

Outcome of Corneal Collagen Cross-Linking in Keratoconus: Introducing the Predictive Factors

Alireza Peyman¹, Awat Feizi², Mazdak Ganjalikhani-Hakemi³, Fahimeh Hosseini-Nasab³, Mohsen Pourazizi¹

¹Isfahan Eye Research Center, Department of Ophthalmology, Isfahan University of Medical Sciences, Isfahan, Iran, ²Department of Biostatistics and Epidemiology, School of Health, Isfahan University of Medical Sciences, Isfahan, Iran, ³Department of Immunology, School of Medicine, Isfahan University of Medical Sciences, Isfahan, Iran

Abstract

Purpose: To evaluate the multiple pretreatment characteristics and topographic factors of keratoconus (KC) patients and their relationship to clinical outcomes of corneal collagen cross-linking (CXL).

Methods: In this prospective study, 61 patients (106 eyes) with KC as candidates for CXL were included. Demographic data including age, sex, place of birth and residence, atopic constitution, family history, rubbing history, sleep apnea, and blood group were collected via a structured checklist. Complete ophthalmologic examination and tear collection to assess tear interleukin 6 (IL-6) and tumor necrosis factor alpha (TNF- α) level were performed. Topometric parameters were evaluated using a rotating Scheimpflug topography device. Changes in best corrected visual acuity (BCVA) and maximum keratometry (K-max) were considered the main predicted variables. Predictive variables were analyzed by univariate and multivariate regression.

Results: The use of multivariate analysis changes in K-max was significantly associated with rubbing frequency (coefficient = 0.94, $P = 0.02$), blood group (coefficient = 4.52, $P = 0.005$), pretreatment corneal asphericity (coefficient = -3.99, $P \leq 0.001$), and pretreatment central keratoconus index (CKI) (coefficient = -55.38, $P = 0.001$). Regarding the changes in BCVA, the multivariate analysis showed a significant association with place of birth (coefficient = -0.08, $P = 0.03$), pretreatment BCVA (coefficient = -0.67, $P < 0.001$), pretreatment central corneal thickness (CCT) (coefficient = -0.005, $P = 0.04$), and pretreatment keratoconus index (KI) (coefficient = 0.53, $P = 0.04$). Other parameters assessed in the multivariable analysis did not appear to have an individual effect on treatment outcomes.

Conclusion: Our results demonstrated that blood group, rubbing of eye, place of birth, corneal asphericity, pretreatment BCVA, CKI, KI, and CCT were statistically associated with the outcome of KC following CXL.

Keywords: Cornea, Corneal topography, Cross-linking reagents, Interleukin 6, Keratoconus, Treatment outcome, Tumor necrosis factor alpha

Address for correspondence: Mohsen Pourazizi, Isfahan Eye Research Center, Department of Ophthalmology, Isfahan University of Medical Sciences, Isfahan, Iran. E-mail: m.pourazizi@yahoo.com

Submitted: 04-Aug-2019; **Revised:** 22-Oct-2019; **Accepted:** 10-Nov-2019; **Published:** 23-Mar-2020

INTRODUCTION

The exact mechanism of keratoconus (KC) is not well understood, but it is commonly accepted that both genetic susceptibility and environmental factors are necessary.¹

Factors associated with KC include a positive family history, atopic constitution, eye rubbing, contact lens use, genetic syndromes such as Down, sleep apnea, blood group, and place of living.^{2,3}

Studies have demonstrated that various cytokines are elevated in KC; these inflammatory markers are interleukin 6 (IL-6), tumor necrosis factor alpha (TNF- α), and matrix metalloproteinase 9 (MMP9).⁴ Among the treatment modalities for KC, corneal collagen cross-linking (CXL) is a promising treatment that may slow or stop the progression of KC and improve subjective and

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How to cite this article: Peyman A, Feizi A, Ganjalikhani-Hakemi M, Hosseini-Nasab F, Pourazizi M. Outcome of corneal collagen cross-linking in keratoconus: Introducing the predictive factors. *J Curr Ophthalmol* 2020;32:19-25.

