• Clinical Research •

# Comparative 2-year outcomes of conventional and accelerated corneal collagen crosslinking in progressive keratoconus

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## Abstract

● **AIM:** To compare the safety and efficacy of conventional versus accelerated (9 mW/cm$^2$) corneal collagen crosslinking (CXL) in progressive keratoconus at the 2-year follow-up.

● **METHODS:** In this prospective study, consecutive progressive keratoconus patients were randomized to receive either conventional CXL (CCXL) or accelerated CXL (ACXL; using hydroxypropyl methylcellulose-assisted riboflavin imbibition for 10min at 9 mW/cm$^2$). Visual, refractive, keratometric, topographic, and aberrometric outcomes and stromal demarcation line depth (DLD) measurements were compared at the end of a 2-year follow-up.

● **RESULTS:** Thirty-two eyes from 32 patients in the CCXL and 27 eyes from 27 patients in the ACXL groups completed 2-year follow-up. At 2y post-CXL, both uncorrected and corrected visual acuities improved significantly in both groups. The improvements in keratometric readings, flattening rate (flattening of the maximum keratometry more than 1 D), 3 topographic indices, and vertical coma were significantly better in the CCXL group compared to the ACXL group ($P$<0.05). The DLD as measured by anterior segment optical coherence tomography or *in vivo* confocal microscopy was better detectable and significantly deeper in the CCXL group compared to the ACXL group. The deeper DLD was found to be significantly correlated with improvements in the mean keratometry measurements. Progression was noted in 11.1% of eyes in the ACXL group, whereas progression was not observed in any patient eye in the CCXL group.

● **CONCLUSION:** In this prospective randomized study, ACXL is less effective in halting the progression of keratoconus at a 2-year follow-up compared to CCXL.

● **KEYWORDS:** corneal collagen crosslinking; keratoconus; corneal ectasia; riboflavin; accelerated crosslinking
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## INTRODUCTION

Keratoconus is a common bilateral, progressive corneal ectatic disorder. Corneal collagen crosslinking (CXL) with riboflavin and ultraviolet-A (UVA), first introduced in 2003 by Wollensak *et al*[1], is the only safe and effective means to halt the progression of this disease. The procedure is based on photopolymerization of the stromal collagen using a photosensitizer (riboflavin) and UVA irradiation that results in corneal stiffening due to an increase in the number of inter- and intra-fibrillar covalent bonds[2].

The treatment protocol introduced by Wollensak *et al*[1] was named as the standard, Dresden, or conventional corneal collagen crosslinking (CCXL) protocol and requires a soaking time of 30min in the riboflavin solution and an illumination time of 30min for 3 mW/cm$^2$ UVA irradiation. This CCXL protocol has been extensively used during the past ten years and has demonstrated its long-term safety and efficacy[3-6]. The major drawback of the conventional protocol is its long treatment duration, which is an hour. Recently, the concept of accelerated CXL (ACXL) was introduced with the purpose of reducing the illumination time by increasing UVA intensity. A number of different ACXL protocols have been proposed based on the Bunsen-Roscoe law of reciprocity. Results from experimental or clinical ACXL studies using these varying protocols are controversial up to date and only short-term data from a few studies is available.

In this study, our aim was to compare the safety and efficacy of CCXL and ACXL (9 mW/cm$^2$) in progressive keratoconus at a 2-year follow-up.

## SUBJECTS AND METHODS

**Ethical Approval** This study was approved by the Institutional Review Board Committee at Ankara University and was

conducted according to the tenets of the Declaration of Helsinki. Written informed consent was obtained from all patients.

**Patient Population** Consecutive patients older than 18y who were diagnosed with progressive keratoconus and scheduled for CXL surgery were enrolled in this prospective study. After enrollment, a computer-generated simple randomization method was used in which patient eyes were randomized to receive either CCXL or ACXL treatment. If both eyes of one patient were progressing, only the worse eye was included in the study.

The diagnosis of keratoconus was based on the presence of slit-lamp biomicroscopy and corneal tomography findings obtained using the Scheimpflug corneal imaging system (Pentacam; Oculus GmbH, Wetzlar, Germany). Progressive keratoconus was documented by serial differential topography and pachymetry analyses. Progression was defined as an increase of at least 1 diopter in the maximum keratometry (Kmax) reading within the past 12mo.

Exclusion criteria included thinnest corneal thickness (TCT) less than 350 μm measured using a Pentacam, central corneal scarring, previous ocular trauma or corneal surgery, a history of herpetic keratitis, or any autoimmune/systemic disease, pregnancy or lactation. Rigid gas permeable contact lens wearers discontinued lens wear for 4wk, and soft contact lens wearers discontinued lens wear 2wk before the baseline ophthalmic examination for CXL.

**Preoperative and Postoperative Evaluation** All patients underwent a detailed ophthalmological examination at baseline, and at 1, 3, and 6mo, and 1 and 2y after receiving CCXL or ACXL treatment. The following were recorded during each visit: uncorrected distance visual acuity (UDVA), best spectacle-corrected distance visual acuity (CDVA), manifest refraction (MR), slit-lamp biomicroscopy findings, corneal tomography (Pentacam; Oculus GmbH, Wetzlar, Germany), and *in vivo* confocal microscopy (IVCM; HRT II, Rostock Cornea Module). One-month postoperatively, the corneal stromal demarcation line depth (DLD) was measured using anterior segment optical coherence tomography (AS-OCT; Visante, Carl Zeiss, Germany) and the corneal stromal devoid of the keratocytes (treatment depth) was measured using IVCM in all patient eyes. On AS-OCT, the relative depth of the central demarcation line [in % of total central corneal thickness (CCT)] was also measured. The visibility of the demarcation line was scored to obtain the accuracy of the measurements (0, line not visible; 1, line unclear, measurements not clearly visible; 2, line clearly visible). Only measurements with a score of 2 were used to measure the DLD and the percentage of the treated cornea in the central cornea using the measurement toolbars of the AS-OCT software. Corneal haze was graded at each visit by slit-lamp biomicroscopy using the scale established by Greenstein *et al*[7].

