Treating keratoconus disease with the cross-linking method

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Abstract

Aim: Keratoconus is a degenerative disease, starting generally at the age of 14-25 years and causing progressive thinning of the cornea. Nowadays, cross-linking is the only procedure used to stop the natural progression of keratoconus. Studied and applied for the first time at Dresden University, a great number of clinical studies have supported its efficacy in halting the progression of keratoconus. The aim of this study was to evaluate the treatment of keratoconus disease with the cross-linking method in Albanian patients.
Methods: This study was conducted at the American Hospital in Tirana and included 81 eyes (75 patients) with progressive keratoconus. Mean age was 23.5 years (range: 15-38 years). A rotating Scheimpflug camera (Pentacam HR, Oculus) was used to diagnose and follow-up the keratoconus before and post cross-linking treatment. The parameters measured included corneal elevation, pachymetry and keratometry.
Results: Central and thinnest values of pachymetry decreased. Central pachymetry values continued to decrease until three years after cross-linking. Thinnest pachymetry values followed the same trend. Flattest, steepest and maximal keratometry significantly reduced six months after cross-linking and continued to reduce even after three years. There was a tendency of stabilizing UCVA after cross-linking. Also, there was a tendency of continuous increasing of BCVA, especially six months after the procedure.
Conclusion: Cross-linking procedure seems effective in reducing corneal radius (flattest, steepest and maximal). Having a flatter cornea in a progressive keratoconus means that the progress of keratoconus is stopped and there is also a remodeling of its surface. Remodeling the cornea also stabilizes visual acuity and even improves best spectacles visual acuity.

Keywords: cornea, cross-linking, keratoconus, keratometry, pachymetry.

Introduction

The cornea is a transparent interface covering the front of the eye. It has the function of protecting the eyeball and also is a powerful refracting surface, providing 2/3 of the eye's focusing power. The adult cornea has a thickness of 500 μm and is comprised of 5 layers: epithelium, Bowman's membrane, stroma, Descemet's membrane and the endothelium. The stroma is the thickest layer composed of collagen fibrils oriented parallel to each-other. It has also transversal fibrils which bond the parallel ones to each-other, giving to the cornea its natural strength. This phenomenon is known as natural cross-linking and it is responsible for the cornea's resistance against deformation. Keratoconus is a bilateral non-inflammatory disease which causes progressive corneal thinning, leading to protrusion, distortion, and scarring of the cornea (1). It is a naturally occurring ocular condition which leads to steepening of the central cornea, increasing myopia, irregular astigmatism, and loss of best spectacle-corrected visual acuity. Corneal thinning normally occurs in the infero-temporal or in the central cornea (2). Exceptional case of superior localizations have also been described (3,4). Keratoconus becomes evident normally during puberty, although the disease has also been found to develop earlier (5) and latter in life (6). It potentially progresses until the fourth decade of life, when it usually stabilizes (6). A study has determined that 50% of non-affected eyes of subjects with unilateral keratoconus will develop the disease in 16 years (7). If left untreated, keratoconus frequently progresses to formation of Descemet's tears (known as Vogt's striae) and corneal perforation, seriously threatening the vision. At this point, corneal transplantation is required to restore useful vision and saving the eye. Corneal Collagen Cross-linking is a treatment for keratoconus and other corneal ectasia which was developed first at the University of Dresden in 1998 (8). In this procedure ultraviolet (UV) light and riboflavin (vitamin B2) drops are used to strengthen the cornea's structure, to slow or halt the progression of keratoconus, preventing deterioration of vision and the need for corneal transplantation. Firstly experimented in porcine and rabbits corneas, the results showed that riboflavin soaked and UVA irradiated corneas were stiffer and more resistant to enzymatic digestion. Investigations also proved that the treated corneas contained high molecular weight polymers of collagen due to fibril cross-linking. Others, *in vitro* investigations, on human and porcine corneas examined the best treatment parameters for standard cross-liking, such as riboflavin concentration, intensity, wavelength of UVA light, and duration of treatment (9). Also, it has been proved that UVA irradiation is not harmful for the endothelium, if the corneal thickness is above 400 μm (10). After the laboratory tests, clinical results were also encouraging. The pilot study included 16 patients with progressive keratoconus that were treated with cross-linking. All of them stopped the progression after treatment. About 70% had flattening of the steepest keratometry, decrease in average and maximum keratometric values and 65% had visual acuity improvement. No complications were reported (8).