Access this article online

Quick Response Code:



Website:
www.jcurrophthalmol.org

DOI:
10.4103/JOCO.JOCO_48_20

objective visual parameters.^{5,6} *In vitro* studies have shown that CXL leads to both biochemical and biomechanical changes in corneal tissue, so that they can modify the natural course of the disease.⁷ The principle goal of CXL is to stabilize the progression of diseases.⁸ On the one hand, successful CXL can prevent the progression of KC and can even cause the ectatic cornea to regress.⁶ On the other hand, worsening in the ocular parameters can occur as a complication of the procedure.⁴ Although some studies have reported significant improvements in topographic, visual, refractive, and aberrometric parameters following CXL, disease progression, severe visual disturbance, and loss of vision have been observed in some patients after CXL; therefore, the clinical results of CXL on improvement of KC progression are variable, and the clinical benefits of CXL can vary among patients.^{4,9-12} As a result, the ability to reliably predict the outcome of performing CXL before the procedure will help clinicians manage their patients' expectations and minimize the exposure to potential side effects. There are limited published data on variable results about factors associated with significant improvement, efficacy, and safety following CXL.^{10,13,14} These factors include preoperative visual acuity, eccentricity of the cone, pretreatment maximum keratometry (K-max), age, and sex.^{10,13} However, no reports on whether these factors play a role in the outcome of CXL independently or are considered a complication of the procedure have been established yet.⁴ There is no single study primarily investigating the association between the most important preoperative basic characteristic, functional, anatomical, and biochemical factors and CXL outcomes. Since the understanding of the factors associated with CXL treatment success is important and can be considered a clinical predictive factor demonstrating the importance of patient selections, here we have designed a study to investigate the value of the aforementioned factors and evaluate the outcome of standard CXL for KC considering medical histories, as well as clinical, ocular, and laboratory parameters. The results of our study with consideration to multiple factors might be useful for ophthalmologists to make the best therapeutic decision for their patients on possible predictive factors for CXL surgery outcome. In addition, clinicians can manage their patients regarding possible predictive factors for CXL surgery outcome.

METHODS

In this prospective and interventional clinical study, patients with KC as candidates for CXL treatment were included in the study at the refractive surgery clinic, Feiz Hospital, affiliated with Isfahan University of Medical Sciences (IUMS), Isfahan, Iran, from September 2018 to April 2019. The Institutional Review Board of IUMS approved the study according to the World Medical Association's code of ethics in accordance with the tenets of the Declaration of Helsinki revised in Brazil in 2013. Signed informed consent was obtained from all the participants after being clearly informed about the study design and setting. All the participants were free to withdraw from the study at any time without affecting their relationship with their

health-care provider. This study was registered in the Iranian Registry of Clinical Trials (IRCT) database (IRCT registration number: IRCT20131229015975N4).

The first author (A. P.) made the diagnosis of KC using Scheimpflug topography device (Pentacam HR; Oculus GmbH, Wetzlar, Germany) based on anterior and posterior elevation, pachymetric, and keratometry.⁸

The participants were surgical-naïve KC patients aged above 12 years who were candidates for CXL. Exclusion criteria were pregnancy, breast feeding, other corneal diseases (e.g., herpetic keratitis and corneal opacities), serious medical conditions, malignancy, hereditary connective tissue disorder (e.g., Marfan disease), collagen vascular disease, severe dry eye, rheumatologic diseases, and patients with poor compliance.

Withdrawal criteria included not showing up in follow-up visits and administration of another treatment protocol or medications by other physicians.

The study was designed in three parts. The first part consisted of data collected using a structured checklist, including demographic data such as age, sex, place of birth and residence, atopic constitution, family history, rubbing history, sleep apnea, and blood group. The second part included complete ophthalmologic examination and tears collection to assess tear IL-6 and TNF- α level. Ocular evaluation included assessment of uncorrected visual acuity, best corrected visual acuity (BCVA), refractive error, slit-lamp microscopic examination, tonometry, funduscopy, keratometric, and topometric parameters using a rotating Scheimpflug topography device (Pentacam HR; Oculus GmbH, Wetzlar, Germany). Topometric parameters included keratoconus index (KI), central keratoconus index (CKI), index of surface variance, index of height asymmetry, index of height decentration (IHD), and index of vertical asymmetry (IVA). The third part included surgical procedures. Evaluation was repeated 1 year after CXL.

