT and LRT automatic grades highly correlate to clinical perception of CNT. We now need to investigate to what degree different cornea pathologies exhibit one or the other form in their subbasal nerve patterns.

Figure 1: Corneal nerves image with the highest tortuosity rank for the short-range (top) and the long-range tortuosity (bottom).

Commercial Relationships: Fabio Scarpa, None; Pedro Guimarães, Alfredo Ruggeri, None
estimation of the size of the epithelial defect (ED) and stromal infiltrate (SI) at the slit-lamp, and is augmented by acquiring external photographs of the ulcer. We introduce an automated method to construct quantitative models of the corneal ulcer from these images, which will facilitate more consistent therapeutic monitoring.

**Methods:** Fourteen patients with a corneal ulcer were examined by 4 separate ophthalmologists at the slit lamp and an expert consensus of the size of the ED and SI were recorded. An external photo of the affected eye was also captured. We developed a graphical user interface to facilitate semi-automated segmentation (delineation) of ED and SI from external photography (Fig 1A). The user initializes seed regions in the foreground (ED or SI) and the surrounding background regions (Fig 1B; foreground in blue, background in red). Random forest tissue classification is used to generate a probability map for the foreground image (Fig 1C). A level-set segmentation is subsequently performed, in which the probability map generated by random forest is used to drive active contour evolution to delineate the boundary of the ED or SI (Fig 1D-F). The area of the ED or SI is then automatically computed. Separately, each photograph was manually traced and the Dice similarity coefficient between manual and automated models computed to assess the accuracy of automated segmentation. We also calculated the intra-class correlation coefficient (ICC) to compare the agreement in ED and SI size between automatically measured and expert consensus.

**Results:** The Dice similarity coefficient between manual and automated segmentation of ED and SI were 0.92 ± 0.03 and 0.89 ± 0.05, respectively (N=14, mean ± SD). The ICC comparing agreement in ED and SI size between automated and expert consensus were 0.96 (95% CI [0.89, 0.98]) and 0.82 (95% CI [0.55, 0.93]), respectively.

**Conclusions:** We describe a novel method for semi-automated analysis of corneal ulcers. Automated measurements of ED and SI size are in good agreement with expert consensus and have the potential to enhance the reliability of therapeutic monitoring.

![Image](image-url)

**Figure 1:** A: external photograph of corneal ulcer. B: seed regions. C: probability map. D-E: active contour evolution. F: final segmentation.

**Commercial Relationships:** Tapan Patel, None; N.Venkatesh Prajna, None; Lakshey Dudeja, None; Nita Valikodath, None; Maria A. Woodward, None

**Support:** NIH K23K23EY023596

**Program Number:** 3529 Poster Board Number: A0382

**Presentation Time:** 3:45 PM–5:30 PM

**Automated analysis of in vivo confocal microscopy images of corneal nerves**

Stuti L. Misra1, Jonathan D. Oakley2, Charles N. McGhee1, Ellen F. Wang1, Dipika Patel1, Patrick M. Tarwater1, Joseph L. Mankowski1, 1Department of Ophthalmology, The University of Auckland, Auckland, New Zealand; 2Voxeleron LLC, Pleasanton, CA; 3Department of Biostatistics, UTHSC School of Public Health, The University of Texas, El Paso, TX; 4Department of Molecular and Comparative Pathobiology, Johns Hopkins University, Baltimore, MD.

**Purpose:** In vivo confocal microscopy of the cornea (IVCM) allows for non-invasive acquisition of two-dimensional images at a cellular level, enabling detailed corneal nerve assessment. Manual quantification, however, is time-consuming and subjective. Automating these measures will provide clinicians with objective tools to process images promptly and provide an ‘in-clinic’ report to patients. In this study, performance of our automated approach relative to expert ground truth in normal and diseased corneas is reported.

**Methods:** The algorithm is an adaption for IVCM imaging of the method reported in [Dorsey et al, Am J Pathology, 2014]. Briefly,
denoising is followed by edge enhancement. Processing continues on localized image patches, and in the principal orientation of the nerves. Integrating in these directions reduces the detection task to 1d while improving sensitivity to the low contrast and fragmented appearance of the nerves. Three confocal images of corneal sub-basal nerves from each of 10 patients with diabetes (M=7, F=3; age range 24-69) and 10 control participants (M=5, F=5; age range 30-73) were acquired and processed (N=60). Manual tracings from three expert readers were performed using ImageJ to obtain total nerve length and density. Bland-Altman analysis presents inter-observer reproducibility, and the Pearson coefficient gave the correlation across groups (Stata, StataCorp, College Station, TX).

**Results:** Comparing the average counts across the readers and the algorithm, the Pearson coefficient reported values of 0.706, 0.862 and 0.934 for controls, diabetes and then all cases, respectively. A Bland-Altman plot (Fig 1) shows the total nerve densities readings versus the algorithm. Overall R² correlation to individual readers was 0.89, 0.80 and 0.87 across all images.

**Conclusions:** The results show strong correlations between manual and automated counting methods. Interestingly the lowest correlation was in control cases. Further analysis showed algorithm sensitivity declined at the periphery of images, where illumination was dim. This could be addressed by cropping the image data, more uniform illumination across the images, or by algorithm improvements.

![Bland-Altman plot](image_url)

Bland-Altman plot of nerve fiber density for the average reader score and the algorithm. R² is 0.88, and the limits of agreement are 0.0016 and -0.0124. Diseased cases are shown as stars, controls as circles.

**Commercial Relationships:** None.

**Program Number:** A0384

**Presentation Time:** 3:45 PM–5:30 PM

**Screening of tear molecular markers for early detection of keratoconus by quantitative mass spectrometry**

**Purpose:** To screen and identify changes in tear fluid proteome that can act as diagnostic markers for early detection in patients with progressing grades of keratoconus (KC) by label-free targeted quantitative mass spectrometry

**Methods:** Tear proteins extracted off Schirmer strips collected from patients at grades I-III (n= 33, 25 and 21) of KC and healthy control subjects(n=40) were subjected to proteinolytic digestion prior to differential labelling by iTRAQ. For screening the disease associated changes, only proteins quantifiable in at least half the patients across the clinical grades of KC are considered. Disease specific alterations in the tear proteome were screened by building secondary ratios of proteins between the lower and higher grades of comparison. A ratio fold difference of ≥1.5 and a two-tailed t-test p values≤0.05 between the grades of comparison is considered to be statistically significant. The differential proteins displaying either an elevated or reducing trend in progressing grades of KC were validated in an independent cohort of patients, control(n=28) and grades I-III (n=23, 25 and 13) by parallel reaction monitoring(PRM). Each patient tear fluid sample was spiked with 5fmol/µl of E.coli beta-galactosidase digest as an internal standard

**Results:** iTRAQ analysis identified altered expression of around 200 proteins in increasing grades of KC. Panther pathway analysis showed glycolytic proteins, proteins on Rho GTPase cytoskeletal regulation, p38/MAPK pathway and FGF signaling to be significantly altered in KC patients 35 proteins satisfying the filter criteria of ratio fold ≥1.5 and two-tailed t-test p values≤0.05 are considered as potential markers for progression of keratoconus. Proteins lactritin (LACRT), cystatin-s(CST4), lactoperoxidase(LPO), Ig alpha 1 chain(IGHA1) were identified to be elevated whereas tear proteins lysozyme C(LYSC), lipocalin1(LCN1) were among those with reduced levels in increasing clinical grades of KC. So far nine proteins--LACRT, LYSC, LCN1, DMBT1, SG2A1, ANXA1, 1433Z, CYTN, and IGHAI were validated with atleast 1 peptide/protein by PRM in an independent cohort of 21(total 90) patients(con-3, GI-III 6 each)

**Conclusions:** Using quantitative mass spectrometry, our study identified a panel of molecular markers that help in screening the progression of KC which can aid in early diagnosis and in distinguishing the disease from other ocular surface conditions

**Commercial Relationships:** None.

**Program Number:** 3533 Poster Board Number: A0385

**Presentation Time:** 3:45 PM–5:30 PM

**Qualitative effects of a novel nano-lipid artificial tear on tear lipid dynamics**

Matthew J. Kowalski, Jennifer S. Fogt, Peter E. King-Smith, Joseph T. Barr. College of Optometry, The Ohio State University, Columbus, OH.