After that, cross-linking became a worldwide used technique. Generally is applied by using Dresden protocol (8) requiring the removal of central 9 nm of corneal epithelium layer, followed by 30 minutes of riboflavin administration, subsequently, UVA light is applied for 30 minutes. The corneal epithelial layer is generally removed to increase penetration of the riboflavin into the stroma (11). During the UV light illumination, riboflavin acts further as a shield during irradiation to the cornea, protecting deeper ocular structures such as the endothelium, lens, and retina from UVA irradiances that are too high (12). Another important role of riboflavin is to prevent corneal dehydration during exposure (13). The combination of riboflavin and UVA light creates 80-95% absorption into the cornea during

cross-linking depending on the concentration and the corneal thickness (12). Given the simplicity and minimal costs of the treatment, cross-linking treatment is also well-suited for developing countries (8). As the Siena Eye Study (14), later studies (15,16), investigated long-term effects of standard cross-linking.

The aim of this study was to evaluate the treatment of keratoconus disease with the cross-linking method in Albanian patients.

Methods

In our institution, namely the American Hospital in Tirana, cross-linking technique is applied from 2009. The patients presented with complains such as: progressive changes in refraction, changing frequently the glasses and not feeling comfortable with them, high astigmatism and myopia, are suspected for keratoconus. These patients were advised to undergo topographic examination with Pentacam instrument which is based on the Scheimpflug working principle, taking 12-50 images of the cornea at different angles using a rotating camera. Anterior and posterior corneal elevations are then measured using topographic analysis, providing useful information in keratoconus diagnostic and grading the severity of keratoconus (15). IV-th grade of keratoconus with pachymetry lower than 360μm, Vogt's striae or corneal hydrops are immediately advised to undergo corneal transplant procedure. The patients, diagnosed in stage 1-3 of keratoconus, with no corneal changes are followed for six months to check the evidence of keratoconus progression and in this case, are advised to undergo cross-linking procedure. Others, already presenting clear evidence of progression in comparison of earlier topographic examination are immediately advised to the cross-linking procedure.

Overall, there were 81 eyes (75 patients) with progressive keratoconus that were included in this study. Average age was 23.5 years (the youngest patient 15 years old and the oldest 38 years old).

A rotating Scheimpflug camera (Pentacam HR, Oculus) was used to diagnose and follow-up the keratoconus before and post cross-linking treatment. Corneal elevation, pachymetry and keratometry were the parameters measured.

The inclusion criteria were as follows: progression of keratoconus resented as an increasing in maximum keratometry (steepest keratometry) at least 0,5 D in six months, preoperative corneal thickness above 400 μm, no corneal scar, no previous corneal surgeries.

Patients underwent cross-linking procedure according to Dresden protocol. After the cross-linking procedure, the patients were followed with three dimensional corneal topography (Pentacam HR Oculus). The parameters followed included: keratometry steepest, flattest, average, corneal pachimetry average and thinnest, uncorrected and best-corrected visual acuity. The follow-up time was 12 months.

Statistical Analysis of the data was performed using SPSS (Statistical Package for Social Sciences, version 20.0). For all numerical variables, measures of central tendency and dispersion were calculated. For variables following the normal distribution, arithmetic means and the respective standard deviations were calculated. Differences between groups were calculated with student's t-test. Correlation between variables was analyzed through coefficients of Kendal's tau. P-values ≤ 0.05 were considered as statistically significant.