Atopic disease is defined as clinical allergy symptoms such as atopic eczema, allergic rhinoconjunctivitis, and/or allergic asthma in combination with a positive allergy (skin prick test). Furthermore, atopic state is defined as a genetic predisposition to react by the skin prick test to common allergens, regardless of clinical symptoms.¹⁵ Positive family history is defined as the existence of documented KC in first-degree and second-degree relatives. The Persian version of Berlin Sleep Questionnaire was used to diagnose obstructive sleep apnea.¹⁶ Eye rubbing was evaluated using a 4-point Likert scale.¹⁷

Non-traumatic tear samples were collected using sterile methods without anesthetic drops or stimulation. Tears were sampled using the Schirmer I method with filter paper (Schirmer Tear Production Measuring Strips; Showa Yakuhin Kako, Tokyo, Japan). The Schirmer strips were stored at -20°C until further use. The Schirmer strips were thawed and eluted overnight at room temperature using 0.5 M NaCl and 0.5% Tween 20 containing 0.05 M phosphate-buffered solution (pH 7.2). The amount of tears obtained was calculated

by considering 1 mm of a wet Schirmer strip to contain 1 μ l of tears. Thus, the end concentration of the eluted solution corresponded to a 20-fold dilution of the original tear sample.

Tear IL-6 level and TNF- α level were measured using a commercial sandwich-type enzyme-linked immunosorbent assay kit for IL-6 and TNF- α .

All the procedures were conducted by one surgeon under sterile conditions, and the total time for the procedure was 30 min. CXL protocols were described elsewhere in detail.⁸

After instilling topical anesthesia (tetracaine 0.5%, SinaDarou, Iran), the central 8.0 mm of the corneal epithelium was removed by mechanical debridement using a blunt blade under sterile conditions. Then, riboflavin solution (0.1% in 20% dextran T500 solution; SinaDarou, Iran) was administered to the cornea topically every 3 min for 30 min. Central cornea (of 8.0 mm diameter) was irradiated using ultraviolet (UV) A 365 nm light (UV-X system, IROC AG, Zurich, Switzerland) with an irradiance of 3 mW/cm². The device was set at a working distance of 5 cm from the corneal surface. After the treatment, a soft bandage contact lens (ACUVUE OASYS[®], Vistakon Pharmaceuticals, and LLC) was placed on the cornea. All the patients received topical antibiotics, ciprofloxacin eye drops (Ciplex[®] 0.30%, SinaDarou, Iran) and topical corticosteroid, betamethasone eye drops (Betasonate[®] 0.1%) every 6 h. After the surgery, all the patients were examined with a slit-lamp on the 3rd day, and 1 week after surgery to evaluate possible complication, epithelial healing, and absence of postoperative infection.¹⁸ The contact lens was removed after epithelial healing, typically 3–5 days postoperatively. Ciplex[®] and Betasonate[®] were continued for another 1 and 3 weeks after removal of bandage lens, respectively.

Data analyses were conducted by the Statistical Package for the Social Sciences (SPSS) version 20 software (IBM Inc., Chicago, IL, USA). $P < 0.05$ was considered statistically significant. Continuous and categorical variables were reported as mean \pm standard deviation (SD) and frequency (percentages). Visual acuity was transformed into a logMAR for statistical analysis. Changes in BCVA logMAR and K-max were considered the main dependent variables.

A nested structure was considered for the data in which the studied eyes were considered nested units in each study participant according to the univariate analysis followed by multivariable linear mixed effects. Generalized estimating equation method was used to account for the correlation between fellow eyes in the regression analysis. Furthermore, regression analysis was used to determine the association of predictors for changes in K-max and BCVA (logMAR). Those predictors with $P < 0.2$ in univariate analyses were entered in the multivariable analysis.

RESULTS

This study included 61 patients (106 eyes) with KC who underwent CXL from September 2017 to September 2018. Of

those, two patients were lost to follow-up (one pregnant woman and one no-show patient in the follow-up period). Therefore, 59 patients (104 eyes) were included in the final analysis, all of whom completed year 1.

The median age was 27 years (range, 12–50 years), and the mean \pm SD age of the patients was 44.53 ± 9.03 years. Fifty-four and one-fifth percent of patients were male. Table 1 depicts patient demographics and preoperative characteristics.