**Purpose:** To assess the effects of a novel nano-lipid artificial tear on tear lipid dynamics using The Ohio State University stroboscopic video color microscope (SVMC).

**Methods:** Normal healthy volunteers underwent SVMC imaging of the central cornea pre- and post-instillation of a novel nano-lipid artificial tear. Image field of view was 6 mm in diameter, and was captured at a resolution of 1400x1100, at a rate of 23 frames per second. Colors observed in images were a result of optical interference in the tear lipid layer, and lipid thickness was determined. Image data was qualitatively graded for extent of lipid spreading and fluidity of lipid between blinks.

**Commercial Relationships:** None.

**Program Number:** 3531 Poster Board Number: A0384

**Presentation Time:** 3:45 PM–5:30 PM

**Screening of tear molecular markers for early detection of keratoconus by quantitative mass spectrometry**

Krishnatej Nishtala1, Lei Zhou1, Rohit Shetty2,3, Kiran kumar4, Roger W. Beuerman5, Arkasubhra Ghosh1,3, GROW Research Laboratory, Narayana Nethralaya Foundation, Bangalore, India; 1Narayana Nethralaya Eye Hospital, Bangalore, India; 3Singapore Eye Research Institute, Singapore, Singapore.

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**Results:** SVCM imaging demonstrated tear lipid increase post-instillation of the artificial tear, where baseline images that appeared grayscale were then enhanced post-instillation with vibrant colors. The fluidity of the post-instillation lipid layer was also enhanced compared to baseline, where areas of high lipid thickness appeared to spread over the ocular surface at a faster rate. Furthermore, spreading of the post-instillation lipid layer appears to persist for a longer duration between blinks, where particles and leading edges of lipid pools appear to continue spreading while the next blink is occurring. In contrast, in pre-instillation images, lipid spreading often plateaus to a halt before the next blink.

**Conclusions:** The Ohio State University SVCM appears to be an effective instrument to assess dynamic tear lipid effects of artificial tears on the ocular surface, and a more quantitative study is required to characterize these dynamic effects.

**Commercial Relationships:** Matthew J. Kowalski, Allergan, Inc (F); Jennifer S. Fogt, Allergan, Inc (F); Peter E. King-Smith, None; Joseph T. Barr, Allergan, Inc (F), Allergan, Inc (C)

**Support:** Allergan, Inc.

**Program Number:** 3533 Poster Board Number: A0386

**Presentation Time:** 3:45 PM–5:30 PM

**Comparative expression of inflammatory markers in corneal tissue of patients with keratoconus vs healthy corneal tissue**

Abigail Kaye, Atzin Robles-Contreras, Oscar Fernandez, Alejandro Babayan Sosa, Regina Velasco, Diana Raya, Elisa Alegria, Cristina Pacheco Del Valle. Fundacion Hospital Nuestra Senora de la Luz, Mexico City, Mexico.

**Purpose:** Keratoconus is defined as a non-inflammatory pathology of the cornea. Recent investigations have found a probable inflammatory role in the pathophysiology, but the amount of inflammatory component has not been clearly determined yet. The aim of the study is to detect overexpression of inflammation and tissue damage markers in the corneal stroma of keratoconus compared with healthy corneal stroma.

**Methods:** A comparative, cross-sectional and prospective study was carried out in Cornea and Refractive Surgery and Biomedical Research Center of the Fundación Hospital Nuestra Señora de la Luz in Mexico City, Pro. Proper informed consent was obtained, corneal tissue sample was collected from patients with advanced keratoconus who underwent penetrating keratoplasty (PK group), and as a control group, stromal lenticules were collected by ReLEx® SMILE (Carl Zeiss, VisMax 2.0®) technique in patients undergoing refractive surgery; ectasic disease was ruled out (SMILE group). Both tissue samples were stored in PBS tubes with lysin buffer inhibitor at -80 degrees Celsius.

Quantification of cytokines and tissue damage markers were performed with a protein array on nitrocellulose membranes using the Proteome Profiler Array-Human Angiogenesis kit (R&D Systems®), following the manufacturer’s instructions. Results were visualized with G-box imaging system, and analyzed the density of each point with the Vision Works LS software. Data were normalized relative to the positive and negative control. We considered as a biological significance more than 2-fold change.

**Results:** Twenty-two patients were included, 7 from the PK group and 15 from the SMILE group. Significant overexpression of angiogenin (3-fold change), dipeptidyl peptidase IV (4-fold change), endostatin (6-fold change), fibroblast growth factor type 1 (3-fold change) and 2 (2-fold change), insulin-like growth factor type 2 (2-fold change), interleukin-8 (3-fold change), metalloproteinases-8 (5-fold change) and 9 (2-fold change), peptidase inhibitor-5 (3-fold change) and urokinase protein (2-fold change) were observed in the PK group compared to the SMILE group.

**Conclusions:** An overexpression of inflammatory proteins in corneal tissue of patients with keratoconus was found compared with healthy corneal tissue. The more significant was endostatin, which has a role in scar formation. Any underexpressed inflammatory protein was found.

**Commercial Relationships:** Paulina Camacho, None; Atzin Robles-Contreras, None; Oscar Fernandez, None; Alejandro Babayan Sosa, None; Regina Velasco, None; Diana Raya, None; Elisa Alegria, None; Cristina Pacheco Del Valle, None

**Program Number:** 3534 Poster Board Number: A0387

**Presentation Time:** 3:45 PM–5:30 PM

**Grading of ocular surface inflammation using anterior segment angiography: pixel densitometry index**

Abigail Kaye, Bernhard Steger, Matthias Brunner, Colin E. Willoughby, Yalin Zeng, Vito Romano. 1Department of Eye and Vision Science, University of Liverpool, London, United Kingdom; 2Department of Corneal and External Eye Diseases, St.Paul’s Eye Unit, Liverpool, United Kingdom; 3Department of Innsbruck, Medical University of Innsbruck, Innsbruck, Austria.

**Purpose:** Clinical grading of ocular surface inflammation is crucial for disease monitoring. Commonly used grading systems exhibit considerable inter- and intra-observer variability, demonstrating the need for an improved grading method. Our studies aim to investigate the grading of ocular surface hyperemia using white pixel densitometry of angiographic images.

**Methods:** Design: Prospective cross sectional study

Setting: Academic tertiary care referral center

Participants: The study group comprised patients with ocular surface inflammation resulting from conjunctivitis and or keratitis. The control group included patients with no history of ocular surface disease.

Interventions: Color photography and indocyanine green angiography (ICGA) of the ocular surface were performed on each patient. Single images were taken up to 3 minutes before and 10 minutes after instillation of phenylephrine hydrochloride 2.5% eye drops.

**Main Outcome and Measures:** Pixel densitometry index (PDI)

**Results:** Fourteen eyes of 14 patients were recruited in the study group and eleven eyes of 11 patients in the control group. In the study group the mean patient age at presentation was 40.7 ± 16.9 years, in the control group 45.1 ± 18.3 years. The PDI before and after instillation of phenylephrine hydrochloride 2.5% eye drops was 71.1 ± 8.1 and 46.1 ± 7.2 respectively (p = 0.03). In the control group, the PDI before and after instillation of phenylephrine hydrochloride 2.5% eye drops was 57.3 ±21.9 and 43.2 ± 15.8 respectively(p = 0.1). The mean visual acuity was 0.30 ± 0.33 logMAR in the study group and it was 0.1 ± 0.18 logMAR in the control group. No patients had any adverse events during angiography.

**Conclusions:** The PDI obtained from anterior segment angiographic images is a useful tool to grade and follow ocular surface inflammation.

