Long Term Evaluation of Posterior Corneal Surface Parameters after Accelerated Corneal Crosslinking with a Comparison with Uncross-Linked Keratoconic Eyes

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Research Article

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Abstract

Purpose: To evaluate the 36 months changes in posterior corneal surface parameters in keratoconic eyes after accelerated corneal cross-linking and to compare the data with uncross-linked progressive and non-progressive keratoconic eyes.

Methods: Thirty five cross-linked, 30 uncross-linked progressive, and 30 uncross-linked non-progressive keratoconic eyes were included. Maximum keratometry (K_{max}), thinnest pachymetry, minimum radius of curvature back (R_{min}back), asphericity back, posterior elevation and corneal densitometry, back corneal higher order aberrations (HOAs), back surface deviation (Db), final D, posterior radius of curvature (PRC) and ‘B’ unit values were recorded at baseline and at the 12, 24, 36 months follow-up. Data were analyzed with repeated measures ANOVA and paired t-tests.

Results: K_{max} and thinnest pachymetry were significantly changed in the cross-linked and progressive uncross-linked groups. R_{min}back, asphericity back, and HOAs did not change in either group. Total posterior corneal densitometry improved; posterior elevation, Db and B unit worsened in the cross-linked group and did not change in the uncross-linked groups. PRC and final D worsened in the cross-linked and progressive uncross-linked groups, and did not change in the non-progressive group.

Conclusion: Despite a decreased K_{max}, the posterior corneal surface parameters, posterior elevation values were determined to have significantly worsened in the cross-linked group and this increase was higher than in progressive uncross-linked eyes.

Introduction

Keratoconus is a bilateral progressive degenerative corneal ectatic disease with stromal collagen matrix alterations that lead to stromal thinning and irregular protrusion of the cornea[1]. This irregular cornea creates an irregular astigmatism deteriorating visual acuity to counting fingers in advanced cases.

Corneal cross-linking (CCL) is the only promising method that can halt the progression of keratoconus[2]. Corneal cross-linking is a technique that uses a combination of riboflavin and ultraviolet-A (UVA) radiation to create covalent bonds within and between both collagen molecules and proteoglycan core proteins. These cross-links aim to restore the biomechanical strength and stability of the cornea[3]. Wollensak et al. developed a standard CCL protocol in 2003[3], while accelerated protocols have been introduced since 2010 to achieve some advantages such as shortened operation time and reduced rate of complications[4]. Accelerated protocols are carried out in 3, 5, and 10 minutes using 30, 18, and 9 mW/cm² irradiance, respectively without changing the cumulative irradiation dose of 5.4 J/cm². In the last meta-analysis of randomized controlled trials by Kobashi et al., accelerated CCL showed a comparable efficacy and safety profile and both methods similarly stopped the disease progression[5].

There are many studies in the literature investigating the outcomes of CCL. The parameters most evaluated are maximum keratometry, central corneal thickness, visual acuity, spherical equivalent, and corneal biomechanical properties. Few studies have examined posterior corneal parameters and they have mainly investigated the effects of standard CCL[6–8]. As far as the authors are aware, there is no study in the literature evaluating the long-term effects of accelerated CCL on all posterior corneal surface parameters.

The aim of this study was to evaluate the long-term effects of accelerated CCL (10 minutes, 9 mW/cm^2) on posterior corneal surface parameters and to compare the posterior corneal surface Scheimpflug tomography measurement changes between cross-linked and uncross-linked keratoconic eyes.

Materials And Methods

Patients

This retrospective observational study was conducted in compliance with the institutional and government review board regulations and informed consent regulations. The study protocol was approved by the Ankara Research and Training Hospital Local Ethics Committee with the number E-2572, and the study was carried out in accordance with the Declaration of Helsinki. Written informed consent was obtained from all participants before performing CCL.

From the large database in the corneal unit, keratoconus patients were reviewed individually. Keratoconus diagnosis was based on the characteristic keratoconus signs in the anterior sagittal curvature maps (eg, an asymmetric bowtie pattern with or without skewed axes or inferior or central steeping) and at least one biomicroscopic sign (eg, a conical protrusion, Vogt striae, and Fleischer ring). Patients were excluded from the study if they were aged < 18 years, had anterior segment surgery history, corneal scarring, corneal haze > grade 1, ocular surface problems, or if they had undergone repeated CCL. Progression was defined as a consistent change in at least 2 of the following 3 parameters where the magnitude of the change was above the normal noise of the testing system: (1) progressive steepening of the anterior corneal surface, (2) progressive steepening of the posterior corneal surface, and (3) progressive thinning and/or an increase in the rate of corneal thickness change from the periphery to the thinnest point[9].

First, 35 eyes of 35 progressive keratoconus patients who underwent CCL treatment and were followed up for at least 36 months after CCL were included as the cross-linked group. To make a comparison, the data of 30 eyes of 30 progressive keratoconus patients who showed progression in the 12th month of follow-up constituted the progressive uncross-linked group, and 30 eyes of 30 patients who did not show progression formed the non-progressive uncross-linked group.