Before the CXL, the mean \pm SD tear levels of IL-6 and TNF- α were 102.47 ± 60.82 pg/mL and 215.22 ± 102.01 pg/mL, respectively [Table 1].

Table 2 shows the changes in visual acuity, keratometric, and topometric parameters between the baseline and 1 year after CXL.

Table 3 provides a univariate correlation between all putative predictors and changes in BCVA logMAR and K-max.

Regarding changes in K-max, notable predictors in the patient's history include birthplace ($P = 0.03$), atopic constitution ($P = 0.01$), rubbing ($P = 0.07$), and blood group ($P = 0.17$). Neither sex nor age influenced the changes of K-max [Table 3]. While regarding the patient's history, only birthplace ($P = 0.01$), place of life ($P = 0.05$), and age ($P = 0.11$) correlated with changes in BCVA (logMAR) [Table 3].

Pretreatment BCVA (logMAR) correlated with changes in K-max ($P = 0.01$) and changes in BCVA (logMAR) ($P < 0.001$) [Table 3].

Corneal asphericity (Q value), astigmatism type, anterior average radii of curvature (ARC), posterior average radii of curvature (PRC), and CKI correlated with changes in K-max [Table 3]. Table 3 summarizes the tomographic parameters associated with changes in BCVA.

Table 4 summarizes the multivariate correlation between the supposed predictors and post-CXL K-max. The use of multivariate analysis changes in K-max was significantly associated with rubbing frequency ($P = 0.02$), blood group ($P = 0.005$), pretreatment corneal asphericity ($P < 0.001$), and pretreatment CKI ($P = 0.001$) [Table 4].

The significant multivariate associations were found between place of birth (city) ($P = 0.03$), place of residence (city) ($P < 0.001$), pretreatment central corneal thickness (CCT) ($P = 0.04$), pretreatment KI ($P = 0.04$), and changes in BCVA (logMAR) [Table 5].

There was no major CXL-related complication including the abscess formation and persisting haziness during study period.

DISCUSSION

The results of our study demonstrated that blood group B⁻, frequent rubbing, lower pretreatment corneal asphericity, and lower pretreatment CKI resulted in higher K-max reduction. Furthermore, rural place of birth, lower pretreatment BCVA

Table 1: Characteristics data of 59 patients enrolled in study; 104 eyes

Variable	Mean (SD) Median (minimum-maximum), n (%)
Age	27.27 (6.28) 27 (12-50)
Sex	
Male	32 (54.2)
Female	27 (45.8)
Laterality	
Unilateral	14 (22)
Bilateral	45 (78)
Place of birth	
City	49 (83.1)
Village	10 (16.9)
Place of life	
City	49 (83.1)
Village	10 (16.9)
Atopic constitution history	
Positive	5 (8.5)
Negative	54 (91.5)
Family history	
Positive	19 (32.2)
Negative	40 (67.8)
Rubbing of eyes	
Never	5 (8.5)
Some time	38 (64.4)
Most time	14 (23.7)
Always	2 (3.4)
Sleep apnea disease	
Positive criteria	7 (11.9)
Negative criteria	52 (88.1)
Blood grouping	
A+	18 (30.5)
A-	2 (3.4)
B+	7 (11.9)
B-	1 (1.7)
AB+	0
AB-	0
O+	19 (32.2)
O-	5 (8.5)
Tear IL-6 level	102.47 (60.82) 86.38 (32.52-416.28)
Tear TNF-a level	215.22 (102.01) 185.48 (48.64-758.08)

SD: Standard deviation, IL-6: Interleukin-6, TNF-a: Tumor necrosis factor alpha

logMAR, thinner pretreatment CCT, and higher pretreatment KI resulted in higher BCVA logMAR reduction. These factors can provide new insights into the pathogenesis of KC following CXL.