**Corneal Tomography** At least 3 topography measurements were taken from each patient eye during each follow-up examination. On the best-quality topography maps, the corneal thickness, sagittal curvature, anterior and posterior elevation data, and tomographic indices were evaluated preoperatively and at all follow-up examinations. The topographic cone location was assessed using previously described methods[8].

**Surgical Technique** All procedures were performed by the same surgeon (Uçakhan ÖÖ) under sterile conditions. After topical anesthesia with proparacaine hydrochloride 0.5%, the central 9.0 mm corneal epithelium was removed by mechanical debridement. After epithelial removal, ultrasound (US) pachymetry (P-1, Takagi Seiko, Japan) was performed to measure the CCT. Five measurements were performed from the central cornea. After the thinnest and thickest measurements were discarded, the mean of the three measurements was recorded as the de-epithelized CCT.

In CCXL with the Dresden protocol, 0.1% riboflavin in 20% dextran T500 solution (MedioCross, Kiel, Germany) was instilled every 2min for 30min. Intraoperative pachymetry was performed before irradiation and UVA light at 365 nm (UV-X system, IROC AG, Switzerland) was applied for 30min at an irradiance of 3.0 mW/cm$^2$. During UVA exposure, riboflavin 0.1% drops continued to be administered every 2min. In ACXL, riboflavin 0.1% with hydroxypropyl methylcellulose (HPMC; Vibex Rapid, Avedro Inc, Waltham, MS, USA) was applied every 2min for 10min. Then, US pachymetry was performed to measure the CCT and 365 nm UVA irradiation at 9 mW/cm$^2$ was administered for 10min (Avedro, Waltham, USA). During UVA exposure, riboflavin 0.1% drops continued to be administered every 2min.

At the end of either procedure, US pachymetry was performed again to measure the CCT and a silicone hydrogel bandage contact lens was placed on the cornea. The bandage contact lens was removed after epithelial healing. Topical antibiotics (Moxifloxacin hydrochloride 0.5%, Vigamox, Alcon, USA) were prescribed 4 times daily for 1wk and topical loteprednol etabonate 0.5% suspension eye drops (Lotemax, Bausch&Lomb, USA) were applied 4 times daily after epithelization with gradual tapering and discontinuation.

**Outcome Measures, Treatment Failure, and Adverse Events** The primary outcome measure was the change in Kmax at the end of the follow-up period compared to baseline. The secondary outcome measures were the changes in the UDVA, CDVA, manifest refraction spherical equivalent (MRSE), keratometric measurements, topographical indices, corneal aberrations, and endothelial cell density (ECD) compared to the baseline.

Treatment failure was defined as an increase in Kmax of more than 1.0 diopter during the follow-up. An adverse event was defined as a loss of 2 or more Snellen lines of CDVA compared to baseline.

**Statistical Analyses** Statistical analyses were performed using SPSS 2 (Version 2.0; SPSS for Windows, IBM, Armonk, NY, USA). Using power and sample size calculation software, enrollment of at least 32 eyes (16 eyes in each group) was determined to be necessary for a meaningful statistical analysis. All continuous variables were presented as means and standard deviations (SD). Visual acuity was converted to logMAR notation for its statistical analysis.

The normality of the data was tested using the Shapiro-Wilk test. Within-group baseline and postoperative parameters were compared using the Mann-Whitney *U* test. Within each group, changes between the pre- and postoperative values were compared using the Wilcoxon signed-rank test. DLD values between the groups were compared using the Mann-Whitney *U* test. Correlation analysis of the DLD and topographic keratometric values were performed using Spearman correlation test. The level of statistical significance was set at $P<0.05$.

## RESULTS

**Patient Characteristics** Sixty-four patients were enrolled in this prospective study. Thirty-four patients received CCXL and 30 patients received ACXL. Two patients (2/34, 5.9%) in the CCXL group and 3 patients (3/30, 10.0%) in the ACXL group were lost to follow-up. Measurements from 32 eyes of 32 patients in the CCXL and 27 eyes of 27 patients in the ACXL groups were eligible for statistical analyses. At baseline, the two groups were matched for age, sex, preoperative visual acuity, refractive, keratometric, pachymetric measurements, cone location, and keratoconus grade[9] ($P>0.05$; Table 1).

During surgery, the mean CCT measurements taken following keratectomy were 422.94±33.72 μm in the CCXL group and 411.59±42.04 μm in the ACXL group. Following 30min of riboflavin-dextran imbibition, the mean CCT was 456.66±54.91 μm in the CCXL group. In the ACXL group, following 10min of riboflavin-HPMC application, the mean CCT was 462.37±30.99 μm.

**Visual Acuity** The mean preoperative and postoperative UDVA and CDVA data are summarized in Figure 1. The mean UDVA and CDVA significantly improved in both groups compared to baseline at 3, 6, 12, and 24mo postoperatively ($P<0.05$; Table 2, Figure 1).

**Manifest Refraction** The mean MRSE and MR cylinder improved significantly in both the CCXL and ACXL groups with no differences found between groups at any time point ($P>0.05$; Table 2).