Ccl Procedure

All CCL procedures were performed in an operating room under topical anesthesia with 0.5% proparacaine hydrochloride eye drops. The corneal epithelium was mechanically removed from the 8.0 mm treatment zone and then ultrasound pachymetry was applied (Sonomed 300P PacScan Pachymeter; Escalon Medical Corp., USA). If the corneal thickness was < 400 µm, hypoosmolar riboflavin was instilled until the cornea had swollen to a thickness exceeding 400 µm. When the corneal thickness exceeds 400 µm, iso-osmolar riboflavin solution (0.1% riboflavin in 20% dextran T500 solution, Meran Medicine, BNM Inc., Turkey) was instilled to the cornea every 2 minutes, for a total of 30 minutes. The cornea was exposed to 370 nm UVA light with a 5.4 J/cm² surface dosage using a commercially available UVA system (UV-X System, Peschke Meditrade GmbH, Huenenberg, Switzerland). The intended 9 mW/cm² surface irradiance was applied for 10 minutes while the riboflavin solution continued to be instilled. The cornea was irrigated with cold water at the end of the procedure, and a silicon-hydrogel bandage contact lens (Acuvue Oasys, Johnson&Johnson, base curve 8.4 mm) was applied. Postoperative treatment included moxifloxacin hydrochloride 0.5% four times a day for one week, fluorometholone 5% eye drops four times a day on a tapering schedule for two weeks, and artificial tears four times a day for one month. The bandage contact lens was removed on the fifth postoperative day.

Ophthalmological Examination

Patients underwent a complete ophthalmological examination, including best spectacle corrected visual acuity (BSCVA), slit-lamp biomicroscopy, fundus evaluation and tomographic analysis with a rotating Scheimpflug corneal tomography device (Pentacam HR, Oculus Optikgeräte GmbH, Wetzlar, Germany). The BSCVA was measured with a Snellen chart by a cornea specialist and converted to Logarithm of the Minimum Angle of Resolution (logMAR) for statistical analysis. The examination and measurement data of all the patients were recorded according to the study design.

The Pentacam HR is a non-invasive tomography device with a single rotating Scheimpflug camera. This device uses a 1.45-megapixel camera to maximally capture 138.000 data points of true elevation and a 475-nm ultraviolet-free blue light-emitting diode light for corneal illumination. All measurements were taken by an expert examiner under scotopic conditions with the natural pupil. Each participant was asked to fixate on an internal target after a complete blink, and the joystick was adjusted until perfect alignment was shown. Acceptable maps had at least 10 mm of corneal coverage without any extrapolated data in the central 8.0 mm zone. Scans not meeting acceptable criteria were repeated.

The most commonly used CCL efficacy indicators of maximum keratometry (K_{max}) and thinnest corneal thickness (TCT) were noted. Minimum radius of curvature (R_{min}back) was noted as the point of maximum posterior corneal curvature. Asphericity (Q_{back}) and the most commonly used higher order aberrations of spherical aberration, vertical and horizontal coma, vertical and oblique trefoil, which pertain to the corneal back surface, were noted.

Corneal light backscatter measurements were recorded via the densitometry software. This software automatically locates the corneal apex and analyzes a surrounding area of 12 mm in diameter. This area is divided into four concentric zones which are 0–2 mm circular zone and 2–6 mm, 6–10 mm, and 10–12 mm annular zones. The analyses also provide densitometric values at three different depths. The anterior layer is the superficial 120 μm region, the posterior layer is the innermost 60 μm region, and the central corneal layer lies between these two layers. For this study, the posterior corneal layer values in all concentric zones were noted. Corneal densitometry values are expressed as the pixel luminance per unit volume in grayscale units. Measurements range from 0 (maximum transparency) to 100 (totally opaque) with regard to the backscattering light degree.

The Belin-Ambrósio Enhanced Ectasia Display (BAD-III) software evaluates the pachymetric progression and the anterior and posterior elevation values of the cornea. The maximum posterior elevation value at the 5.0 mm central cornea ($MaxPost_{elev}$) and at the thinnest point ($ThinnestPost_{elev}$) were examined according to the best fit sphere (BFS) reference and to the enhanced BFS reference surface. The deviation of normality of the back elevation (Db) and overall deviation of normality (final D) were also noted from the BAD-III software.

The Belin ABCD keratoconus grading system is incorporated in the Pentacam software as part of the topometric/keratoconus grading display. This system uses the anterior and posterior radius of curvature (ARC and PRC) taken from the 3.0-mm zone centered on the thinnest point. The classification parameters are ‘A’ for ARC, ‘B’ for PRC, ‘C’ for thinnest pachymetry, and ‘D’ for distance visual acuity. The PRC and ‘B’ parameters were recorded from this grading system.