**Commercial Relationships:** Abigail Kaye; Bernhard Steger, None; Matthias Brunner, None; Colin E. Willoughby, None; Yalin Zeng, None; Vito Romano, None

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Program Number: 3535 Poster Board Number: A0388
Presentation Time: 3:45 PM–5:30 PM
Quantification of Inflammatory Cells in the Corneal Sub-basal Layer in Type 2 Diabetes by In Vivo Confocal Microscopy (IVCM)
Reza A Badian1,2, Tor Utheim1,4, Stephan Allgeier1, Xu Liu3, Bernd Köhler4, Neil S. Lagali1. 1Faculty of Vision and Health Sciences, University College of Southeast Norway, Kongsberg, Norway; 2The Norwegian Dry Eye Clinic, Oslo, Norway; 3Department of Medical Biochemistry, Unit of Regenerative Medicine, Oslo University Hospital, Oslo, Norway; 4Department of Ophthalmology, Vestre Viken Hospital Trust, Drammen, Norway; 5Institute of Applied Computer Science/ Automation, Karlsruhe Institute of Technology, Karlsruhe, Germany; 6Oe yelegesenteret, Tromso, Norway; 7Department of Ophthalmology, Faculty of Health Sciences, Linköping University, Linköping, Sweden.

Purpose: To quantify and compare mature dendritic cells (mDCs), immature dendritic cells (iDCs) and globular cells (GCs) in the sub-basal layer of the central cornea in healthy and Type 2 diabetes mellitus (T2DM) patients, using IVCM obtained large-area mosaic images.

Methods: 82 subjects, including healthy and T2DM patients were included in the study. The central cornea in both eyes of all study subjects (163 eyes) was imaged by IVCM, using a Heidelberg HRT 3 with Rostock corneal module. IVCM images were used to make the best possible mosaics that were subsequently analyzed by an experienced examiner using image J. Mosaics were subjected to multiple-parameter quantitative analyses with respect to different types of inflammatory cells (mDCs, imDCs, GCs).

Results: Average size of mosaics was 6.0 mm², which consisted of an average of 37 single confocal fields of view. Mature DCs were the least prevalent cells compared to immature DCs and globular cells. The ratio between both the number of imDCs and globular cells to the total number of inflammatory cells did not show any significant difference between healthy and T2DM patients. Interestingly, the ratio of mDCs/Type I cells to the total number of inflammatory cells showed a significant increase (P < 0.006, Mann-Whitney) in patients with T2DM compared to healthy subjects.

Conclusions: The corneal sub-basal layer can be imaged using IVCM to produce large-area mosaics. The analysis of the sub-basal layer can provide information on the presence and distribution of different types of inflammatory cells (mDCs, imDCs and GCs) and their characteristics. Inflammatory cells of mature dendritic phenotype and morphology could be a marker for increased inflammation or immune activation in type 2 diabetes. The possibility to non-invasively assess inflammation in diabetes may improve management of the condition.

Commercial Relationships: Reza A Badian, None; Tor Utheim, None; Stephan Allgeier, None; Xu Liu, None; Bernd Köhler, None; Neil S. Lagali, None

Program Number: 3536 Poster Board Number: A0390
Presentation Time: 3:45 PM–5:30 PM
Phase-resolved fluorometer for fluorescence lifetime measurements in the human eye
Alex Meyer1, Asnika Sridhar1, Ramesh Babu2, Uday B. Kompella1, Sangly P. Srinivas1. 1Optometry, Indiana University, Bloomington, IN; 2Computer Science, DSCE, Bangalore, India; 3Pharmaceutical Sciences, University of Colorado, Denver, CO.

Purpose: To report on the enhancements to an ocular fluorometer for performing lifetime-based pO2 measurements in the human eye.

Methods: A slit lamp has been modified to perform ocular fluorimetry. To enable depth-resolved measurements, an adjustable collection slit is placed confocal to the excitation slit (i.e., the slit of the illumination optics) along the emission axis. The measurements of fluorescence lifetime (T) are accomplished in the frequency domain (FD) so that tan φ = ωT, where ω = 2πf and φ is the phase delay in the emission relative to excitation; f is frequency of the excitation sine wave. To accomplish high-speed modulation of the excitation, we replaced the halogen lamp of the slit lamp with an LED. For measurements of T, we employed a lock-in amplifier (SR830) whose reference was coupled to the sync signal of a function generator which provided a sine wave input for driving the LED. The signal input of the lock-in was coupled to the output of a red-sensitive photomultiplier (R928). The excitation frequency was set up at f = 1/(2π) (T1 * T2) where T1 and T2 are expected lifetimes in the

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**Results:** The fluorometer could be maneuvered to measure fluorescence from the tears, cornea, and a/c easily. The excitation and emission slits were positioned confocal to each other and could be ascertained readily with the sighting optics situated in between the detector and collection slit. We have performed initial experiments with formulations of Ru phenanthroline bound to silica and dispersed in silicone ($\lambda_{ex} = 440 \text{ nm}$, $\lambda_{em} > 600 \text{ nm}$) and Pd-porphyrin ($\lambda_{ex} = 520 \text{ nm}$, $\lambda_{em} > 600 \text{ nm}$). As is characteristic of the frequency-domain method, the $pO_2$-sensitive phase delay, and modulation data could be acquired in excess of 100 Hz yielding a high temporal resolution in the measurements of $pO_2$. Calibration with mixtures of $O_2$ and $N_2$ indicated high quenching constants for formulations of Ru and porphyrin. The optimal excitation frequencies for the two dyes were 33 and 4 kHz, respectively.

**Conclusions:** The new FD spot fluorometer is capable of $pO_2$ measurements with high sensitivity and temporal resolution as required for measurements of rapid $pO_2$ dynamics underneath contact lenses. The sighting-optics enables easy positioning of the excitation and emission slits to be confocal which is necessary for high depth resolution.

**Commercial Relationships:** Alec Meyer, None; Asnika Sridhar, None; Ramesh Babu, None; Uday B. Kompella, None; Sangly P. Srinivas, None

**Support:** CTSI and FRSP (PI-SP)

**Program Number:** 3539 **Poster Board Number:** A0391
**Presentation Time:** 3:45 PM–5:30 PM
**The use of TeraHertz scanning system as a quantitative tool in the evaluation of corneal edema**


**Purpose:** To evaluate the feasibility and accuracy of using the Terahertz scanning system as a quantitative tool in the evaluation of corneal edema

**Methods:** Compressed collagen sheets that contained different water concentrations ranging from 8% to 92% were first scanned with the TPS Spectra 3000 Teraview Terahertz scanning system. The peak intensity of the curve from the spectral domain was recorded and used to establish the standard curve. Porcine eyes ($n = 50$) with different extent of corneal edema resulting from different time duration after procurement (0 day to 4 days), were then scanned with the Terahertz and anterior segment optical coherence tomography systems, to obtain the hydration level and central corneal thickness (CCT), respectively. The correlation between the hydration level and CCT was also analyzed.

**Results:** The Terahertz scanning system had the capacity to differentiate different extents of corneal stromal edema (Figure). For the corneas with the procurement time of 0, 1, 2, 3 and 4 days, the mean peak signal intensity was 159951, 224761±6223, 250290±5665, and 327236±6368 atomic units, and the corresponding hydration level was 75.3±7.8%, 79.2±6.2%, 84.7±8.2%, 89.7±10.9%, and 96.8±10.1%, respectively. The mean CCT was 682±45, 745±68, 836±48, 1006±66, and 1225±62 $\mu m$, respectively. The peak signal intensity was positively and significantly correlated with CCT measurements ($r = 0.91$, $P = 0.002$).

**Conclusions:** The Terahertz scanning system has the potential to provide a novel tool to quantitatively evaluate corneal hydration status. It may be helpful in monitoring the disease progression in patients with corneal edema.
Comparison of keratometry measurements using the iDesign® Advanced WaveScan Studio System, manual and auto-keratometers

Janice Tarrant, Ying Wang, Sanjeev Kasthurirangan. Abbott Medical Optics, Martinez, CA.