**Keratometry Readings** At month 24, compared to baseline, the flattening of the mean keratometry (Km), steep keratometry (Ks), flat keratometry (Kf), and Kmax were statistically significant in the CCXL group ($P=0.029$, $P=0.001$, $P=0.043$, and $P=0.007$, respectively; Table 2). All keratometric improvements except Kmax were significantly improved in the CCXL group compared to the ACXL group ($P<0.05$; Figure 2).

At month 24, the mean flattening in Kmax were -0.9±1.1 D in the CCXL group and -0.4±0.9 D in the ACXL group. The flattening rate (flattening of the Kmax more than 1 D) was 43.7% (14/32) versus 29.6% (8/27) in the CCXL and ACXL groups, respectively. The flattening rate was significantly higher in the CCXL group compared to the ACXL group ($P=0.03$).

**Table 1 Baseline demographic characteristics, visual, refractive, keratometric, pachymetric values, and cone location of the patients in the CCXL group and ACXL group** mean±SD

| Parameters | CCXL (32 eyes, 32 patients) | ACXL (27 eyes, 27 patients) | *P* |
|---|---|---|---|
| Age (y) | 23.13±3.8 | 24.69±5.0 | 0.494 |
| Sex (male:female) | 20:12 | 17:10 | 0.311 |
| Visual acuity (logMAR) | | | |
| UDVA | 0.74±0.6 | 0.61±0.5 | 0.520 |
| CDVA | 0.23±0.2 | 0.19±0.1 | 0.539 |
| Refractive error (D) | | | |
| MR sphere | -3.4±4.8 | -3.3±5.2 | 0.757 |
| MR cylinder | -3.8±2.6 | -3.2±2.0 | 0.220 |
| MRSE | -5.4±5.4 | -5.1±5.5 | 0.837 |
| Pentacam parameters | | | |
| Kmax (D) | 54.7±7.3 | 56.0±5.4 | 0.357 |
| Km (D) | 48.3±4.2 | 48.7±4.7 | 0.778 |
| Ks (D) | 50.1±4.8 | 50.7±5.0 | 0.599 |
| Kf (D) | 46.8±3.8 | 46.9±4.6 | 0.988 |
| Kas (D) | 3.2±1.9 | 3.8±2.0 | 0.454 |
| CCT (μm) | 463.9±31.3 | 457.8±42.3 | 0.518 |
| TCT (μm) | 445.4±37.2 | 439.1±45.0 | 0.420 |
| Cone location, *n* (%) | | | |
| Central | 28 (87.5) | 23 (85.2) | 0.227 |
| Paracentral | 4 (12.5) | 4 (14.8) | 0.309 |
| Keratoconus grade, *n* (%) | | | |
| Grade I | 18 (56.3) | 14 (51.9) | |
| Grade II | 9 (28.1) | 10 (37.0) | |
| Grade III | 2 (6.2) | 0 | |
| Grade IV | 3 (9.4) | 3 (11.1) | |

CCXL: Conventional corneal collagen crosslinking; ACXL: Accelerated corneal collagen crosslinking; UDVA: Uncorrected distance visual acuity; CDVA: Corrected distance visual acuity; MR: Manifest refraction; MRSE: Manifest refraction spherical equivalent; Kmax: Maximum keratometry; Km: Mean keratometry; Ks: Steep keratometry; Kf: Flat keratometry; Kas: Keratometric astigmatism; CCT: Central corneal thickness; TCT: Thinnest corneal thickness.

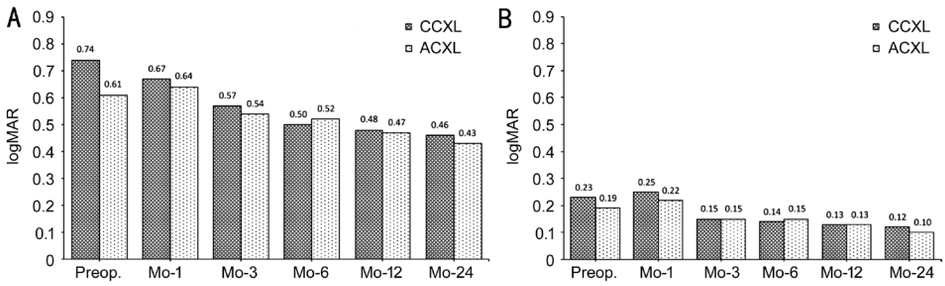


**Figure 1 Changes in UDVA and CDVA** A: Change in the mean UDVA over time in the CCXL group and ACXL group; B: Change in the mean CDVA over time in the CCXL group and ACXL group.

**Table 2 Preoperative and month 24 postoperative visual, refractive, keratometric indices, corneal thickness, elevation measurements, topographic indices, and anterior corneal aberrations in the CCXL group and ACXL group** mean±SD