Statistical Analysis

Statistical analysis was performed using SPSS for Windows software (version 22.0, IBM Corp.). Conformity of the data to normal distribution was analyzed with the Kolmogorov-Smirnov test. Descriptive statistics were recorded as mean ± standard deviation (SD) values. Longitudinal analyses were performed with repeated-measures analysis of variance (ANOVA) and the Bonferroni post-hoc test. The assumption of sphericity was tested with Mauchly’s test of sphericity. When the significance level was greater than the a priori α level ($p > 0.05$), sphericity was assumed and the value from the univariate test table was used. When the significance level was less than or equal to the a priori α level ($p \leq 0.05$), sphericity could not be assumed and Wilk’s λ test value from the multivariate test table was used. Paired t-tests were performed to determine whether the difference between two measurements of the same eye was significant. Between-group comparisons of the three groups were performed using the one-way ANOVA test. When the overall ANOVA model was significant, the Bonferroni post-hoc test was conducted to determine the pairwise comparison of the means that were significantly different. A value of $p < 0.05$ was considered statistically significant.

Results

The mean age of the patients was 22.0 ± 3.55 years in the cross-linked group, 23.29 ± 7.36 years in the progressive uncross-linked group and 32.02 ± 7.55 years in the non-progressive uncross-linked group.

Visual Acuity, Topography And Aberrometry Changes After Ccl

Table 1 shows the visual and corneal topographic and aberrometric parameters at baseline and 12, 24, and 36 months after CCL. BSCVA significantly improved at 36 months after CCL. BSCVA at post-CCL 12 months was significantly better than at baseline in all post-CCL measurements. K_{max} and TCT were significantly decreased at 36 months after CCL. The Bonferroni posthoc test revealed that all postoperative values were significantly lower than baseline values. The R_{min} and Q values of the back cornea did not significantly change after CCL. None of the higher-order aberrations of the corneal back surface changed at 36 months after CCL.

Table 1
Visual and topographic and aberrometric parameters over time in the cross-linked group

Variables	Preoperative	Postoperative 12. month	Postoperative 24. month	Postoperative 36. Month	p value*	p value**
BSCVA (logMAR)	0.51 ± 0.31	0.22 ± 0.14	0.44 ± 0.50	0.46 ± 0.65	**< 0.001**	**< 0.001** 1.00 1.00
K_{max} (D)	56.63 ± 4.96	54.66 ± 5.12	53.82 ± 4.64	53.71 ± 4.87	**< 0.001**	**< 0.001** **< 0.001** **< 0.001**
TCT (µm)	463.33 ± 41.48	427.93 ± 47.84	427.06 ± 51.37	426.03 ± 53.35	**< 0.001**	**< 0.001** **< 0.001** **< 0.001**
R_{min}back (mm)	4.33 ± 0.45	4.23 ± 0.45	4.27 ± 0.44	4.27 ± 0.43	0.14	
Q_{back}	-0.99 ± 0.27	-1.06 ± 0.30	-1.02 ± 0.26	-1.02 ± 0.30	0.49	
Spherical aberration (µm)	0.09 ± 0.12	0.12 ± 0.15	0.11 ± 0.13	0.11 ± 0.14	0.29	
Vertical coma (µm)	-0.08 ± 0.07	-0.09 ± 0.07	-0.07 ± 0.06	-0.07 ± 0.05	0.28	
Horizontal coma (µm)	-0.008 ± 0.05	0.012 ± 0.06	0.013 ± 0.06	0.01 ± 0.05	0.73	
Vertical trefoil (µm)	-0.0004 ± 0.02	-0.006 ± 0.05	0.007 ± 0.04	0.010 ± 0.04	0.12	
Oblique trefoil(µm)	0.03 ± 0.03	0.03 ± 0.06	0.02 ± 0.05	0.02 ± 0.04	0.47	

BSCVA:best spectacle corrected distance visual acuity, K_{max} : maximum keratometry, TCT: thinnest corneal thickness, R_{min}back: smallest radius of curvature at the back of the cornea, Q_{back}: asphericity value at the back of the cornea

* repeated-measures analysis of variance

** bonferroni post-test analysis between baseline and 12,24, and 36 months respectively after crosslinking

Corneal Densitometry Changes After Ccl

Table 2 shows the posterior corneal densitometric parameters at baseline and at 12, 24, and 36 months after CCL. Densitometric values in all the zones and total posterior layer significantly improved at 36 months after CCL. The Bonferroni posthoc test revealed that the densitometric values were significantly different between baseline and post-CCL 12th month in the 0–2 and 10–12 mm zones, and significantly different between baseline and post-CCL 24th month in the 6–10 mm zone.