Results

The parameters before the cross-linking treatment are presented in Table 1.

Table 1. Parameters before the cross-linking treatment

Variables	Average±SD	Minimum	Maximum
Pak_central_preop	467.09±33.70	341	554
Pak_thinnest_preop	444.83±33.52	313	526
Kerat_flatest_preop	46.68±4.41	39.2	62.4
Kerat_steepest_preop	50.58±4.93	42.3	67.9
Kmax_preop	56.46±6.30	45.4	78.2
UCVA_preop	0.20±0.18	0.01	1
BCVA_preop	0.41±0.21	0.01	1

The comparison of medium values of PAK after cross-linking is exhibited in Table 2.

Table 2. Comparison of medium values of PAK (central corneal thickness) after cross-linking

Comparison groups		Average±SD	P-value*
Comparison couple I	Pak_central_preop	466.99±34.14	<0.001
	Pak_central_1 week	457.54±32.26	
Comparison couple II	Pak_central_1 week	457.54±32.26	0.004
	Pak_central_1month	452.47±34.00	
Comparison couple III	Pak_central_1month	452.26±33.93	0.169
	Pak_central_3month	450.39±35.50	
Comparison couple IV	Pak_central_3month	450.48±35.27	0.970
	Pak_central_6month	450.53±34.12	
Comparison couple V	Pak_central_6month	450.53±34.12	0.541
	Pak_central_12month	451.25±34.19	
Comparison couple VI	Pak_central_12month	451.25±34.19	<0.001
	Pak_central_24month	445.05±36.57	
Comparison couple VII	Pak_central_24month	445.05±36.57	<0.001
	Pak_central_36month	440.61±35.65	

*Student's t-test for two couples

Analyzing the values through student's t-test for two couples, there is a statistically important difference between average values of PAK central before treatment and after the first week ($p<0.001$); the first week and the first month after treatment ($p=0.004$); the 12^{th} month compared to the 24^{th} month ($p<0.001$) and the 24^{th} month compared to the 36^{th} month after cross-linking ($p<0.001$), when a significant reduction in average values of central pachymetry is evident (PAK central) (Table 2).

Based on student's t-test for two couples, there is a statistically important difference between the average values of PAK thinnest before cross-linking and one week after cross-linking ($p<0.001$), first week and first month after procedure ($p=0.015$), first month and third month ($p=0.002$), 12^{th} and 24^{th} month ($p=0.030$), 24^{th} and 36^{th} month after cross-linking ($p=0.012$), resulting in a significant reduction of medium values of PAK thinnest (Table 3).

Table 3. Comparison of medium values of thinnest pachymetry (PAK thinnest) after cross-linking

Comparison parameters		Average±SD	P-value
Comparison couple I	Pak_thinnest_preop	444.99±34.00	<0.001
	Pak_thinnest_1week	436.06±34.86	
Comparison couple II	Pak_thinnest_1week	436.06±34.86	0.015
	Pak_thinnest_1month	431.52±33.74	
Comparison couple III	Pak_thinnest_1month	431.11±33.70	0.002
	Pak_thinnest_3month	427.39±34.75	
Comparison couple IV	Pak_thinnest_3month	427.63±34.58	0.449
	Pak_thinnest_6month	428.72±35.51	
Comparison couple V	Pak_thinnest_6month	428.72±35.51	0.432
	Pak_thinnest_12month	430.25±35.28	
Comparison couple VI	Pak_thinnest_12month	430.25±35.28	0.030
	Pak_thinnest_24month	426.43±37.73	
Comparison couple VII	Pak_thinnest_24month	426.43±37.73	0.012
	Pak_thinnest_36month	422.40±35.82	

Based on the student's t-test for two couples, there is a statistically important difference between the medium values of steepest keratometry before cross-linking and one week after cross-linking (p=0.005); first month and 3rd month after procedure (p<0.001); 3rd month and 6th month (p<0.001); 6th month and 12th month (p<0.001); 12th month and 24th (p<0.001); and 24th and 36th month (p<0.001), where a statistically important reduction is seen in medium values of steepest keratometry. There is no evidence of a statistically important reduction between first week and first month after treatment (p=0.596) (Table 4).