| Parameters | CCXL | | | ACXL | | | Intergroup difference |
|---|---|---|---|---|---|---|---|
| | Preop. | Mo-24 | *P* | Preop. | Mo-24 | *P* | *P*-value |
| UDVA (logMAR) | 0.74±0.6 | 0.46±0.4 | <0.001 | 0.61±0.5 | 0.43±0.5 | <0.001 | 0.082 |
| CDVA (logMAR) | 0.23±0.2 | 0.12±0.1 | <0.001 | 0.19±0.1 | 0.10±0.1 | 0.001 | 0.351 |
| MR sphere (D) | -3.4±4.8 | -3.4±4.4 | 0.244 | -3.3±5.2 | -3.0±5.2 | 0.109 | 0.744 |
| MR cylinder (D) | -3.8±2.6 | -3.3±2.2 | 0.027 | -3.2±2.0 | -2.9±2.8 | 0.043 | 0.607 |
| MRSE (D) | -5.4±5.4 | -4.7±5.4 | 0.037 | -5.1±5.5 | -4.7±5.7 | 0.049 | 0.873 |
| Kmax (D) | 54.7±7.3 | 53.8±7.2 | 0.007 | 56.0±5.4 | 55.6±5.4 | 0.050 | 0.491 |
| Km (D) | 48.3±4.2 | 48.03±4.3 | 0.029 | 48.7±4.7 | 48.7±5.0 | 0.475 | 0.047 |
| Ks (D) | 50.1±4.8 | 49.7±5.0 | 0.001 | 50.7±5.0 | 50.6±5.3 | 0.211 | 0.01 |
| Kf (D) | 46.8±3.8 | 46.2±4.0 | 0.043 | 46.9±4.6 | 46.7±4.9 | 0.971 | 0.045 |
| Kas (D) | 3.2±1.9 | 3.2±2.0 | 0.675 | 3.8±2.0 | 3.6±1.9 | 0.212 | 0.439 |
| CCT (μm) | 463.9±31.3 | 462.7±35.5 | 0.674 | 457.8±42.3 | 467.1±44.6 | 0.001 | 0.004 |
| TCT (μm) | 445.4±37.2 | 443.3±39.9 | 0.471 | 439.1±45.0 | 448.4±47.0 | 0.001 | 0.006 |
| MAE (μm) | 26.3±15.1 | 22.6±14.3 | 0.001 | 27.5±9.4 | 24.6±7.2 | 0.007 | 0.731 |
| ISV | 81.96±22.7 | 77.04±23.5 | <0.001 | 74.53±40.7 | 71.66±39.3 | 0.046 | 0.065 |
| IVA | 0.82±0.3 | 0.75±0.3 | <0.001 | 0.59±0.5 | 0.53±0.4 | 0.044 | 0.072 |
| KI | 1.17±0.1 | 1.03±0.1 | <0.001 | 1.20±0.1 | 1.18±0.1 | 0.015 | <0.001 |
| CKI | 1.06±0.05 | 1.0±0.07 | <0.001 | 1.05±0.03 | 1.04±0.03 | 0.056 | <0.001 |
| Rmin | 6.21±0.7 | 6.24±0.8 | 0.063 | 6.06±0.5 | 6.11±0.5 | 0.059 | 0.334 |
| IHA | 32.75±31.5 | 29.82±25.0 | 0.746 | 34.26±21.2 | 31.94±25.8 | 0.280 | 0.670 |
| IHD | 0.1±0.07 | 0.09±0.07 | <0.001 | 0.11±0.04 | 0.1±0.04 | 0.002 | <0.001 |
| VC (μm) | -1.81±1.5 | -1.59±1.53 | <0.001 | -2.0±1.4 | -1.84±1.3 | 0.044 | 0.03 |
| SA (μm) | -0.15±0.8 | -0.12±0.7 | 0.570 | -0.05±0.5 | -0.01±0.5 | 0.156 | 0.867 |

CCXL: Conventional corneal collagen crosslinking; ACXL: Accelerated corneal collagen crosslinking; UDVA: Uncorrected distance visual acuity; CDVA: Corrected distance visual acuity; MR: Manifest refraction; MRSE: Manifest refraction spherical equivalent; Kmax: Maximum keratometry; Km: Mean keratometry; Ks: Steep keratometry; Kf: Flat keratometry; Kas: Keratometric astigmatism; CCT: Central corneal thickness; TCT: Thinnest corneal thickness; MAE: Maximum anterior elevation; ISV: Index of surface variance; IVA: Index of vertical asymmetry; IHA: Index of height asymmetry; IHD: Index of height decentration; CKI: Central keratoconus index; KI: Keratoconus index; Rmin: Minimum sagittal curvature in the central 8.0 mm; VC: Anterior corneal vertical coma; SA: Anterior spherical aberration.

**Corneal Thickness and Topographic Indices** The mean CCT and TCT values decreased significantly in both groups at postoperative month 1 ($P$=0.015 and $P$=0.05 for CCXL and ACXL groups, respectively) and then gradually increased over time until month 24. The mean CCT and TCT values improved to baseline levels at postoperative month 24 in the CCXL group and at month 6 in the ACXL group (Table 2).

At month 24, the mean flattening in the maximum anterior elevation (MAE) was significantly different from baseline in the CCXL and ACXL groups (Table 2). There was no shift in the topographic localization of the cone apex in either group. Four of seven tomographic indices evaluated improved significantly in both groups, while one index [central keratoconus index (CKI)] improved significantly only in the CCXL group, compared to baseline ($P$<0.001; Table 2). The improvements in the mean KI, CKI, and IHD were significantly greater in the CCXL group compared to the ACXL group ($P$<0.001; Table 2).

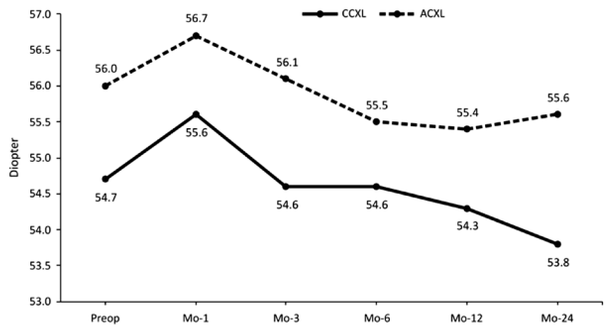


**Figure 2 Change in the mean Kmax over time in the CCXL group and ACXL group.**

The mean anterior vertical coma (VC) decreased significantly compared to baseline in both groups. However, the improvement in VC was significantly greater in the CCXL group compared to the ACXL group ($P$=0.03).

**Endothelial Cell Counts** At month 24, compared to baseline, there was no loss of endothelial cells in either group ($P$>0.05).