Table 2
Posterior corneal densitometric parameters over time in the cross-linked group

Variables	Baseline	Postoperative 12.month	Postoperative 24.month	Postoperative 36.month	p value*	p value**
0–2 mm (GSU)	8.48 ± 1.17	9.64 ± 1.79	8.43 ± 1.40	8.45 ± 1.87	**< 0,001**	**0.01** 1.00 1.00
2–6 mm (GSU)	9.09 ± 0.85	9.35 ± 1.06	8.55 ± 0.89	8.87 ± 1.28	**< 0.001**	1.00 0.17 1.00
6–10 mm (GSU)	8.79 ± 0.79	8.53 ± 0.97	8.11 ± 0.95	8.34 ± 1.21	**0.008**	1.00 **0.01** 0.42
10–12 mm (GSU)	13.37 ± 3.61	11.88 ± 2.31	12.24 ± 2.81	11.73 ± 2.56	**0.04**	**0.04** 0.56 0.06
Total (GSU)	9.53 ± 0.85	9.53 ± 0.98	8.95 ± 0.98	9.18 ± 1.35	**0.01**	1.00 0.08 1.00
GSU:gray scale units						
* repeated-measures analysis of variance						
** bonferroni post-test analysis between baseline and 12,24, and 36 months respectively after crosslinking						

Posterior Surface Elevation Changes After Ccl

Table 3 shows the posterior corneal surface elevation and ectasia indices at baseline and at 12, 24, and 36 months after CCL. The maximum and thinnest point posterior elevation values, Db, and final D parameters significantly increased after CCL. The Bonferroni posthoc test analysis revealed that Db value was significantly higher than baseline at post-CCL 24 and 36 months, and the final D value was higher than baseline at the 12th and 24th months. The PRC value significantly decreased and its Belin unit 'B' significantly increased after CCL. Bonferroni posthoc test analysis revealed significantly lower PRC and higher 'B' at the postoperative 24th month compared to baseline values.

Table 3
Posterior corneal surface elevation and ectasia indices parameters over time in the cross-linked group

Variables	**Baseline**	**Postoperative 12.month**	**Postoperative 24.month**	**Postoperative 36.month**	**p value***	**p value****
$MaxPost_{elev}$ (μm)	64.10 ± 18.24	70.36 ± 18.31	71.13 ± 19.01	72.10 ± 20.26	**0.01**	0.05 0.09 0.08
Db	8.02 ± 3.70	9.10 ± 3.62	9.29 ± 3.39	9.24 ± 3.74	**0.03**	0.06 **0.03** **0.01**
Final D	8.33 ± 2.52	9.44 ± 2.54	9.44 ± 2.74	9.11 ± 3.24	**0.005**	**0.003** **0.005** 0.66
$ThinnestPost_{elev}$ (μm)	46.10 ± 19.36	51.70 ± 18.47	52.73 ± 24.79	52.50 ± 23.49	**0.04**	0.05 0.07 0.05
PRC (mm)	4.98 ± 0.42	4.88 ± 0.43	4.83 ± 0.40	4.88 ± 0.48	**0.001**	0.09 **0.007** 0.19
B value (Belin unit for PRC)	4.12 ± 1.71	4.52 ± 1.83	4.75 ± 1.71	4.59 ± 1.94	**0.012**	0.14 **0.006** 0.10

$MaxPost_{elev}$: maximum posterior elevation at the 5.0 mm central cornea Db: deviation of normality of the back elevation $ThinnestPost_{elev}$: maximum posterior elevation at the thinnest point PRC: Posterior radius of curvature

* repeated-measures analysis of variance

** bonferroni post-test analysis between baseline and 12,24, and 36 months respectively after crosslinking

Comparison of Visual acuity, Topography and Aberrometry changes in the three groups

Table 4 shows the visual acuity and topography and aberrometry changes at the 12-month follow-up examination of the three groups. BSCVA did not significantly change in the uncross-linked group and improved in the cross-linked group ($p < 0.001$). K_{max} significantly decreased in the cross-linked group and significantly increased in the progressive uncross-linked group ($p < 0.001$). TCT significantly decreased in both the cross-linked and progressive uncross-linked groups ($p < 0.001$) and the decrease was greater in the cross-linked group ($p < 0.001$). R_{min} and Q back value changes were similar in all the groups. Table 5 shows the wavefront cornea back aberrometric parameter changes at the 12-month follow-up examination in all three groups. The aberrometry values did not change at 12 months in all the groups and the change values did not differ between the groups ($p > 0.05$ for all values).

Table 4
Visual and topographic parameters at baseline and at 12 months in all the groups

Variables	Group	Baseline	12. month follow-up	Mean change	p value*	p value**	p value†
BSCVA (logMAR)	Cross-linked	0.51 ± 0.31	0.22 ± 0.14	-0.28 ± 0.31	**< 0.001**	**< 0.001**	**< 0.001** **< 0.001** 1.00
	Progressive uncross-linked	0.18 ± 0.19	0.18 ± 0.20	0.001 ± 0.11	0.93		
	Nonprogressive uncross-linked	0.17 ± 0.21	0.17 ± 0.20	-0.004 ± 0.02	0.32		
K_{max} (D)	Cross-linked	56.63 ± 4.96	54.66 ± 5.12	-1.96 ± 2.19	**< 0.001**	**< 0.001**	**< 0.001** **< 0.001** **< 0.001**
	Progressive uncross-linked	54.14 ± 4.90	55.73 ± 5.17	1.54 ± 1.80	**< 0.001**		
	Nonprogressive uncross-linked	52.80 ± 5.24	52.69 ± 5.01	-0.11 ± 0.61	0.24		
TCT (μm)	Cross-linked	463.33 ± 41.48	427.93 ± 47.84	-35.40 ± 26.40	**< 0.001**	**< 0.001**	**< 0.001** **< 0.001** 0.05
	Progressive uncross-linked	448.40 ± 91.60	439.53 ± 90.97	-8.96 ± 14.74	**0.001**		