Table 4. Comparison of Steepest Keratometry after cross-linking

Comparison of keratometry		Average±SD	P-value
Comparison couple I	kerat_steepest_preop	50.70±5.04	0.005
	kerat_steepest_1week	51.19±5.12	
Comparison couple II	kerat_steepest_1week	51.19±5.12	0.596
	kerat_steepest_1month	51.07±5.15	
Comparison couple III	kerat_steepest_1month	51.03±5.05	<0.001
	kerat_steepest_3month	50.22±4.80	
Comparison couple IV	kerat_steepest_3month	50.18±4.78	<0.001
	kerat_steepest_6month	49.57±4.93	
Comparison couple V	kerat_steepest_6month	49.57±4.93	<0.001
	kerat_steepest_12month	48.71±4.51	
Comparison couple VI	kerat_steepest_12month	48.71±4.51	<0.001
	kerat_steepest_24month	47.75±4.43	
Comparison couple VII	kerat_steepest_24month	47.75±4.43	<0.001
	kerat_steepest_36month	46.34±4.39	

Based on the student's t-test for two couples, there is a statistically important difference between the average values of maximal keratometry (Kerat_kmax) before cross-linking and one week after cross-linking (p=0.008); first and third month (p<0.001); third month and 6th month (p<0.001); 6th month and 12th

month (p<0.001); 12th month and 24th month (p=0.002); and 24th and 36th month (p<0.001), where a statistically important reduction is seen in average values of maximal keratometry (Kerat_Kmax). There is no evidence of a statistically important reduction between first week and first month after treatment (p=0.917) (Table 5).

Table 5. Comparison of maximal keratometry after cross-linking

Parameters		Average±SD	P-value
Couple I	Kmax_preop	56.57±6.42	000.1
	Kerat_Kmax_1 week	57.09±6.23	
Couple II	Kerat_Kmax_1 week	57.09±6.23	0.917
	Kerat_Kmax_1 month	57.11±6.19	
Couple III	Kerat_Kmax_1 month	57.07±6.07	<0.001
	Kerat_Kmax_3 month	56.00±5.90	
Couple IV	Kerat_Kmax_3 month	55.94±5.89	<0.001
	Kerat_Kmax_6 month	55.08±5.90	
Couple V	Kerat_Kmax_6 month	55.08±5.90	<0.001
	Kerat_Kmax_12 month	53.14±5.15	
Couple VI	Kerat_Kmax_12 month	53.14±5.15	0.002
	Kerat_Kmax_24 month	52.24±5.00	
Couple VII	Kerat_Kmax_24 month	52.24±5.00	<0.001
	Kerat_Kmax_36 month	50.53±4.80	

Based on the student's t-test for two couples, there is a statistically important difference between the average values of UCVA (uncorrected visual acuity) in first month and third month (p<0.001) and first month (p=0.052); 3rd month and sixth month (p=0.214); sixth month and 12th month (p=0.135); and 12th month and 24th month (p=0.157) (Table 6).

and 24th month and 36th month (p=0.002), where a statistically important increasing is seen in average values of UCVA. There is no evidence of statistically important changes between first week

Table 6. Comparison of values UCVA (uncorrected visual acuity)

Comparing parameters		Average±SD	P-value
Couple I	UCVA_preop	0.19±0.18	0.754
	UCVA_1week	0.22±0.60	
Couple II	UCVA_1week	0.22±0.60	0.052
	UCVA_1month	0.23±0.59	
CoupleIII	UCVA_1month	0.17±0.14	0.001
	UCVA_3month	0.21±0.17	
Couple IV	UCVA_3month	0.22±0.17	0.214
	UCVA_6month	0.23±0.18	
Couple V	UCVA_6month	0.23±0.18	0.135
	UCVA_12month	0.25±0.18	
Couple VI	UCVA_12month	0.25±0.18	0.157
	UCVA_24month	0.26±0.19	
Couple VII	UCVA_24month	0.26±0.19	0.002
	UCVA_36month	0.29±0.18	