**Demarcation Line Depth–Anterior Segment OCT** Demarcation line measurements of 1 eye in the CCXL group and 10 eyes in the ACXL group were suboptimal and therefore excluded from the analysis. The demarcation line after 1mo was clearly visible (score of 2) in 25 of 31 eyes (80.6%) in the CCXL group and in 9 of 17 eyes (52.9%) in the ACXL group. The ACXL group had a significantly lower occurrence rate of a clear demarcation line compared to the CCXL group ($P$=0.02). In eyes with a visible demarcation line (score of 2), the mean depth of the corneal stromal demarcation line was 301.2±56.3 μm (59.1% of the mean CCT; range, 170 to 430 μm) in the CCXL group and 203.3±38.4 μm (39.9% of the mean CCT; range, 150 to 260 μm) in the ACXL group. The inter-group difference was statistically significant ($P$<0.001). The DLD correlated was significantly correlated with the Km measurements ($r$=-0.372, $P$=0.03), whereas, the DLD did not correlate with the Kmax ($r$=-0.307, $P$=0.07; Figure 3).

**Demarcation Line Depth–*in vivo* Confocal Microscopy** The transition zone from the stroma with apoptotic keratocytes to normal keratocytes was considered the demarcation line on IVCM. Seven eyes in the CCXL and 6 eyes in the ACXL group were excluded from this analysis because of suboptimum IVCM measurements. The mean IVCM DLD was at 317.48±47.2 μm (76.4% of the CCT; range, 235 to 402 μm) in the CCXL group and 212.33±32.1 μm (47.9% of the CCT; range, 170 to 298 μm) in the ACXL group. The inter-group difference was found to be statistically significant ($P$<0.001).

**Slit-Lamp Biomicroscopy Findings** The epithelial defect healed by postoperative day 4 in all patient eyes. At postoperative week 1, 4 eyes (12.5%) in the CCXL group and 5 eyes (18.5%) in the ACXL groups had grade 1+ haze, which resolved to normal levels with frequent use of topical corticosteroid eye drops. Sterile infiltrates were observed in 2 eyes (6.2%) of 2 patients in the CCXL group and 2 eyes (7.4%) of 2 patients in the ACXL group. The infiltrates resolved in all eyes with frequent use of topical corticosteroid eye drops.

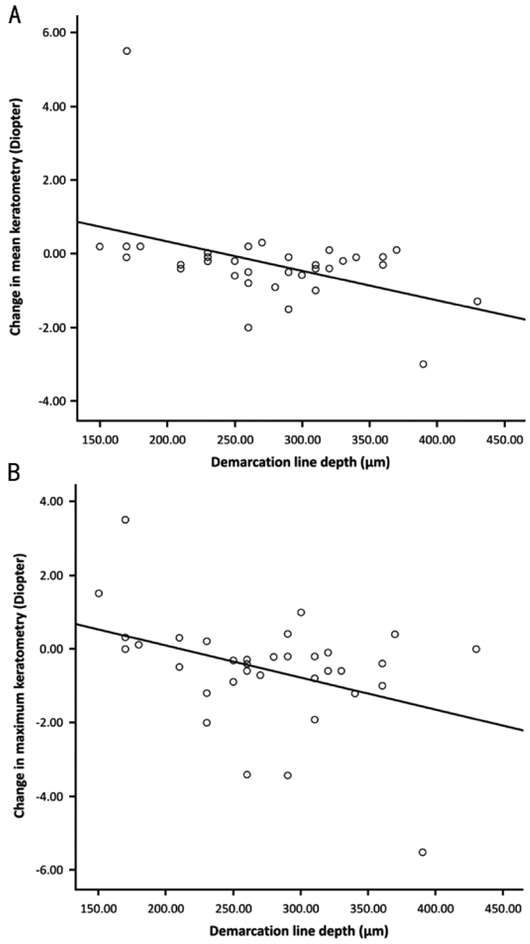


**Figure 3 Correlation analysis of the DLD with the Km and the Kmax** A: Correlation between the DLD (as measured by AS-OCT) and the mean Km; B: Correlation between the DLD (as measured by AS-OCT) and the mean Kmax.

**Progression** At the end of postoperative month 24, progression of at least 1 D in the Kmax was seen in 3 of 27 eyes (11.1%) in the ACXL group patients, whereas no progression was noted in any patient eye in the CCXL group.

## DISCUSSION

In this study, compared to baseline, keratometric, topographic, and aberrometric improvements were significantly improved and the corneal flattening rate was significantly higher in the CCXL group compared to the ACXL group. The mean DLD was also significantly more clear and deeper following CCXL compared to ACXL, which was measured using AS-OCT or IVCM. The deeper DLD correlated with more flattening in the Km readings. Finally, at the end of 2y, 11.1% of eyes in the ACXL group had progression of at least 1 D in Kmax, whereas no eye progressed in the CCXL group. Therefore, although both CCXL and ACXL (9 mW/cm$^2$) were found to