BSCVA:best spectacle corrected distance visual acuity, K_{max} : maximum keratometry, TCT: thinnest corneal thickness, R_{min}back: smallest radius of curvature at the back of the cornea, Q_{back}: asphericity value at the back of the cornea

* Paired t-test analysis for postoperative 12. month and baseline comparison

** One-way ANOVA test analysis for comparison of the mean change values between groups

† Bonferroni posttest analysis between cross-linked and progressive uncross-linked, cross-linked and nonprogressive uncross-linked, progressive uncross-linked and nonprogressive uncross-linked groups, respectively

Variables	Group	Baseline	12. month follow-up	Mean change	p value*	p value**	p value†
	Nonprogressive uncross-linked	449.55 ± 50.58	450.91 ± 50.56	1.17 ± 8.46	0.30		
R_{min} back	Cross-linked	4.33 ± 0.45	4.23 ± 0.45	-0.10 ± 0.27	0.05	0.15	
	Progressive uncross-linked	4.53 ± 0.56	4.41 ± 0.50	-0.11 ± 0.20	**0.005**		
	Nonprogressive uncross-linked	4.86 ± 0.58	4.84 ± 0.58	-0.01 ± 0.11	0.39		
Q back	Cross-linked	-0.99 ± 0.27	-1.06 ± 0.30	-0.06 ± 0.21	0.12	0.19	
	Progressive uncross-linked	-0.85 ± 0.43	-0.89 ± 0.43	-0.02 ± 0.11	0.09		
	Nonprogressive uncross-linked	-0.74 ± 0.37	-0.74 ± 0.38	0.02 ± 0.22	1.00		

BSCVA:best spectacle corrected distance visual acuity, K_{max} : maximum keratometry, TCT: thinnest corneal thickness, R_{min}back: smallest radius of curvature at the back of the cornea, Q_{back}: asphericity value at the back of the cornea

* Paired t-test analysis for postoperative 12. month and baseline comparison

** One-way ANOVA test analysis for comparison of the mean change values between groups

† Bonferroni posttest analysis between cross-linked and progressive uncross-linked, cross-linked and nonprogressive uncross-linked, progressive uncross-linked and nonprogressive uncross-linked groups, respectively

Table 5
Wavefront cornea back aberrometric parameters at baseline and at 12 months in all the groups

Variables	Group	Baseline	12. month follow-up	Mean change	p value*	p value**
Spherical aberration	Cross-linked	0.09 ± 0.12	0.12 ± 0.15	0.02 ± 0.10	0.16	0.35
	Progressive uncross-linked	0.04 ± 0.16	0.06 ± 0.17	0.01 ± 0.06	0.19	
	Nonprogressive uncross-linked	-0.01 ± 0.13	-0.01 ± 0.12	0.0007 ± 0.01	0.84	
Vertical coma	Cross-linked	-0.08 ± 0.07	-0.09 ± 0.07	-0.005 ± 0.05	0.57	0.34
	Progressive uncross-linked	0.51 ± 0.23	0.53 ± 0.25	0.01 ± 0.10	0.32	
	Nonprogressive uncross-linked	0.35 ± 0.24	0.37 ± 0.25	0.01 ± 0.03	0.05	
Horizontal coma	Cross-linked	0.008 ± 0.05	0.01 ± 0.06	0.003 ± 0.02	0.39	0.43
	Progressive uncross-linked	-0.03 ± 0.24	-0.06 ± 0.26	-0.02 ± 0.16	0.41	
	Nonprogressive uncross-linked	0.006 ± 0.13	0.009 ± 0.13	0.003 ± 0.02	0.55	
Vertical trefoil	Cross-linked	-0.0004 ± 0.02	-0.006 ± 0.05	-0.005 ± 0.04	0.49	0.15
	Progressive uncross-linked	-0.02 ± 0.17	0.01 ± 0.18	0.04 ± 0.15	0.13	
	Nonprogressive uncross-linked	0.03 ± 0.12	0.03 ± 0.10	0.0009 ± 0.08	0.95	
Oblique trefoil	Cross-linked	0.03 ± 0.03	0.03 ± 0.06	0.002 ± 0.05	0.81	0.68
	Progressive uncross-linked	-0.14 ± 0.13	-0.17 ± 0.18	-0.02 ± 0.14	0.41	
	Nonprogressive uncross-linked	-0.15 ± 0.12	-0.15 ± 0.16	-0.004 ± 0.11	0.85	

* Paired t-test analysis for postoperative 12. month and baseline comparison

** One-way ANOVA test analysis for comparison of the mean change values between groups

Comparison of Posterior Corneal Densitometry changes in the three groups

Table 6 shows the posterior corneal densitometry changes at the 12-month follow-up examination in the three groups. A significant posterior densitometry increase only occurred in the 0–2 mm zone in the cross-linked group ($p = 0.002$) and the amount of change created a significant difference between the groups ($p = 0.01$). The uncross-linked groups did not show significant densitometry changes at 12 months.