Purpose: To assess the agreement between keratometry measurements taken with a full gradient corneal topographer, the iDesign® Advanced WaveScan Studio™ (AWS) System, and manual and auto-keratometers.

Methods: A retrospective analysis was performed of pre-operative keratometry data from 167 right eyes of 170 subjects who underwent LASIK correction of myopic refractive errors. The mean age of the subjects was 32.3 ± 8.3 years (range: 18 to 58 years) with slightly more males (54.7%) than females. Outcome measures comprised steep (Ks) and flat (Kr) keratometry, steep keratometry axis, mean keratometry (Kmean), corneal astigmatism (C) and the J1 and J2 components. Measurements were made on 60 eyes with manual keratometry, 107 eyes with auto-keratometer and on all eyes with iDesign® System topography. Paired t-tests and Bland-Altman plots with 95% limits of agreement were used to evaluate the difference between measurements.

Results: The differences between the devices in all outcome measures were small and not clinically significant. Pre-operative iDesign® Ks ranged from 40.675 to 47.735 D. Ks values were 44.13 ± 1.30 D and 44.24 ± 1.33 D for manual and iDesign® devices respectively (p = 0.05, n = 60) and 44.14 ± 1.49 D and 44.22 ± 1.41 D for auto and iDesign® devices respectively (p = 0.04, n = 107). C values were 1.71 ± 1.31 D and 1.74 ± 1.27 D for manual and iDesign® devices respectively (p = 0.37 and 1.59 ± 1.31 D and 1.65 ± 1.35 D for auto and iDesign® devices respectively (p = 0.005). For manual and iDesign® devices, J1 values were 0.74 ± 0.72 D and 0.77 ± 0.69 D respectively (p = 0.28) and 0.65 ± 0.72 D and 0.67 ± 0.74 D for auto and iDesign® devices respectively (p = 0.02). For manual and iDesign® devices, J2 values were 0.02 ± 0.30 D and 0.02 ± 0.31 D respectively (p = 0.91) and -0.02 ± 0.34 D and 0.02 ± 0.37 D for auto and iDesign® devices respectively (p = 0.002). The 95% limits of agreement were similar for the paired device analyses and slightly smaller between the auto-keratometer and iDesign® topographer.

Conclusions: The iDesign® AWS System produced comparable keratometry measurements to manual and auto-keratomers in pre-refractive surgery eyes.

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Program Number: 3540 Poster Board Number: A0393
Presentation Time: 3:45 PM–5:30 PM

Limitations of Portable Cameras for Detecting Anterior Segment Pathology

Maria A. Woodward, Leslie M. Niziol, David C. Musch, Paul P. Lee. Ophthalmology and Visual Sciences, University of Michigan, Ann Arbor, MI.

Purpose: While posterior eye diseases cause the majority of visual impairment in the US, anterior segment (AS) eye diseases cause the majority of visits to eye-care professionals. We evaluated the diagnostic accuracy of detecting corneal ulcers, opacities, and abrasions using external photographs from two portable AS cameras for telemedicine purposes.

Methods: A prospective study of patients >= 18 years old with a clinical diagnosis in one eye of corneal pathology. When possible, photographs of both eyes were obtained. Gold-standard diagnosis was determined from a slit-lamp exam by a cornea specialist. iTouch 5S and Nidek VersaCammm cameras were used to obtain a series of images including multiple gazes with white light and fluorescein with blue light. Three cornea specialists interpreted the images for presence, suspicion, or absence of AS pathology. Sensitivity and specificity to detect AS disease compared to gold standard diagnosis were calculated, stratified by camera and grader. Reliability of diagnosis was evaluated with weighted kappa statistics. Grader confidence in diagnosis was assessed on a scale from 1 (not at all) to 10 (most confident).

Results: 188 eyes (108 patients) were photographed. By gold standard diagnosis, 59 eyes (31%) had corneal scars, 34 (18%) had ulcers, 13 (7%) had abrasions, and 82 (44%) were normal. Sensitivity on photographs. Additional work is needed to improve the ability to detect AS pathology remotely.

Commercial Relationships: Maria A. Woodward, None; Leslie M. Niziol, None; David C. Musch, None; Paul P. Lee, None
Support: NIH Grant K23EY023596;

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The association between central and peripheral corneal astigmatism based on Fourier analysis in normal eyes

Vinod Maseedupally, Kerry Ho, Kevin Tran, Helen A. Swarbrick, School of Optometry and Vision Science, The University of New South Wales, Sydney, NSW, Australia.

**Purpose:** With the increasing popularity of large diameter rigid lenses such as orthokeratology lenses, characterizing peripheral corneal shape in relation to the central cornea has become very important. We investigated the association between central and peripheral corneal astigmatism using Fourier analysis.

**Methods:** Three composite corneal topography maps were obtained from 55 healthy participants (26M, 31F; 21±2 years) using the Medmont E300 corneal topographer (V 7.12, Melbourne, Australia). To generate composite maps, topography was captured in the primary, nasal, temporal, inferior and superior gazes. The topographer’s built-in software then combined these maps into one single composite map. Right eye axial power maps from the topographer were imported into a custom written MATLAB program (V 8.5 Mathworks Inc., USA). Fast Fourier Transformation was applied to the axial power data to decompose astigmatism into regular and irregular components in the central (2.5 to 4 mm diameter) and peripheral (9 to 10 mm diameter) annular zones. The root mean square (RMS) for regular, irregular and total astigmatism was determined. Based on normality, the Wilcoxon signed rank test and Spearman rank correlations were used to investigate differences and associations between the two regions.

**Results:** The mean Sim K astigmatism was 1.19±0.83D. The mean RMS for central regular, irregular and total astigmatism was 0.37±0.26D, 0.07±0.04D and 0.13±0.08D respectively. The mean RMS for peripheral regular, irregular and total astigmatism was 0.70±0.43D, 0.46±0.25D and 0.52±0.27D respectively; these were all significantly greater than in the central region (all p<0.001).

There was a strong correlation between SimK and RMS central total astigmatism (r=0.89, p<0.001). A strong correlation was also noted between RMS central irregular and central total astigmatism (r=0.67, p<0.001), and between RMS peripheral irregular and peripheral total astigmatism (r=0.96, p<0.001).

**Conclusions:** Corneal astigmatism is significantly greater in the peripheral than central region. Eyes with higher amounts of astigmatism tend to have higher amounts of irregular astigmatism in the central and peripheral regions. The influence of these differences in corneal shape in central and peripheral regions on large diameter contact lens fitting is yet to be investigated.

**Commercial Relationships:** Vinod Maseedupally, Kerry Ho, None; Kevin Tran, None; Helen A. Swarbrick, None

**Support:** This research was funded through the Australian Research Council Linkage Project Scheme with support from Industry partners Bausch + Lomb (Rochester, NY), BE Enterprises Pty Ltd (Brisbane, Australia) and Capricornia Contact Lens Pty Ltd (Brisbane, Australia).

**Program Number:** 3543 Poster Board Number: A6396 Presentation Time: 3:45 PM–5:30 PM Evaluation of corneal tomography parameters with Pentacam

Adriana Ribeiro De Almeida, Rayssa M. Léda, Eduardo D. Feijó, Augusto Pereira, André P. Bittencourt. Hospital Oftalmológico de Anápolis, Anápolis, Brazil.

**Purpose:** To analyze corneal tomography parameters with Pentacam in patients cared for at Ophthalmology Hospital of Anápolis.

**Methods:** This cross-sectional study analyzed data from Pentacam (Oculus Optikgeräte GmbH, Wetzlar, Germany) of 90 patients (180 eyes), who had this exam at Ophthalmology Hospital of Anápolis from July 2015 to August 2016. Inclusion criteria: age between 12 and 80 years. Exclusion criteria: previous eye surgery. A triage was performed using the KISA% index, in which a percentage of 60% or less indicates the absence of keratoconus, between 60 and 100% indicates suspected keratoconus and greater than or equal to 100% indicates keratoconus (KCN). Several parameters acquired from Pentacam were analyzed and differences between the KCN and KCN-suspect groups and the normal group were assessed.