**Table 3 Design and outcomes of studies with a standardized protocol and at least 1-year follow-up using ACXL at 9 mW/cm$^2$**

| First author, year | Study design | Eye/patient | Mean age (range, y) | Follow-up (mo) | Duration (min)/ intensity (mW/cm$^2$) | Riboflavin/ soaking time (min) | Apparatus | ΔCDVA (logMAR) | ΔMRSE (D) | ΔKs (D) | ΔKmax (D) | ΔCCT (μm) | DLD (μm) | ΔECD (cell/ mm$^2$) |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Elbaz, 2014[13] | Retrospective | 16/14 | 24.9 (17.1-38.3) | 12 | 10/9 | 0.1% RF in 20% dex/30 | UV-X 2000 | -0.01 | 0.25 | 0.02 | -0.06 | N/A | N/A | N/A |
| Shetty, 2014[14] (pediatric) | Prospective | 30/18 | 12.7 (11-14) | 24 | 10/9 | 0.1% RF in 20% dex/30 | Avedro KXL | -0.12[a] | -0.95 | -2.07[a] | N/A | N/A | N/A | No loss |
| Brittingham, 2014[20] | Retrospective | 81 eyes | 28.6 | 12 | 30/3 | 0.1% RF in 20% dex/20 | UV-XTM 2000 | N/A | N/A | -0.8[b] | -0.76[b] | N/A | 323[b] | N/A |
| | Comparative | 50 eyes | 26.1 | | 10/9 | | UV-XTM 1000 | N/A | N/A | +0.52[b] | +0.72[b] | N/A | 245[b] | N/A |
| Shetty, 2015[15] | Prospective | 36/36 | 22.8 (14-33) | 12 | 30/3 | 0.1% RF in 20% dex/30 | Avedro KXL | -0.05[a] | -0.85[a] | -1.32[a,b] | N/A | N/A | 280 | -166[a] |
| | Randomized comparative | 36/36 | 23.1 (13-33) | | 10/9 | | | -0.15[a] | -1[a] | 0.67[a,b] | N/A | N/A | 292 | -187[a] |
| Ng, 2016[21] | Retrospective | 14/12 | 36.1 | 12 | 30/3 | 0.1% RF in 20% dex/25 | UV-X 1000 | -0.13[a] | 0.23 | N/A | -1.8[a,b] | -2.1 | 282[b] | N/A |
| | Comparative | 12/12 | 32.6 | | 10/9 | | UV-X 2000 | 0.02 | 0.98 | N/A | -0.3[b] | 2.1 | 209[b] | N/A |
| Toker, 2017[19] | Retrospective | 34/34 | 21.1 (13-30) | 12 | 30/3 | 0.1% RF in HPMC/20 | CCL-Vario | -0.11[a] | 0.96 | -0.74[b] | -2.15[a] | N/A | 266 | No loss |
| | Comparative | 45/45 | 22.4 (14-33) | | 10/9 | | Avedro KXL | -0.12[a] | 0.49 | -0.94[b] | -1.64[a] | N/A | 273 | No loss |
| Hagem, 2017[23] | Prospective | 20 eyes | N/A | 12 | 30/3 | 0.1% RF in HPMC/20 | UV-X 1000 | -0.11[a] | N/A | N/A | -1.4[a] | N/A | 442[b,c] | No loss |
| | Randomized comparative | 20 eyes | | | 10/9 | | UV-X 2000 | -0.09[a] | N/A | N/A | -0.5 | N/A | 317[b,c] | No loss |
| Baenninger, 2017[16] (pediatric) | Retrospective | 39/31 | 16.3 | 12 | 30/3 | 0.1% RF in 20% dex/30 | UV-X | 0.14 | N/A | N/A | 1.49 | N/A | N/A | N/A |
| | Comparative | 39/30 | 15.5 | | 10/9 | | CCL-365 Vario | 0.19 | N/A | N/A | 0.71 | N/A | N/A | N/A |
| Vounotrypidis, 2018[17] | Retrospective | 131/101 | 26.9 | 36 | 30/3 | 0.1% RF in 20% dex/30 | UV-X 1000 | -0.09 | N/A | N/A | -0.3[a] | N/A | N/A | N/A |
| | Comparative | 282/215 | 27.5 | | 10/9 | | UV-X 2000 | -0.03 | N/A | N/A | -1.4[a] | N/A | N/A | N/A |
| Sarac, 2018[18] (pediatric) | Retrospective | 38/29 | 15.0 (11-17) | 24 | 30/3 | 0.1% RF in 20% dex/30 | UVA system (Meran Tip) | 0.00 | 0.44 | -0.22 | -0.61 | N/A | N/A | N/A |
| | Comparative | 49/35 | 14.9 (10-17) | | 10/9 | | | -0.07 | 0.46 | -0.59[a] | -1.01[a] | N/A | N/A | N/A |
| Our study | Prospective | 32/32 | 24.6 (19-38) | 24 | 30/3 | 0.1% RF in 20% dex/30 | UV-X 1000 | -0.13[a] | -0.7[a] | -0.4[a,b] | -0.9[a] | -1 | 310[b] | No loss |
| | Randomized comparative | 27/27 | 23.1 (18-34) | | 10/9 | 0.1% RF in HPMC/10 | Avedro KXL | -0.07[a] | -0.4[a] | -0.1[b] | -0.4[a] | 10[a] | 240[b] | No loss |

N/A: Not applicable. [a]Statistically significantly different compared to baseline; [b]Statistically significant difference between CCXL and ACXL (9 mW/cm$^2$) groups; [c]Measurement with IVCM.

be safe procedures with no loss of endothelial cell density or significant adverse events, CCXL was more effective in halting the progression of keratoconus.

Accelerated CXL protocols were introduced with the hope of delivering an equal total dose of irridation as in CCXL in a shorter period of time while also achieving similar efficacy as in CCXL. However, as of today, there is no scientific evidence regarding the efficacy of any uniform ACXL protocol. *Ex-vivo* studies have shown conflicting results regarding the effectiveness of ACXL at 9 mW/cm$^2$[10-12].