Although achievement of treatment success is probably associated with preoperative individual characteristics of patients, preoperative corneal topography and preoperative visual acuity are important. Few studies with variable results have been conducted to evaluate predictive factor in the improvement or progression of KC after CXL.^{10,13,14,19}

Table 2: The changes in mean different parameters between the baseline and at 1-year after collagen cross-linking

Parameters	Mean (SD)		P
	Before CXL	After CXL	
UCVA (logMAR)	0.52 (0.30)	0.46 (0.27)	0.045
BCVA (logMAR)	0.23 (0.16)	0.21 (0.18)	0.13
K-max (D)	52.29 (5.80)	51.94 (6.01)	0.003
CCT	473.22 (40.59)	466.29 (43.85)	<0.001
Deviation index	6.81 (2.98)	7.02 (2.82)	<0.001
Corneal asphericity (Q value)	-0.71 (0.46)	-0.63 (0.55)	0.001
ARC	6.98 (0.57)	7.07 (0.57)	<0.001
PRC	5.28 (0.54)	5.26 (0.57)	<0.001
ISV	66.25 (30.97)	64.23 (31.37)	0.006
IVA	0.705 (0.38)	0.67 (0.38)	0.009
KI	1.17 (0.10)	1.15 (0.11)	<0.001
CKI	1.037 (0.03)	1.035 (0.04)	0.023
IHA	27.17 (22.28)	26.37 (24.49)	0.463
IHD	0.093 (0.05)	0.086 (0.05)	<0.001

CXL: Corneal collagen cross-linking, SD: Standard deviation, UCVA: Uncorrected visual acuity, BCVA: Best corrected visual acuity, K-max: Maximum keratometry, CCT: Central corneal thickness, ARC: Anterior average radii of curvature, PRC: Posterior average radii of curvature, ISV: Index of surface variance, IVA: Index of vertical asymmetry, KI: Keratoconus index, CKI: Central keratoconus index, IHA: Index of height asymmetry, IHD: Index of height decentration

Currently, there are three clinical studies providing additional insights into factors associated with CXL outcomes in KC patients.^{3,20,21} The results of the study conducted by Wisse *et al.* confirmed the role of cone eccentricity in improvement of corneal curvature following CXL. Furthermore, they demonstrated that visual acuity following CXL could be accurately predicted based on pretreatment visual acuity. According to the study conducted by Wisse *et al.*, age, sex, and K-max are debated as independent factors for predicting CXL outcome in KC.³

There are some studies reporting some characteristics influencing CXL outcomes for KC as a clue for patient selection. The limitation of most of these studies is focus on limited topographic data and visual acuity.^{9,10} Our study included multiple demographic and topographic factors associated with two cytokines. A major point is attention to univariate analysis to predict factor of CXL outcome, since many potential predictors are interrelated. In the study conducted by Greenstein and Hersh, the only independent predictor of a change in the postoperative BCVA after CXL was the preoperative BCVA.¹⁴ Moreover, in the study conducted by Viswanathan and Males *et al.*, visual acuity outcome could be predicted based on pretreatment visual acuity.⁴

In another study, Badawi *et al.* demonstrated that worse BCVA, higher K-max, and relative thinner corneas were associated with greater improvement, while multivariate evaluation revealed a strong interrelation with preoperative BCVA only.²⁰

Table 3: Univariate linear regression of the baseline predictive factors and its significance on the treatment outcomes