**Results:** There were 20 males (22.2%) and 70 females (77.8%) in the group of 90 patients with a mean age of 36.43 years (SD 14.0 years). The study showed that 81.1% of eyes had no KCN according to the KISA% index, 6.1% had suspected KCN and 12.8% had KCN. There was a significant difference in steep keratometry (K2), maximum axial curvature (Kmax), thinnest corneal location (Pachy Min), thickness at the apex of the cornea (Paqui Apex), Y coordinate of the thinnest corneal location (TLx), asphericity coefficient (QV), progress index maximum (ProgMax) and average (ProgAvg), Ambrosio relational thickness maximum (ARTMax), posterior elevation (PE) in patients with KCN compared to the normal group (p<0.05). As for the KCN-suspect group, only Kmax, ProgMax and ARTMax were statistically significant compared to the normal group (p<0.05).

**Conclusions:** The corneal tomography analysis with Pentacam in this study showed a statistically significant difference between the Pentacam parameters in patients with KCN and suspected KCN compared to patients with normal eyes.

**Table I.** Mean values for Pentacam parameters by study group

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Keratoconus</th>
<th>Suspected Keratoconus</th>
<th>Normal</th>
<th>P Value*</th>
</tr>
</thead>
<tbody>
<tr>
<td>K1</td>
<td>44.05±3.27</td>
<td>44.24±1.44</td>
<td>43.49±1.93</td>
<td>0.47±0.209</td>
</tr>
<tr>
<td>K2</td>
<td>48.10±2.75</td>
<td>46.17±1.49</td>
<td>45.05±1.94</td>
<td>0.00±0.063</td>
</tr>
<tr>
<td>Kmax</td>
<td>47.21±3.56</td>
<td>46.34±2.24</td>
<td>44.71±2.33</td>
<td>0.00±0.026</td>
</tr>
<tr>
<td>Paqui Min</td>
<td>-60.13±37.69</td>
<td>524.27±39.44</td>
<td>523.11±34.45</td>
<td>0.00±0.913</td>
</tr>
<tr>
<td>ProgMax</td>
<td>472.26±38.87</td>
<td>533.73±37.22</td>
<td>529.36±34.13</td>
<td>0.00±0.679</td>
</tr>
<tr>
<td>TLx</td>
<td>-0.11±0.88</td>
<td>-0.11±0.80</td>
<td>-0.04±0.73</td>
<td>0.09±0.782</td>
</tr>
<tr>
<td>TLy</td>
<td>-0.53±0.31</td>
<td>-0.46±0.45</td>
<td>-0.36±0.35</td>
<td>0.05±0.395</td>
</tr>
<tr>
<td>ECC</td>
<td>0.63±0.54</td>
<td>0.27±0.58</td>
<td>0.39±0.30</td>
<td>0.05±0.518</td>
</tr>
<tr>
<td>QV</td>
<td>-0.09±0.57</td>
<td>-0.61±0.41</td>
<td>-0.24±0.19</td>
<td>0.01±0.204</td>
</tr>
<tr>
<td>ProgAvg</td>
<td>1.75±0.46</td>
<td>1.16±0.17</td>
<td>1.02±0.27</td>
<td>0.00±0.421</td>
</tr>
<tr>
<td>ProgMax</td>
<td>2.53±0.77</td>
<td>1.66±0.39</td>
<td>1.33±0.38</td>
<td>0.00±0.027</td>
</tr>
<tr>
<td>ARTMax</td>
<td>200.43±72.30</td>
<td>347.74±99.30</td>
<td>419.45±107.98</td>
<td>0.00±0.034</td>
</tr>
<tr>
<td>KLs</td>
<td>0.24±0.62</td>
<td>0.16±0.56</td>
<td>0.39±0.86</td>
<td>0.42±0.394</td>
</tr>
<tr>
<td>KLy</td>
<td>-1.32±1.66</td>
<td>-0.11±0.63</td>
<td>-0.42±0.60</td>
<td>0.05±0.434</td>
</tr>
<tr>
<td>PE</td>
<td>24.35±20.30</td>
<td>2.45±8.98</td>
<td>4.46±5.34</td>
<td>0.00±0.258</td>
</tr>
</tbody>
</table>

*Keratoconus versus normal group: Keratoconus suspect versus normal group

**Commercial Relationships:** Adriana Ribeiro De Almeida, None; Rayssa M. Léda, None; Eduardo D. Feijó, None; Augusto Pereira, None; André P. Bittencourt, None

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Methods: A video camera was attached to the ESP to view the instrument’s focusing spots from a second direction. The operator can then position the ESP at a more repeatable distance (Z) from the eye. X/Y alignment is handled via the original ESP focusing system. A blue LED ring light, operated via a footswitch, was also attached to the camera to give a live view of tear fluorescence. When consistent fluorescence coverage was observed, the operator switches off the ring light and collects data immediately with the original flash system. To investigate repeatability, five maps were collected for the right eyes of 3 subjects using A) the original ESP focusing technique (ESP) and B) our new camera and focusing technique (NEWf). Maps for each subject for each technique were filtered to remove artefacts then averaged to derive a standard deviation map (SD at each map grid point). To compare ESPf with NEWf, the SD maps for all subjects were averaged and split into ‘corneal’ (central 10 mm diameter) and ‘limbal/scleral’ (outside 10 mm diameter) regions.

Results: The ‘corneal’ average SD was 8.6 um for ESPf and 4.8 um for NEWf (44% reduction). The ‘limbal/scleral’ average SD was 19.6 um for ESPf and 11.1 um for NEWf (43% reduction).

Conclusions: The hardware modifications to the ESP have enhanced focusing precision and reduced variability between maps for the eyes tested. By viewing tear film fluorescence prior to collecting data, we have also ensured that tear film artefacts have less impact on data quality.

Figure 1: camera and ringlight on ESP (a), image showing focusing spots and poor fluorescein coverage (b), SD map for ESPf (c) and SD map for NEWf (d)

Commercial Relationships: Brett A. Davis, None; Pryntha V. Rajasingam, None; Michael J. Collins, None; Alyra J. Shaw, None; Hamish J. McNeill, None
**Purpose:** Measuring corneal clarity in healthy eyes is important because it could help when planning treatments. This study aims to standardize and investigate the changes in corneal clarity with age. Densitometry software for the Oculus Pentacam was used to examine corneal clarity at different age groups.

**Methods:** A total of 192 eyes of 97 healthy participants were included in this cohort comparative non-randomized cross-sectional study. Central Manchester University Hospitals NHS Foundation Trust, Manchester, UK and NREC local ethics committee approved this study. An Oculus Pentacam was used to image cornea for healthy participants grouped by sex and age (between 10 and 70 years old). Data from the densitometry output has been used to determine the clarity in concentric zones and different depths of the cornea.

**Results:** Corneal densitometry across all age groups showed significant differences between groups when we divided the corneal into layers: anterior, central and posterior (p<0.05). We also showed significant differences when the cornea was divided in to 0-2 mm, 2-6 mm, and 6-10 mm concentric zones (p<0.05). The most striking increase in densitometry values occurred with age in all 3 layers was in the periphery (6-10 mm) (p<0.05). Additionally, we showed that the 10-20 year age group had lower clarity than the 20-30 age group (p<0.05) and after 30 years the cornea shows a steady progression of increased densitometry values. Densitometry of the anterior and posterior layer was shown to be the least stable with aging.

**Conclusions:** This study is the first study to our knowledge that has standardized corneal densitometry value in healthy control eyes, which include teenage densitometry values. These values for corneal densitometry, as well as subdivisions based on layer and surface area, might provide a standardized stage for use in further studies and clinical practice. This study established that relation between corneal densitometry and age is differed when cornea divided by layers and zone. This study suggested that there are other factors play an essential role in corneal densitometry level as well as age.