Table 6
Posterior corneal densitometric parameters at baseline and at 12 months in all the groups

Variables	Group	Baseline	12. month follow-up	Mean change	p value*	p value**	p value†
0–2 mm (GSU)	Cross-linked	8.48 ± 1.17	9.64 ± 1.79	1.16 ± 1.89	**0.002**	**0.01**	0.46 **0.008** 0.30
	Progressive uncross-linked	8.35 ± 1.12	8.87 ± 1.59	0.51 ± 1.65	0.09		
	Nonprogressive uncross-linked	8.23 ± 1.50	8.00 ± 0.98	-0.23 ± 1.69	0.45		
2–6 mm (GSU)	Cross-linked	9.09 ± 0.85	9.35 ± 1.06	0.26 ± 1.41	0.31	0.16	
	Progressive uncross-linked	8.76 ± 0.98	9.22 ± 1.21	0.45 ± 1.43	0.09		
	Nonprogressive uncross-linked	8.43 ± 1.55	8.18 ± 0.93	-0.24 ± 1.54	0.39		
6–10 mm (GSU)	Cross-linked	8.79 ± 0.79	8.53 ± 0.97	-0.26 ± 1.41	0.21	0.40	
	Progressive uncross-linked	8.84 ± 1.50	9.00 ± 1.45	0.15 ± 1.30	0.51		
	Nonprogressive uncross-linked	9.05 ± 1.75	8.83 ± 1.39	-0.22 ± 1.48	0.42		
10–12 mm (GSU)	Cross-linked	13.37 ± 3.61	11.88 ± 2.31	-1.49 ± 2.86	**0.008**	**0.04**	0.05 0.19 1.00
	Progressive uncross-linked	13.31 ± 4.83	13.44 ± 4.24	0.13 ± 2.52	0.78		
	Nonprogressive uncross-linked	13.07 ± 3.04	12.83 ± 2.06	-0.24 ± 2.31	0.56		
Total (GSU)	Cross-linked	9.53 ± 0.85	9.53 ± 0.98	0.003 ± 1.26	0.98	0.28	

* Paired t-test analysis for postoperative 12. month and baseline comparison

** One-way ANOVA test analysis for comparison of the mean change values between groups

† Bonferroni posttest analysis between cross-linked and progressive uncross-linked, cross-linked and nonprogressive uncross-linked, progressive uncross-linked and nonprogressive uncross-linked groups, respectively

Variables	Group	Baseline	12. month follow-up	Mean change	p value*	p value**	p value†
	Progressive uncross-linked	9.35 ± 1.22	9.69 ± 1.23	0.33 ± 1.36	0.18		
	Nonprogressive uncross-linked	9.35 ± 1.69	9.12 ± 1.05	-0.23 ± 1.55	0.41		

* Paired t-test analysis for postoperative 12. month and baseline comparison

** One-way ANOVA test analysis for comparison of the mean change values between groups

† Bonferroni posttest analysis between cross-linked and progressive uncross-linked, cross-linked and nonprogressive uncross-linked, progressive uncross-linked and nonprogressive uncross-linked groups, respectively

Comparison of Posterior Corneal Surface Elevation and Ectasia Indices changes in the three groups

Table 7 shows the posterior corneal surface elevation and ectasia indices changes at the 12-month follow-up examination in the three groups. Maximum posterior elevation at the 5.0 mm central cornea and at the thinnest point, and the deviation of normality of the back elevation values significantly increased in the cross-linked group, and did not significantly change in the uncross-linked groups ($p = 0.01$, $p = 0.01$, $p = 0.09$, respectively). Overall deviation of normality significantly increased and posterior radius of curvature significantly decreased in the cross-linked and progressive uncross-linked groups, and did not change in the non-progressive uncross-linked group ($p < 0.001$ and $p = 0.008$). The amount of change was higher in the cross-linked group than in the progressive uncross-linked group for both final D and Db. The B value significantly increased in the cross-linked group and did not significantly change in the uncross-linked groups ($p = 0.01$).