Table 7. Comparison of BCVA (best corrected visual acuity) after cross-linking

Comparing parameters		Average±SD	P-value
Couple I	BCVA_preop	0.41±0.20	<0.001
	BCVA_1week	0.27±0.19	
Couple II	BCVA_1week	0.27±0.20	<0.001
	BCVA_1month	0.33±0.20	
Couple III	BCVA_1month	0.33±0.20	<0.001
	BCVA_3month	0.43±0.20	
Couple IV	BCVA_3month	0.43±0.20	<0.001
	BCVA_6month	0.51±0.19	
Couple V	BCVA_6month	0.51±0.19	<0.001
	BCVA_12month	0.57±0.18	
Couple VI	BCVA_12month	0.57±0.17	<0.001
	BCVA_24month	0.60±0.17	
Couple VII	BCVA_24month	0.60±0.17	<0.001
	BCVA_36month	0.67± 0.15	

Based on the student's t-test for two couples, there is a statistically important difference between the average values of BCVA after cross-linking for all the comparison period (Table 7).

There is a tendency of continuous increasing of BCVA especially starting six months after procedure and continuing even after three years with 2/10 (Snellen chart).

Discussion

The main parameters which define the topographic corneal shape are the radius of corneal curvature. Generally, two of them, perpendicular to each-other, are used to topographically characterize a certain cornea (the flattest and the steepest keratometry). Another keratometry value, corresponding to the apex of the cone or the point of maximal corneal elevation is recorded in Pentacam examination referring as maximal keratometry (Kmax).

As the Siena Eye Study (14), later studies (15,16), investigated long-term effects of standard cross-linking. Three hundred and sixty-three eyes were treated and monitored over four years, producing reliable long-term results proving the efficacy of the procedure in terms of long-term stability of the cornea by halting the progression of keratoconus, and proving the safety of the procedure (14,17-19). The use of cross-linking was also employed in other forms of corneal ectasia, even in iatrogenic after refractive surgery (20).

In our study, the flattest, steepest and maximal radius of the cornea was taken from the anterior curvature sagittal map of the cornea. The corneal thickness values, central and thinnest, were taken also from this map. With the advancement of keratoconus: steepest, flattest and Kmax increase. Central and thinnest values of pachymetry decrease. Central pachymetry values continue to decrease until three years after cross-linking. This phenomenon is known as "Corneal shrinking". Cornea stiffens and becomes stronger, opposing to the deforming tendency of the keratoconus. Thinnest pachymetry values follow the same tendency as central pachymetry. They continue to decrease until three years after cross-linking. Cornea stiffens and becoms stronger, opposing to the deforming tendency of the keratoconus. Flattest keratometry significantly reduces six months after cross-linking and continues to reduce even after three years (flattening 3.8 D). Steepest keratometry significantly reduces six months after cross-linking and continues to reduce even after three years (flattening 3.36 D). Maximal

keratometry significantly reduces six months after cross-linking and continues to reduce even after 3 years (flattening 6 D). There is a tendency of stabilizing UCVA after cross-linking and even an increasing 1/10, three years after the procedure. Also, there is a tendency of continuous increasing of BCVA especially starting six months after the procedure and continuing even after three years with 2/10.

Conflicts of interest: None declared.

Conclusion

Cross-linking procedure seems effective in reducing corneal radius (flattest, steepest, maximal). Having a flatter cornea in a progressive keratoconus means that the progress of keratoconus is stopped and there is also a remodeling of its surface.
Remodeling the cornea also stabilizes visual acuity and even improves best spectacles visual acuity.