To date, a handful of clinical studies have been performed using ACXL at 9 mW/cm$^2$. Table 3 shows a list of these studies, which used a standard protocol and at least 12mo follow-up. Similar to the *ex-vivo* studies, these studies have also reported conflicting outcomes regarding the efficacy of ACXL at 9 mW/cm$^2$. In 6 studies that used dextran-assisted riboflavin imbibition for 30min[13-18] and 1 study that used HPMC-assisted riboflavin imbibition for 20min[19], ACXL (9 mW/cm$^2$) was found to be similar to CCXL. However, two studies utilizing HPMC-assisted riboflavin imbibition for 20min[20] and 25min[21] reported more significant improvements in the Kmax measurements using the CCXL protocol. Previously, stromal demarcation lines observed at AS-OCT were put forward to use as a tool to monitor the efficacy of the CXL procedure[22]. In 2 studies, the DLD was found to be similar in ACXL group (9 mW/cm$^2$) and CCXL group[15,19]. However, in 3 studies[20-21,23], the DLD was significantly deeper using the CCXL protocol compared to the ACXL (9 mW/cm$^2$) protocol.

The difference in outcomes of the different ACXL studies utilizing the same UVA intensity may be hypothesized to be related to the type of solvent used in the riboflavin solutions. Indeed, very little is known about the effect of imbibition properties of riboflavin solutions containing dextran or HPMC. The colloid osmotic pressure of the riboflavin solution containing dextran 20% is high due to the abundant hydrophilic hydroxyl groups that lead to marked intraoperative reduction of CCT during the CXL procedure[24-25]. Using a riboflavin solution containing 0.5% of the polysaccharide HPMC has been suggested as an alternative. Because HPMC does not contain water-binding hydroxyl groups, riboflavin-HPMC solutions are known to swell the stroma faster and more effectively during the CXL procedure[26]. Despite the advantages of enhanced and faster stromal penetration of riboflavin-HPMC, it is not clear whether CXL with the use of HPMC is less effective due to the formation of a lower number of inter-fibrillar crosslinks, particularly in the anterior stroma[27-28]. The more prominent appearance of the demarcation line with the standard protocol that utilized riboflavin-dextran has been hypothesized to be due to the steeper distribution gradient of riboflavin-dextran, which tapers off rapidly in the posterior stroma, and also more UVA transmission. However, significant swelling of corneas during HPMC-assisted imbibition together with less UVA transmission results in shallow and faint demarcation lines at the AS-OCT.

Even in studies that report similar efficacy, the topographical flattening and smoothening effect of CCXL was found to be superior to that of ACXL at 9 mW/cm$^2$ regardless of the type of riboflavin-solvent used[15,19]. On the other hand, clinical outcomes, primarily in terms of efficacy in preventing progression, differ considerably, even using the same type of riboflavin solvent with the same imbibition period and utilizing the same irradiation intensities. This finding may point to factors other than riboflavin solution and/or imbibition duration playing a role in the efficacy of CCXL procedures, or may simply reveal the need for better conducted prospective studies before the efficacy of CXL with UVA intensities at 3 and 9 mW/cm$^2$ are suggested to be the same.

As of today, the clinical implications of the putative lower crosslinking of HPMC-treated corneas with shallower and less detectable stromal demarcation lines remain controversial. The long-term efficacy of ACXL at 9 mW/cm$^2$ using HPMC-assisted riboflavin imbibition for 10min is also unknown. To our knowledge, our study is the first prospective randomized study that compares the 2-year outcomes of ACXL at 9 mW/cm$^2$ using HPMC-assisted riboflavin imbibition for 10min to conventional CXL using dextran-assisted riboflavin imbibition for 30min, also known as the conventional protocol. In conclusion, in this study, ACXL (9 mW/cm$^2$) using HPMC-assisted riboflavin imbibition for 10min was found to be less effective in halting the progression of keratoconus at 2-year follow-up compared to CCXL. Progression was also noted only in the ACXL group (11.1%), whereas no patient eye progressed in the CCXL group. Our results suggest that, although ACXL protocols have the advantage of reduced treatment time and less reduction in corneal thickness, the CCXL protocol has better therapeutic efficacy. We stress the need for further randomized controlled studies with long-term follow-up, more number of patients, and uniform protocols to establish the efficacy of ACXL at 9 mW/cm$^2$.

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**REFERENCES**

1 Wollensak G, Spoerl E, Seiler T. Riboflavin/ultraviolet-A-induced

collagen crosslinking for the treatment of keratoconus. *Am J Ophthalmol* 2003;135(5):620-627.

2 Mazzotta C, Balestrazzi A, Traversi C, Baiocchi S, Caporossi T, Tommasi C, Caporossi A. Treatment of progressive keratoconus by riboflavin-UVA-induced cross-linking of corneal collagen: ultrastructural analysis by Heidelberg Retinal Tomograph II *in vivo* confocal microscopy in humans. *Cornea* 2007;26(4):390-397.

3 O'Brart DP, Chan E, Samaras K, Patel P, Shah SP. A randomised, prospective study to investigate the efficacy of riboflavin/ultraviolet A (370 nm) corneal collagen cross-linkage to halt the progression of keratoconus. *Br J Ophthalmol* 2011;95(11):1519-1524.

4 Wittig-Silva C, Chan E, Islam FMA, Wu T, Whiting M, Snibson GR. A randomized, controlled trial of corneal collagen cross-linking in progressive keratoconus. *Ophthalmology* 2014;121(4):812-821.

5 O'Brart DP, Kwong TQ, Patel P, McDonald RJ, O'Brart NA. Long-term follow-up of riboflavin/ultraviolet A (370 nm) corneal collagen cross-linking to halt the progression of keratoconus. *Br J Ophthalmol* 2013;97(4):433-437.

6 Hashemi H, Seyedian MA, Miraftab M, Fotouhi A, Asgari S. Corneal collagen cross-linking with riboflavin and ultraviolet a irradiation for keratoconus: long-term results. *Ophthalmology* 2013;120(8):1515-1520.