Table 7
Posterior corneal surface elevation and ectasia indices parameters at baseline and 12. months follow-up in all groups

Variables	Group	Baseline	12. month follow-up	Mean change	p value*	p value**	p value†
$MaxPost_{elev}$ (μm)	Cross-linked	64.10 ± 18.24	70.36 ± 18.31	6.26 ± 12.15	**0.008**	**0.01**	0.06 **0.02** 1.00
	Progressive uncross-linked	55.53 ± 21.23	56.03 ± 18.94	0.50 ± 10.45	0.79		
	Nonprogressive uncross-linked	42.03 ± 16.33	41.50 ± 17.26	-0.53 ± 3.67	0.43		
Db	Cross-linked	8.02 ± 3.70	9.10 ± 3.62	1.07 ± 2.18	**0.01**	0.09	
	Progressive uncross-linked	7.42 ± 4.73	25.36 ± 7.71	0.28 ± 1.72	0.37		
	Nonprogressive uncross-linked	5.06 ± 3.39	5.27 ± 3.36	0.20 ± 1.02	0.28		
Final D	Cross-linked	8.33 ± 2.52	9.44 ± 2.54	1.11 ± 1.55	**< 0.001**	**< 0.001**	0.06 **< 0.001** 0.05
	Progressive uncross-linked	7.77 ± 3.16	8.28 ± 3.35	0.50 ± 1.00	**0.004**		
	Nonprogressive uncross-linked	6.96 ± 2.86	6.91 ± 2.97	-0.05 ± 0.55	0.54		
$ThinnestPost_{elev}$ (μm)	Cross-linked	46.10 ± 19.36	51.70 ± 18.47	5.60 ± 10.89	**0.009**	**0.01**	0.09 **0.01** 1.00

* Paired t-test analysis for postoperative 12. month and baseline comparison
** One-way ANOVA test analysis for comparison of the mean change values between groups
† Bonferroni posttest analysis between cross-linked and progressive uncross-linked, cross-linked and nonprogressive uncross-linked, progressive uncross-linked and nonprogressive uncross-linked groups, respectively

Variables	Group	Baseline	12. month follow-up	Mean change	p value*	p value**	p value†
	Progressive uncross-linked	46.76 ± 19.74	47.43 ± 21.79	0.66 ± 9.59	0.70		
	Nonprogressive uncross-linked	36.20 ± 13.91	35.20 ± 14.84	-1.00 ± 4.16	0.19		
PRC (mm)	Cross-linked	4.98 ± 0.42	4.88 ± 0.43	-0.10 ± 0.21	**0.01**	**0.008**	0.66 **0.007** 0.30
	Progressive uncross-linked	5.06 ± 0.48	4.99 ± 0.49	-0.05 ± 0.15	**0.02**		
	Nonprogressive uncross-linked	5.22 ± 0.47	5.23 ± 0.47	0.01 ± 0.08	0.35		
B value (Belin unit for PRC)	Cross-linked	4.12 ± 1.71	4.52 ± 1.83	0.40 ± 0.93	**0.02**	**0.01**	0.70 **0.01** 0.41
	Progressive uncross-linked	3.87 ± 1.93	4.10 ± 2.03	0.19 ± 0.68	0.05		
	Nonprogressive uncross-linked	3.25 ± 1.74	3.21 ± 1.75	-0.04 ± 0.32	0.39		
* Paired t-test analysis for postoperative 12. month and baseline comparison							
** One-way ANOVA test analysis for comparison of the mean change values between groups							
† Bonferroni posttest analysis between cross-linked and progressive uncross-linked, cross-linked and nonprogressive uncross-linked, progressive uncross-linked and nonprogressive uncross-linked groups, respectively							

Discussion

The posterior corneal surface evaluation first began with the measurement of the radius of curvature and central corneal thickness using some techniques such as purkinje image photography and pachymetry [10–11]. Orbscan scanning slit topography (Bausch&Lomb, Orbtek Inc., Salt Lake City, UT) and later Pentacam Scheimpflug imaging provided more detailed information about the posterior surface than had been previously possible. The reliability and the validity of the posterior cornea measurements with Orbscan have been questioned in many cases and it has been concluded that Orbscan posterior cornea results may be an artifact [12–13]. Scheimpflug imaging is capable of measuring 25,000 true elevation points and may detect posterior corneal elevations with mathematical reconstruction [14]. Scheimpflugderived posterior elevation measurements have acceptable reproducibility in both keratoconic and cross-linked keratoconic eyes [15].

There are many studies in the literature which have investigated the changes in BSCVA, K_{max}, and TCT parameters after CCL. The current study results are consistent with those of recent studies that have reported improved K_{max}, BSCVA and decreased TCT values after accelerated CCL [16–18]. Visual acuity changes in the current study were not significant in the uncross-linked groups. K_{max} increased and TCT decreased in the progressive uncross-linked group as a consequence of progression. Corneal thickness was measured thinner in the cross-linked group than in the progressive uncross-linked group despite a flatter anterior surface. In a recent study by the current authors, the CCT measurement difference between the Pentacam and ultrasound pachymetry was largest in the CCL group, followed by the keratoconus and control groups (-20.9 ± 21.5, -10.6 ± 20.3, and 0.4 ± 6.8 μm), and it was concluded that pachymetric measurements from Scheimpflug must be interpreted with extreme caution [19].