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**Commercial Relationships:** Khaled Alzahrani, None; Fiona Carley, None; Arun Brahma, None; Debbie Morley, None; M Chantal Hillarby, None

**Program Number:** 3547 **Poster Board Number:** A0400

**Presentation Time:** 3:45 PM - 5:30 PM

**Change in intraocular pressure following cataract extraction is unrelated to preoperative anterior chamber angle, and to lens thickness as measured by Pentacam**

**Richard Hession, Bradford Tannen.** Ophthalmology, Mount Sinai School of Medicine, New York, NY.

**Purpose:** Cataract extraction can lower intraocular pressure (IOP). Whether the decrease in IOP is related to preoperative anterior chamber parameters is unclear. A retrospective chart review was performed to examine the relationship between preoperative anterior chamber angle (ACA), lens thickness, and change in average IOP following cataract extraction.

**Methods:** A retrospective chart review was conducted for patients who underwent uncomplicated cataract extraction with posterior chamber intraocular lens implantation by one experienced surgeon over a two-year interval (2014-2015). Data collected included preoperative ACA and lens thickness, as measured by Pentacam, as well as average preoperative IOP and average postoperative IOP, measured by applanation tonometry. All pre- and post-operative IOP measurements were taken either before the initiation of, or after the termination of all perioperative topical medication. Patients with angle-closure glaucoma, open-angle glaucoma with prior IOP-lowering surgery, or with a change in IOP lowering medications over the study period were excluded. Regression analysis was performed to analyze the data.

**Results:** Charts of 85 patients were reviewed. 47 patients (69 eyes) were included in the ACA analysis. Of these patients, 14 (19 eyes) had a lens thickness measurement available for analysis. Average preoperative ACA was 34.3 degrees. Average lens thickness was...
The stereopsis that supports 3D vision in keratoconus. This knowledge may help in clinical or surgical decisions for vision rehabilitation. The aim of the present work is to evaluate the motor and sensorial aspects of vision on patients with keratoconus.

**Methods:** A series of patients with keratoconus, confirmed by corneal tomography and graded by the ABCD grading system, were evaluated for binocular vision and extraocular muscle motor function. Best-corrected visual acuity (BCVA) with glasses was measured in ETDRS chart at 4 meters. Binocular vision status for distance was checked using simple and alternate cover tests, red striate Maddox (for simultaneous perception), 6° base down prism (for fusion), and a prism bar (for fusional convergence). Stereopsis was measured using the Titmus Fly Test.

**Results:** Forty-eight patients, with a mean age of 19.3 ± 7.3 years old, with a female: male ratio of 25:23 were evaluated. Considering the eyes with the worst vision, 2 (4%) were classified as A0 (Kmax <46.5), 3 (6%) as A1 (Kmax <48), 24 (50%) as A2 (Kmax <53), 3 (6%) as A3 (Kmax <55) and 17 (35%) as A4 (Kmax >55). Six (13%) patients had exotropia and 12 (25%) had heterophoria. Nine (20%) out of 46 had no simultaneous perception, 10 (22%) out of 46 had no binocular fusion and 10 (29%) out of 34 had a fusional convergence of less than 15°. For stereopsis, 8 (20%) out of 45 had stereopsis equal or less than 63° and 23 (51%) out of 45 had 400° or worse. Patients whose BCVA in the worse eye was less than 0.2 (categories D3 <0.2 or D4 <0.05) had a significantly higher frequency of exotropia (p<0.05; OR: 12; 95% CI, 2.1-68.1), absence of simultaneous perception (P<0.05), and absence of fusion (p<0.05; OR: 12; 95% CI, 2.3-61.7).

**Conclusions:** In our study, keratoconus patients presented a higher frequency of impaired binocular vision than the general population. It indicates a worse 3D vision in these individuals. Future studies are needed to elucidate the factors associated with the loss of binocularity in this disease, such as the age of onset of ectasia or high order aberrations. Testing for 3D vision may be helpful to consider in the guidelines of treatments for patients with keratoconus.

**Commercial Relationships:** Rosalia Maria Antunes-Foschini, None; Eduardo M. Rocha, None

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**3.63 mm. Average pre- and post-operative IOP was 14.1 and 12.3 mmHg, respectively. Average pre- and post-operative IOP for the lens thickness subgroup was 13.5 and 11.8 mmHg, respectively. Average change in IOP was -1.8 mmHg. Change in IOP for the lens thickness subgroup was -1.5 mmHg. Change in IOP was not related to preoperative ACA (p=0.75, R²=0.0015), or to lens thickness (p=0.08, R²=0.1675).

**Conclusions:** Average IOP decreased following uncomplicated cataract extraction. In this study, the postoperative decrease in IOP was unrelated to preoperative ACA, and to lens thickness, as measured by Pentacam. Cataract extraction can aid in IOP control; however, neither the preoperative ACA, nor lens thickness can be used to predict the amount of IOP reduction that will result from the procedure.

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**Commercial Relationships:** Richard Hession, None; Bradford Tannen, None

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**Program Number: 3550 Poster Board Number: A0403**

**Presentation Time:** 3:45 PM–5:30 PM

**Topographic Elevation Data to Design Scleral Lenses**

Louise Sclafani1, David Slater1, Bruno Lay2, Christine W. Sindt2.

1Ophthalmology, University Of Chicago, Chicago, IL; 2Ophthalmology, University Of Iowa, Iowa City, IA; 3Eyeprint Prosthetic, Lakewood, CO; 4Adcis, St. - Contest, France.

**Purpose:** Ocular Impression technology has been used to effectively create elevation specific scleral lens designs. We propose the elevation data from scanning tomography (Pentacam, Oculus DE) can be used to create an elevation specific scleral lens, without the need for time-intensive contact lens fitting or ocular impressions.

**Methods:** Scheimpflug technology was used to collect 5 scans: 1 central, 2 horizontal and 2 vertical exams in primary gaze. Each Sub-exam uses 50 segmental images, so in total 250 Images are captured during the acquisition phase. The 5 examinations are stitched together to create one 3D shape file covering 17mm of the cornea and sclera. There is no fixation displacement during image capture, so tilt reconstruction is not necessary. The software was first validated against known reference test surfaces, and then compared to ocular impression technology 3-D scans. Finally, lenses were generated, using the elevation specific technology in the EyePrint Design software, and evaluated for ocular fit, vision and comfort.

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A 3-D geometrical model of the cornea was constructed. It was used to calculate the shape and size of the region of biomechanical weakening in keratoconic corneas.

**Purpose:** To develop a spatial model of keratoconic biomechanical properties and to develop an artificial intelligence method to estimate the shape and size of the affected zone using the model.

**Methods:** A 3-D geometrical model of the cornea was constructed using tomography (OCULUS Optikgerate Gmbh, Germany). The model included the epithelium and stroma. Transverse fiber dependent material model was used (Sinha Roy et al., J Mech Behav Biomed Mat, 2015). To model KC, it was assumed that: (a) the disease caused local biomechanical degeneration; (b) the strength of the stroma in KC was determined by the residual collagen network that was unaffected by the disease. Thus, the material properties were multiplied by a linear “factor” that varied spatially over the cornea to model localized steepening (Sinha Roy et al., IOVS, 2011). The model was used to compute the shape and size of the region of localized weakening in progressive keratoconus using artificial intelligence (AI) and finite element method (FEM). By evaluating the “factor” at several locations in the stroma, the AI automatically adjusted the factor at each location so as to minimize the difference between the measured anterior corneal curvature of the progressed stage of KC and the FEM estimate of the same.

**Results:** Figure 1 shows the outcomes of the model, when applied to a progressive KC case. The patient progressed to KC two years after the 1st measurement (year 2010) and continued thereafter (2013 and later). The eye had normal topography at 1st visit. Figure 1A shows the axial curvature measured in 2015. Figure 1B shows the model outcome at the same time point. Figure 1C, D and E shows the AI predicted spatially varying map of the “factor” for years 2013, 2014 and 2015, respectively. A “factor” of 1 (Figure 1C, D and E) indicated no change in material properties. For e.g., if at a given location in the cornea, the computed factor was 0.5 and the modulus of the cornea in the unaffected region was 1 MPa, then the modulus of the cornea at that location due to diseases progression was 0.5 MPa (1 x 0.5).