References

1. Krachmer JH, Feder RS, Belin MW. Keratoconus and related noninflammatory corneal thinning disorders. Surv Ophthalmol 1984;28:293-322.
2. Auffarth GU, Wang L, Völcker HE. Keratoconus evaluation using the Orbscan topography system. J Cataract Refract Surg 2000;26:222-8.
3. Prisant O, Legeais, Renard G. Superior keratoconus. Cornea 1997;16:693-4.
4. Weed KH, McGhee CN, MacEwen CJ. Atypical unilateral superior keratoconus in young males. Cont Lens Anterior Eye 2005;28:177-9.
5. Rahmen W, Anwar S. An unusual case of keratoconus. J Pediatr Ophthalmol Strabismus 2006;43:373-5.
6. Rabinowitz YS. Keratoconus. Surv Ophthalmol 1998;42:297-319.
7. Rabinowitz YS, Yang H, Rasheed K, Li X. Longitudinal analysis of the fellow eyes in unilateral keratoconus. Invest Ophthalmol Vis Sci 2003;44:1311.
8. Wollensak G, Spoerl E, Seiler T. Riboflavin/ ultraviolet-a-induced collagen crosslinking for the treatment of keratoconus. Am J Ophthalmol 2003;135:620-7.
9. Koller T, Mrochen M, Seiler T. Complication and failure rates after corneal crosslinking. J Cataract Refract Surg 2009;35:1358-62.
10. Ashwin PT, McDonnell PJ. Collagen cross-linkage: a comprehensive review and directions for future research. Br J Ophthalmol 2010;94:965-70.
11. Spoerl E, Huhle M, Seiler T. Induction of cross-links in corneal tissue. Exp Eye Res 1998;66:97-103.
12. Spoerl E, Mrochen M, Sliney D, Trokel S, Seiler T. Safety of UVA-riboflavin cross-linking of the cornea. Cornea 2007; 26:385-9.
13. Aurich H, Wirbelauer C, Jaroszewski J, Hartmann C, Pham DT. Continuous measurement of corneal dehydration with online optical coherence pachymetry. Cornea 2006;25:182-4.
14. Caporossi A, Mazzotta C, Baiocchi S, Caporossi T. Long-term results of riboflavin ultraviolet a corneal collagen cross-linking for keratoconus in Italy: the Siena eye cross study. Am J Ophthalmol 2010;149:585-93.
15. Ishii R, Kamiya K, Igarashi A, Shimizu K, Utsumi Y, Kumanomido T. Correlation of corneal elevation with severity of keratoconus by means of anterior and posterior topographic analysis. Cornea 2012;31:253-8.
16. Wittig-Silva C, Whiting M, Lamoureux E, Lindsay RG, Sullivan LJ, Snibson GR. A randomized controlled trial of corneal collagen cross-linking in progressive keratoconus: preliminary results. J Refract Surg 2008;24:S720-5.
17. Kohlhaas M, Spoerl E, Schilde T, Unger G, Wittig C, Pillunat LE. Biomechanical evidence of the distribution of cross-links in corneas treated with riboflavin and ultraviolet A light. J Cataract Refract Surg 2006;32:279-83.
18. Vinciguerra P, Albe E, Trazza S, Rosetta P, Vinciguerra R, Seiler T, et al. Refractive, topographic, tomographic, and aberrometric analysis of keratoconic eyes undergoing corneal cross-linking. Ophthalmology 2009;116:369-78.
19. Caporossi A, Baiocchi S, Mazzotta C, Traversi C, Caporossi T. Parasurgical therapy for keratoconus by riboflavin-ultraviolet type A rays induced cross-linking of corneal collagen; preliminary refractive results in an Italian study. J Cataract Refract Surg 2006;32:837-45.
20. Vinciguerra P, Camesasca FI, Albe E, Trazza S. Corneal collagen cross-linking for ectasia after excimer laser refractive surgery: 1-year results. J Refract Surg 2010;26:486-97.