The decrease in posterior surface asphericity and minimum radius of curvature was not significant in the long term, and the amounts of change were similar in the cross-linked and progressive uncross-linked groups at postoperative 12 months. In a comparative study of changes of corneal curvatures at 1 year post-CCL by Safarzadeh et al, curvatures of the posterior corneal surface were reported to be significantly increased [8]. This study can be considered to add to the literature that CCL does not have a positive or negative effect on back surface curvatures while it significantly improves anterior surface curvatures at a 36-month longitudinal follow-up.

Posterior HOAs are not calculated by extracting the anterior HOAs from the entire corneal aberrations because the incident rays on the posterior surface have a deformed wavefront caused by anterior surface refraction. Therefore, evaluation of both anterior and posterior corneal surface HOAs allows increased understanding of the optical quality of keratoconic eyes [20]. In studies by Hassan and Iselin et al., aberrations of the posterior corneal surface did not change at 7 and 24 months after CCL [6, 21]. In the present study, higher-order aberrations of the posterior corneal surface did not significantly change in a post-CCL follow-up period of 36 months similar to the study by Iselin et al. Greenstein et al. compared HOA changes at 1 year post-CCL between crosslinked and control ectatic eyes, and concluded that there was a mean decrease in posterior corneal HOAs in the treatment group while there was a mean increase in the control group [22]. In contrast to that study, the current study results showed that CCL did not add additional benefit to posterior corneal HOAs.

Compromised regularity of cells in keratoconic eyes increases the densitometry values of these corneas. With this knowledge, corneal densitometry changes have been investigated as an outcome of CCL. Greenstein et al. reported that the mean densitometry values peak at 1 month after CCL, but decreased throughout 12 months, and did not completely return to the baseline value at the postoperative 12th month [23]. Alnawaiseh et al. found that densitometry values including the posterior 0–2 and 2–6 mm zones were stable at 12 months post-CCL, and decreased below the preoperative values at the post-CCL 24th month [24]. Similarly in the current study, posterior densitometry values in the 0–2 and 2–6 mm zones were higher in the 12th month and lower in the 36th month compared to the baseline values. In the 12th month comparison between the groups, corneal densitometry increased in both the cross-linked and progressive uncross-linked groups. Although the densitometric change at the 12th month resembled ongoing ectatic change, posterior surface backscattering values improved 36 months after CCL.

The definition of progression in the 'Global consensus on keratoconus and ectatic disease' includes progressive steepening of the posterior corneal surface as one of the three parameters [9], so posterior surface evaluation is as important as anterior corneal surface. In a study by Kırgız et al., it was found that apical keratoscopy back increased 6 months after CCL and was associated with increased visual acuity [16]. Hashemi et al. reported that the decrease in posterior R_{min} and the increase in posterior elevation values were significant at 4 years after 5-minute accelerated CCL while the changes were not significant after standard CCL [25]. Tian et al. found that posterior elevation values slightly decreased 1 year after accelerated transepithelial CCL, but the parameters were significantly increased at the 3-year follow up [26]. It has been concluded that the posterior surface of the cornea may not be adequately cross-linked with accelerated CCL. In the current study, posterior surface elevation parameters significantly worsened at 36 months after CCL. In addition, the posterior elevation value changes were higher in the cross-linked group than in the progressive uncross-linked group. This suggested that posterior elevation increases could not be explained solely by ongoing ectatic changes.

There were considered to be three most likely reasons for the worsened posterior surface elevation measurements found after CCL. The first was that ectatic changes were ongoing due to inadequate cross-linking to the posterior cornea but this is unlikely due to the large difference between the cross-linked and progressive uncross-linked groups. The second possible reason was that tissue shrinkage and thinning induced by cross-linking may subsequently rearrange the corneal sequence, so more anterior and more elevated corneal regions may be measured at the post-CCL visits compared to baseline. The third reason could be that it is a result of a technical limitation of topography devices. Corneal topographers that measure the anterior and posterior corneal surface with optical methods depend on the pathway of the reflected waves of light and need a clear cornea structure for precise measurements [27]. For example, thinner corneal thickness values after CCL may be found as a result of a change in the refractive index of the stroma leading to false thinner results with optical ray tracing. Unreliable posterior surface measurements because of refractive index changes and demarcation line artifacts may be another reason for highly elevated posterior surface values.

There were some limitations to this study related to the retrospective design of the study. The primary limitation is the relatively small sample size but a large database was checked to identify patients with regular follow up of at least 36 months after CCL and the quality of measurements was also checked. A second limitation is that there was no comparison with a standard CCL protocol because heavily accelerated CCL is performed in our busy tertiary care hospital. The third limitation is that corneal changes could not be compared with different topography devices.

In conclusion, visual acuity improved, anterior corneal surface was flattened and posterior corneal surface was elevated at 36 months after accelerated CCL and the posterior corneal surface elevations were higher than than in the progressive uncross-linked eyes. Further studies are needed to reveal the exact cause of posterior surface elevation 36 months after CCL.

Declarations

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Data availability statement

Due to the nature of this research, participants of this study did not agree for their data to be shared publicly, so supporting data is not available.

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