**Conclusions:** An algorithmic approach using conventional tomography was developed to estimate the region of biomechanical weakening in KC cornea. The method showed the irregular shape and size of the region of biomechanical weakening, which may be used to plan corneal crosslinking procedures.
Increased Prevalence of Homosexuals in the Keratoconus Population
Lisa Marten¹, Susan L. Johnson⁴, Matthew A. Reilly⁴, William E. Sponsel¹, South Texas Eye Institute, San Antonio, TX; ²Dr. William E. Sponsel, MD, San Antonio, TX; ³Biomedical Engineering, The Ohio State University, Columbus, OH; ⁴UIW Rosenberg School of Optometry, San Antonio, TX.

Purpose: Keratoconus is a rare disease and is estimated as a prevalence of 54.5 cases per 100,000. Although recent studies have reviewed the genetic component of Keratoconus, no specific gene has been identified. Homosexuality (self-reported) is 3.4% of the adult population in the United States. Furthermore, no specific gene for homosexuality has been found, yet we observed an increase number of patients who are keratoconic and homosexual. We performed a retrospective study in order to learn more about the prevalence of keratoconus amongst homosexual patients.

Methods: We conducted a retrospective chart review and selected subjects from the entire keratoconus population in the South Texas Eye Institute seen within the last 4 years (n=84, M=48, F=36). From this group, we determined their sexual orientation by self-report or listed a same sex significant other on our intake form. Our control group (n=252) consists of age and sex matched patients without keratoconus. Exclusion criteria included patients under the age of 18, patients with Down’s syndrome, and subjects with any other systemic disease. Risk factor was calculated for both study and control groups via linear regression model: y=m₁x₁+m₂x₂+m₃x₃+b. The categories for analysis are 1=age, 2=gender, 3=sexual orientation, m=relative risk associated with each category (Kennedy 1986; Gallup 2012), and b=risk in each population not identified as homosexual.

Results: The database of patients at South Texas Eye Institute revealed a self-reported homosexual population of 9.52% among those with keratoconus (n=8, F=5, M=3), and only 1.19% were homosexual in matched controls, (n=3, F=1, M=2). Our findings demonstrated that there was an increased prevalence of homosexuals in our keratoconus population compared to the control group (p= 0.00018, 95% confidence interval 0.237-0.7499, mean effect=0.4934). Thus, the risk of keratoconus is about 50% higher for a homosexual (range 25-75%).

Conclusions: There is a statistically significant increased prevalence of homosexual patients in the Keratoconus population in our South Texas clinic, which warrants further research to see if this is reflected in the overall keratoconus population. These findings suggest that there may be a keratoconic/homosexual genetic component which potentially places the homosexual population at a higher risk for Keratoconus.

Commercial Relationships: Lisa Marten, Susan L. Johnson, None; Matthew A. Reilly, None; William E. Sponsel, None

Program Number: 3554 Poster Board Number: A0407
Presentation Time: 3:45 PM–5:30 PM
Total, corneal, and internal high order aberrations in monocular dioplia patients without anterior segment abnormality
Young Joo Park¹,², Joo Hyun Kim³, Hyun Sun Jeon³, Hee Kyung Yang³,², Jeong-Min Hwang³,², Won Ryang Wee¹, Joon-Young Hyon¹,². ¹Department of Ophthalmology, Seoul National University Hospital, Seoul, Korea (the Republic of); ²Department of Ophthalmology, Seoul National University Bundang Hospital, Seongnam, Korea (the Republic of).

Purpose: To evaluate the optical performance and wavefront profiles in the eyes presenting monocular dioplia without cornea and lens abnormalities

Methods: This retrospective study included 27 eyes of 16 patients who presented monocular dioplia with no apparent corneal abnormalities on slit lamp examination. Patients were subclassified into Clear lens group (16 eyes of 8 patients) and Haze lens group (12 eyes of 8 patients) according to the lens status on slit lamp biomicroscopy. Total high order aberration (HOA), coma, and trefoil up to 6th order in internal, corneal, and total optical path difference (OPD), and Strehl ratios at mesopic condition and at 5 mm pupil size were evaluated using OPD-Scan III.

Results: The average age was 35.3 ± 19.7 (23.3 ± 12.3 with Clear lens group and 51.2 ± 16.3 with Haze lens group, p < 0.001). Although there was no apparent abnormalities in cornea and lens on slit lamp examination, clear lens group showed increased root mean square (RMS) of HOA (0.57 ± 0.294, reference value 0.32 ~ 0.42) and decreased Strehl ratio (-2.900 ± 0.278 by natural logarithm, reference value -2.60 ~ -1.90). There was no significant difference between Clear lens group and Haze lens group in all parameters except in the trefoil and HOA values of the total OPD in the condition with 5 mm-sized pupil (p=0.030 and p=0.028 respectively). However, the HOAs of mesopic pupil were significantly higher than the HOAs of 5 mm pupil (p=0.002 in total, p=0.003 in corneal, p=0.003 in internal OPD) in Clear lens group.

Conclusions: The results suggest that the relative increase of HOA in the mesopic condition may induce monocular dioplia in the eyes without apparent anterior segment abnormality.

Commercial Relationships: Young Joo Park, None; Joo Hyun Kim, None; Hyun Sun Jeon, None; Hee Kyung Yang, None; Jeong-Min Hwang, None; Won Ryang Wee, None; Joon-Young Hyon, None

Program Number: 3555 Poster Board Number: A0408
Presentation Time: 3:45 PM–5:30 PM
Atopic dermatitis as a high risk of recurrent protrusion after penetrating keratoplasty in keratoconic eyes
Masahiro Yamaguchi, Yoshimune Hiratsuka, Toshinari Funaki, Satoru Nakatani, Akira Matsuda, Akira Murakami. Ophthalmology, Juntendo University, Bunkyo-ku, Japan.

Purpose: To assess the effect of atopic dermatitis on the recurrence of keratoconic protrusion following penetrating keratoplasty.

Methods: A retrospective single-center analysis was performed on 111 keratoconic eyes that underwent penetrating keratoplasty between January 2000 and December 2015. Of these, 18 eyes were diagnosed with atopic dermatitis (AD). The mean follow-up period was 8.7 years (range, 1.5-19 years). The recurrence of keratoconic protrusion was analyzed using the Kaplan-Meier method and compared between eyes with and without AD.

Results: The incidence of keratoconic protrusion was 4/18 eyes (22.2%) in the AD group and 5/93 eyes (5.4%) in the non-AD group (p = 0.014). The median time to keratoconic protrusion was 3.7 years (95% CI, 0.4-7.0 years) in the AD group and 7.5 years (95% CI, 2.7-12.2 years) in the non-AD group (p = 0.018). The recurrence rate of keratoconic protrusion at 5 years was 11.1% (95% CI, 2.3-20.0%) in the AD group and 3.2% (95% CI, 0.4-6.0%) in the non-AD group (p = 0.035).

Conclusions: Atopic dermatitis is a high risk of recurrent keratoconic protrusion after penetrating keratoplasty. Further studies are needed to elucidate the underlying mechanisms and develop preventive strategies.
Purpose: To detect whether if atopic dermatitis affects to recurrent protrusion after keratoplasty in keratoconic eye.

Methods: 54 eyes of 50 patients who were diagnosed keratoconus, who were received keratoplasty in Juntendo University from 2006 to 2015 and who were conducted suture removal, were included in this study. Criteria for diagnosing protrusion were defined by existence of clinical findings of keratoconus, or corneal topographic change (localized 1D steeping of K-max in the axial power in a year, asymmetric inferior protrusion and inferior thinning). A Cox proportional hazards model was used to determine the influence of donor, recipient and surgical factors on protrusion. Kaplan Meier survival estimates was also conducted the long-term probability of recurrent protrusion.