Variable	Changes in K-max (D)			Changes in BCVA (logMAR)		
	Regression coefficient	95% CI	P	Regression coefficient	95% CI	P
History						
Age	0.019	-0.052-0.092	0.58	0.004	-0.001-0.009	0.11*
Male sex	-0.159	-0.964-0.645	0.69	-0.040	-0.104-0.023	0.21
Place of birth (city)	1.1992	0.115-2.282	0.03*	-0.105	-0.189--0.021	0.01*
Place of residence (city)	0.294	-0.791-1.380	0.59	-0.083	-0.166-0.0006	0.052*
Atopic constitution (no)	1.766	0.361-3.171	0.01*	0.0126	-0.101-0.127	0.82
Family history (yes)	-0.103	-1.540-1.332	0.88	-0.029	-0.133-0.075	0.57
Rubbing frequency	0.574	-0.051-1.199	0.07*	-0.009	-0.063-0.043	0.72
Sleep apnea (no)	-0.377	-1.673-0.917	0.56	0.0305	-0.072-0.133	0.55
Blood group						
A+	0.935	-0.662-2.533	0.17*	-0.020	-0.142-0.101	0.88
A-	0.967	-1.566-3.500		-0.110	-0.303-0.083	
B+	-0.594	-2.608-1.419		-0.026	-0.173-0.120	
B-	3.329	0.041-6.617		-0.086	-0.337-0.165	
O+	0.738	-0.849-2.327		-0.038	-0.162-0.085	
O-	Ref	Ref		Ref	Ref	
Biochemical						
IL-6	0.002	-0.003-0.009	0.40	0.0001	-0.0003-0.0007	0.49
TNF-a	-0.001	-0.005-0.002	0.54	0.00002	-0.0003-0.0003	0.89
Functional						
Pretreatment UCVA (logMAR)	0.615	-0.741-1.972	0.37	-0.0937	-0.1946-0.0071	0.06*
Pretreatment BCVA (logMAR)	3.079	0.711-5.448	0.01*	-0.4258	-0.600--0.251	<0.001*
Anatomical						
Pretreatment K-max (D)	-0.041	-0.112-0.029	0.24	0.008	0.003-0.014	0.002*
Pretreatment CCT	0.001	-0.007-0.011	0.72	-0.0008	-0.001-0.00002	0.057*
Pretreatment thinnest point	0.001	-0.008-0.011	0.71	-0.0009	-0.001--0.0000	0.03*
Pretreatment deviation index	0.019	-0.110-0.150	0.76	0.009	-0.001-0.019	0.09*
Pretreatment corneal asphericity (Q value)	-0.799	-1.711-0.112	0.08*	-0.063	-0.133-0.006	0.07*
Pretreatment astigmatism type						
ATR	-0.960	-1.887--0.033	0.10*	0.032	-0.043-0.109	0.61
Oblique	-0.206	-1.196-0.782		0.032	-0.046-0.112	
WTR	Ref	Ref		Ref	Ref	
Pretreatment ARC	0.595	-0.168-1.359	0.12*	-0.033	-0.093-0.027	0.27
Pretreatment PRC	0.737	-0.048-1.523	0.06*	-0.036	-0.1005-0.028	0.26
Pretreatment ISV	-0.006	-0.020-0.007	0.38	0.0006	-0.0004-0.0017	0.22
Pretreatment IVA	-0.263	-1.404-0.877	0.64	0.053	-0.035-0.142	0.23
Pretreatment KI	-1.296	-5.527-2.934	0.54	0.227	-0.106-0.561	0.17*
Pretreatment CKI	-9.084	-21.49-3.32	0.14*	0.210	-0.789-1.209	0.67
Pretreatment IHA	-0.011	-0.031-0.008	0.24	-0.0003	-0.0018-0.0012	0.67
Pretreatment IHD	-2.646	-10.41-5.11	0.50	0.324	-0.289-0.938	0.29

K-max: Maximum keratometry, BCVA: Best corrected visual acuity, CI: Confidence interval, IL-6: Interleukin-6, TNF-a: Tumor necrosis factor alpha, UCVA: Uncorrected visual acuity, CCT: Central corneal thickness, ATR: Against the rule, WTR: With the rule, ARC: Anterior average radii of curvature, PRC: Posterior average radii of curvature, ISV: Index of surface variance, IVA: Index of vertical asymmetry, KI: Keratoconus index, CKI: Central keratoconus index, IHA: Index of height asymmetry, IHD: Index of height decentration, Ref: Set as reference for analysis, *Those predictors with $P < 0.2$ in univariate analyses were entered in the multivariable analysis

Contrary to the mentioned study, Koller *et al.* demonstrated that there was no independent preoperative indicator for unwanted outcome.¹⁰

Greenstein *et al.* reported that male patients and patients with a central cone location seemed to benefit more from CXL treatment in terms of K-max regression.²²

In addition, Koller *et al.* found that a higher baseline K-max was associated with a greater degree of flattening.¹³

Contrary to some studies, we did not observe any correlation between age, gender, and the change in K-max after the treatment.