7 Greenstein SA, Fry KL, Bhatt J, Hersh PS. Natural history of corneal haze after collagen crosslinking for keratoconus and corneal ectasia: Scheimpflug and biomicroscopic analysis. *J Cataract Refract Surg* 2010;36(12):2105-2114.

8 Greenstein SA, Fry KL, Hersh PS. Effect of topographic cone location on outcomes of corneal collagen cross-linking for keratoconus and corneal ectasia. *J Refract Surg* 2012;28(6):397-405.

9 Krumeich JH, Kezirian GM. Circular keratotomy to reduce astigmatism and improve vision in stage I and II keratoconus. *J Refract Surg* 2009;25(4):357-365.

10 Schumacher S, Oeftiger L, Mrochen M. Equivalence of biomechanical changes induced by rapid and standard corneal cross-linking, using riboflavin and ultraviolet radiation. *Invest Ophthalmol Vis Sci* 2011;52(12):9048-9052.

11 Wernli J, Schumacher S, Spoerl E, Mrochen M. The efficacy of corneal cross-linking shows a sudden decrease with very high intensity UV light and short treatment time. *Invest Ophthalmol Vis Sci* 2013;54(2):1176-1180.

12 Hammer A, Richoz O, Arba Mosquera S, Tabibian D, Hoogewoud F, Hafezi F. Corneal biomechanical properties at different corneal cross-linking (CXL) irradiances. *Invest Ophthalmol Vis Sci* 2014;55(5):2881-2884.

13 Elbaz U, Shen C, Lichtinger A, Zauberman NA, Goldich Y, Chan CC, Slomovic AR, Rootman DS. Accelerated (9-mW/cm2) corneal collagen crosslinking for keratoconus-a 1-year follow-up. *Cornea* 2014;33(8):769-773.

14 Shetty R, Nagaraja H, Jayadev C, Pahuja NK, Kurian Kummelil M, Nuijts RM. Accelerated corneal collagen cross-linking in pediatric patients: two-year follow-up results. *Biomed Res Int* 2014;2014:894095.

15 Shetty R, Pahuja NK, Nuijts RM, Ajani A, Jayadev C, Sharma C, Nagaraja H. Current protocols of corneal collagen cross-linking: visual, refractive, and tomographic outcomes. *Am J Ophthalmol* 2015;160(2):243-249.

16 Baenninger PB, Bachmann LM, Wienecke L, Thiel MA, Kaufmann C. Pediatric corneal cross-linking: comparison of visual and topographic outcomes between conventional and accelerated treatment. *Am J Ophthalmol* 2017;183:11-16.

17 Vounotrypidis E, Athanasiou A, Kortüm K, Kook D, Shajari M, Priglinger S, Mayer WJ. Long-term database analysis of conventional and accelerated crosslinked keratoconic mid-European eyes. *Graefes Arch Clin Exp Ophthalmol* 2018;256(6):1165-1172.

18 Sarac O, Caglayan M, Uysal BS, Uzel AGT, Tanriverdi B, Cagil N. Accelerated versus standard corneal collagen cross-linking in pediatric keratoconus patients: 24 months follow-up results. *Cont Lens Anterior Eye* 2018;41(5):442-447.

19 Toker E, Çerman E, Özcan DÖ, Seferoğlu ÖB. Efficacy of different accelerated corneal crosslinking protocols for progressive keratoconus. *J Cataract Refract Surg* 2017;43(8):1089-1099.

20 Brittingham S, Tappeiner C, Frueh BE. Corneal cross-linking in keratoconus using the standard and rapid treatment protocol: differences in demarcation line and 12-month outcomes. *Invest Ophthalmol Vis Sci* 2014;55(12):8371-8376.

21 Ng AL, Chan TC, Cheng AC. Conventional versus accelerated corneal collagen cross-linking in the treatment of keratoconus. *Clin Exp Ophthalmol* 2016;44(1):8-14.

22 Seiler T, Hafezi F. Corneal cross-linking-induced stromal demarcation line. *Cornea* 2006;25(9):1057-1059.

23 Hagem AM, Thorsrud A, Sandvik GF, Råen M, Drolsum L. Collagen crosslinking with conventional and accelerated ultraviolet-A irradiation using riboflavin with hydroxypropyl methylcellulose. *J Cataract Refract Surg* 2017;43(4):511-517.

24 Kymionis GD, Kounis GA, Portaliou DM, Grentzelos MA, Karavitaki AE, Coskunseven E, Jankov MR, Pallikaris IG. Intraoperative pachymetric measurements during corneal collagen cross-linking with riboflavin and ultraviolet A irradiation. *Ophthalmology* 2009;116(12):2336-2339.

25 Mazzotta C, Caragiuli S. Intraoperative corneal thickness measurement by optical coherence tomography in keratoconic patients undergoing corneal collagen cross-linking. *Am J Ophthalmol* 2014;157(6):1156-1162.

26 Vetter JM, Tubic-Grozdanis M, Faust M, Lorenz K, Gericke A, Stoffelns BM. Effect of various compositions of riboflavin eye drops on the intraoperative corneal thickness during UVA-cross-linking in keratoconus eyes. *Klin Monbl Augenheilkd* 2011;228(6):509-514.

27 Ahearne M, Yang Y, Then KY, Liu KK. Non-destructive mechanical characterisation of UVA/riboflavin crosslinked collagen hydrogels. *Br J Ophthalmol* 2008;92(2):268-271.

28 Ehmke T, Seiler TG, Fischinger I, Ripken T, Heisterkamp A, Frueh BE. Comparison of corneal riboflavin gradients using dextran and HPMC solutions. *J Refract Surg* 2016;32(12):798-802.