Results: Recurrent protrusion was detected in 5 eyes of 5 cases (11.1%), occurred in 3.6±4.3±6.9 years after surgery. Univariate analysis in a Cox proportional hazards model revealed atopic dermatitis had 14.7±8 times higher (p<0.01), although recipient age, graft age, graft size, removal duration, and suture type had no statistical difference. Multivariate analysis showed atopic dermatitis had 14.0±7 times higher risk without effect of suture removal duration. Kaplan Meier survival estimates in 10-year follow up showed 65.0±% of patients with atopic dermatitis accompanied protrusion, compared to 2.5±% of those who were without atopic dermatitis (p=0.001, Log-rank test).

Conclusions: Our results revealed strong correlation between atopic dermatitis and protrusion after keratoplasty in keratoconic eyes.

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Presentation Time: 3:45 PM–5:30 PM
Exome sequencing analysis in three Bedouin families with Keratoconus

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Purpose: 1 in 2000 people are affected by Keratoconus (KT). Despite its prevalence within the general population, researchers have been unable to identify a confirmed genetic cause of KT. It has been documented that Keratoconus seems to occur more frequently and aggressively in Israel and Saudi Arabia. In order to identify a genetic cause of KT, we performed exome sequencing in three Bedouin families with a family history of KT.

Methods: We obtained a total of 16 samples from 3 Bedouin families with a history of KT. These samples were genotyped using an Affymetrix Genome-Wide SNP 5.0 microarray. Using MERLIN, we analyzed the SNP data for regions consistent with segregation within each of the families under both dominant and recessive models of inheritance. The SNP data was also analyzed for regions of autozygosity using PLINK to identify regions of the genome shared between the affected family members in each of the families. One KT sample from each of the families was chosen for exome sequencing. Variants were filtered based upon predicted function, variant quality, population prevalence, and commonly called unfounded variants (based upon our local set of 1000+ exomes) to create a final variant list. The variant list was annotated with corneal expression (http://genome.uiowa.edu/otdb) to assist in prioritizing potential candidates.

Results: No plausible variants were identified within the three families in genes previously reported to cause KT. In addition, no single gene with plausible disease-causing variants was shared across all three families. We identified a few genes with variants that are shared between two of the three families. Upon further inspection, however, variants in these genes were identified as technical artifacts. We also identified three regions of autozygosity that are shared between the three families. However, analysis of these regions failed to provide strong candidate genes or plausible mutations.

Conclusions: Our data supports the genetic heterogeneity of KT even within relatively isolated populations. As we proceed with our analysis, we will focus on variants within each of the families. We are also working with our collaborator in Israel to ascertain additional family members from our KT families, and to ascertain additional families with KT. This will allow us to further narrow the intervals of the genome in search of the causative mutations.

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Presentation Time: 3:45 PM–5:30 PM
Arginase, P4H and iNOS expression and the effect of iNOS inhibitor 1400W on urea, hydroxyproline and nitrite formation of keratoconus keratocytes

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Purpose: Keratoconus (KC) is a stromal disease of the cornea, which leads to conical shape and loss of vision in later stages. Arginase is a cytoplasmatic enzyme that catalyzes the conversion of L-arginine to urea and ornithine, which serves as a precursor for the synthesis of proline and hydroxyproline (determinate parts of collagen strands of the corneal stroma). The competing enzyme for the substrate L-arginine is nitric-oxid synthase (iNOS). In a previous study we determined decreased arginase activity, urea and hydroxyproline concentration in KC keratocytes. The purpose of our present study was to evaluate arginase, prolyl-4-hydroxylase and iNOS expression in KC keratocytes and to determine the effect of the selective iNOS inhibitor 1400W on urea, hydroxyproline and nitrite (NO) formation of these cells.

Methods: Primary human KC (n=8) and normal (n=8) keratocytes were isolated by digestion in collagenase A from human corneal buttons, and cultured in DMEM/Ham’s F12 medium supplemented with 5% fetal calf serum. Arginase and iNOS were evaluated using qPCR and Western blot analysis. The amount of prolyl-4-hydroxylase was determined using flow cytometry. Following 10, 20 and 40μM iNOS inhibitor 1400W treatment, urea, hydroxyproline and NO concentration in KC keratocytes were assessed using colorimetric assays.

Results: KC keratocytes showed a 6-fold increased iNOS mRNA expression compared to normal cells, whereas Arginase expression remained unchanged. Arginase could not be detected using Western blot analysis, and iNOS expression showed no differences in
normal and KC keratocytes. The amount of prolyl-4-hydroxylase was decreased in KC keratocytes compared to controls \((p=0.038)\). Using all of the above inhibitor concentrations, urea \((p<0.01)\) and NO \((p<0.007)\) concentrations were significantly decreased in KC keratocytes, but hydroxyproline concentration remained unchanged \((p>0.09)\).

**Conclusions:** Our results indicate that the expression of the urea cycle enzymes is altered in KC keratocytes. The iNOS inhibitor 1400W decreases urea and NO formation without changes in hydroxyproline concentration in these cells. Therefore, its topical use does not seem to be a potential treatment option for keratoconus patients. Further investigations concerning the regulatory processes in collagen synthesis are needed to fully understand the pathogenesis of keratoconus.

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**Presentation Time:** 3:45 PM–5:30 PM

**Keratoconus Endophenotypes in a Healthy Aging Population**

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**Purpose:** Keratoconus is a complex condition; little is known about genetic factors underlying its susceptibility. Use of disease endophenotypes in population-based studies has identified genetic variants associated with various eye conditions, such as central corneal thickness (CCT) and keratoconus. This study aimed to access topographic measures that have been suggested as keratoconus risk indices – Keratoconus Prediction Index (KPI), apical gradient of curvature (AGC) and Symmetry Index (SI) – as possible endophenotypes in a healthy aging population.

**Methods:** Corneal topography and wavefront data from the Visionix VX120 autorefractor was analysed for 1430 twins (91% female, mean age 58.9, age range 18.5–91.7) from the TwinsUK cohort. STATA14 was used for exploring normality tests, correlations (Pearson, Spearman and Lin’s concordance correlation coefficients), association between age, CCT, and rank normalised AGC and SI (linear regression), and for conducting t-test for differences in means of predictors between KPI groups. Falconer’s formula was used to calculate broad-sense heritability \((H^2)\) for topographic measures in 414 monozygotic and 300 dizygotic twin pairs.

**Results:** CCT was normally distributed \((P=0.3)\), had a mean \([SD]\) of 537.4\(\mu m\) [34.2\(\mu m\)], decreased with age \((\beta=-0.17, P=0.01)\), and had a \(H^2\) of 0.72. AGC and SI were not normally distributed \((P<0.0001)\), did not vary with age, had median \([range]\) of 1.3 [-16.1:19.6] and 0.33 [-14.2:15.5], and had a \(H^2\) of 0.12 and 0.37 respectively. AGC and SI were not correlated with CCT or KPI but were weakly correlated with each other \((\rho=0.13)\). KPI followed trimodal distribution with 30% of participants having at least one eye predicted to be affected, although none of them had keratoconus. These individuals were on average older, had thinner corneas and lower K \((P=0.003, P=0.02\) and \(P<0.0001\) respectively). The \(H^2\) of KPI \((\rho_c)=0.60\). Correlation between both eyes was strong for all variables: CCT \((r^2=0.82)\), AGC and SI \((\rho=0.62\) both), and KPI \((\rho_c=0.55)\).

**Conclusions:** This study shows that although KPI is heritable, it is a poor keratoconus endophenotype as it mislabels 30% of individuals as cases, and is also a semi-quantitative measure. While AGC and SI had better distributions, they had lower heritability and did not strongly correlate with KPI or other predictors. Although CCT is known to not be an ideal endophenotype, it may still be the most reliable endophenotype for keratoconus to date.

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