In agreement with our finding, the result of the study by Hashemi *et al.* showed a relatively high prevalence of KC in rural areas of Iran.²³ One reason for the higher prevalence of KC in rural areas could be attributed to some factors related to rural life, such as agricultural activities and more frequent exposure to the sunlight and UV light every day, which is one

Table 4: Multivariate linear regression of the baseline predictive factors and its significance on the maximum keratometry changes

Variable	Changes in K-max (D)		
	Regression coefficient	95% CI	P
Place of birth (city)	0.882	-0.359-2.124	0.16
Atopic constitution	0.706	-0.763-2.175	0.33
Rubbing frequency	0.948	0.126-1.769	0.02
Blood group			
A+	2.729	0.975-4.484	0.005
A-	1.620	-1.350-4.591	
B+	0.524	-1.501-2.549	
B-	4.522	1.615-7.429	
O+	2.027	0.385-3.669	
O-	Ref	Ref	
Pretreatment BCVA (logMAR)	2.707	-0.516-5.930	0.09
Pretreatment corneal asphericity (Q value)	-3.99	-6.038-1.950	<0.001
Pretreatment astigmatism type			
ATR	-0.453	-1.463-0.555	0.27
Oblique	-0.935	-2.096-0.226	
WTR	Ref		
Pretreatment ARC	0.756	-0.529-2.043	0.24
Pretreatment PRC	-0.455	-1.906-0.996	0.52
Pretreatment CKI	-55.38	-85.91-24.84	0.001

K-max: Maximum keratometry, CI: Confidence interval, BCVA: Best corrected visual acuity, ATR: Against the rule, WTR: With the rule, ARC: Anterior average radii of curvature, PRC: Posterior average radii of curvature, CKI: Central keratoconus index, logMAR: Logarithm of the minimum angle of resolution

Table 5: Multivariate linear regression of the baseline predictive factors and its significance on the best corrected visual acuity (logMAR) changes

Variable	Changes in BCVA (logMAR)		
	Regression coefficient	95% CI	P
Age	0.002	-0.0026-0.007	0.34
Place of birth (city)	-0.081	-0.158-0.005	0.03
Place of residence (city)	-0.002	-0.075-0.069	0.94
Pretreatment UCVA (logMAR)	0.045	-0.066-0.157	0.41
Pretreatment BCVA (logMAR)	-0.677	-0.930-0.424	<0.001
Pretreatment K-max (D)	-0.009	-0.021-0.003	0.14
Pretreatment CCT	-0.005	-0.008-7.60	0.04
Pretreatment thinnest point	0.003	-0.0006-0.0082	0.09
Pretreatment deviation index	0.016	-0.015-0.049	0.29
Pretreatment corneal asphericity (Q value)	-0.054	-0.163-0.055	0.32
Pretreatment KI	0.536	-0.029-1.02	0.04

BCVA: Best corrected visual acuity, CI: Confidence interval, UCVA: Uncorrected visual acuity, K-max: Maximum keratometry, CCT: Central corneal thickness, KI: Keratoconus index, logMAR: Logarithm of the minimum angle of resolution

of the reported risk factors of KC.² Another important reason could be attributed to ethnic differences.²⁴

Regarding the influence of blood groups and KC, since blood groups are genetic entities, blood group antigens have been shown to be expressed by corneal epithelium, but not the stroma or endothelium in a similar pattern with the individual's red blood cell (RBC) phenotype.²⁵ These findings suggest a possible link between KC and RBC.²⁶

Recent studies have shown a significant role of proteolytic enzymes, cytokines, and free radicals. The majority of studies in the tears of patients with KC have found increased levels of IL-6, TNF- α , and MMP.^{4,27} Whether these factors play a role in the effectiveness and consequences of CXL treatment has not been established yet. According to our study, there was no correlation between levels of IL-6 and TNF- α with changes in K-max and BCVA.

Our study had several limitations. First, although it provides useful information about proper patient selection for the CXL, the data of predictors are often not generalizable to patients outside the study population; therefore, this study should be validated in future studies to implement in clinical practice. Second, only two outcomes (changes in K-max and changes in BCVA) were included in our study.

Although our study had some limitations, the primary strength of our study is the evaluation of multiple topometric and keratometric parameters.

In conclusion, we found that changes in anatomical outcome (K-max) after CXL were significantly associated with blood group B⁻, frequent rubbing, lower corneal asphericity, and lower CKI. Furthermore, changes in functional outcome (BCVA) correlate with rural place of birth, lower pretreatment BCVA logMAR, thinner CCT, and higher KI. These factors can provide new insights into the pathogenesis of KC following CXL and provide new insights into prediction of CXL and better patient selection. However, further future and validation studies are needed.

Financial support and sponsorship

Nil.

Conflicts of interest

There are no conflicts of interest.

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