Journal of Biomechanics 93 (2019) 209–212

Contents lists available at ScienceDirect

Journal of Biomechanics

journal homepage: www.elsevier.com/locate/jbiomech
www.JBiomech.com

Short communication

Two-year changes in corneal stiffness parameters after accelerated corneal cross-linking

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ARTICLE INFO

Article history:
Accepted 14 June 2019

Keywords:
Accelerated corneal cross-linking
Corneal stiffness
Dynamic corneal response
9 mW/cm^2 protocol
18 mW/cm^2 protocol

ABSTRACT

The objective of this non-randomized trial was comparison of two-year changes in dynamic corneal response (DCR) between 18 mW/cm^2 (5- min) and 9 mW/cm^2 (10-min) cross-linking (CXL) protocols, using novel stiffness parameters and correlating them to clinical indices. The two groups were evaluated before and 2 years after the procedure using Corvis ST (Oculus Optikgeräte GmbH, Germany) and DCR parameters such as deformation amplitude ratio at 1 mm and 2 mm (DA ratio-1 mm and DA ratio-2 mm) and integrated radius and stiffness parameters at A1 (SP-A1). Two-year follow-up was completed for 16 of the 30 eyes in the 5-min group and 21 of the 25 eyes in the 10-min group; data from those who were lost to follow-up was not included in the analyses. Mean age at baseline was 21.7 ± 4.9 and 21.5 ± 5.2 years in the 5- and 10-min groups, respectively (P = 0.895). At 2 years after CXL, in the 5-min group, the reduction in integrated radius (−1.12 ± 1.27 mm, P = 0.003) was significant, and the increase in SP-A1 (7.11 ± 14.86 mmHg/mm, P = 0.075) was borderline, while in the 10-min group, the decrease in DA ratio-2 mm (−0.43 ± 0.58, P = 0.003) and integrated radius (−1.89 ± 1.72 mm, P < 0.001), and increase in SP-A1 (7.67 ± 10.92 mmHg/mm, P = 0.004) were significant. In both groups, the strongest and significant correlation was observed between DCR parameters and changes in radius of curvature. In conclusion, results indicated corneal strengthening with both protocols especially with the 9 mW/cm^2. Corvis ST indices can provide "in vivo" biomechanical evidence on the efficacy of CXL that may occur prior to clinical indices.

1. Introduction

Corneal crosslinking (CXL) is used to halt the progression of corneal ectasia (Wollensak, 2006). Studies have evaluated the outcomes of various accelerated CXL protocols. An outcome of interest is corneal biomechanics [corneal hysteresis (corneal viscoelastic response) and resistance factor (total resistance against deformation)] which have been studied using the Ocular Response Analyzer (ORA) after CXL (Hashemi et al., 2015) and it has been suggested that the area under the peak in the second applanation could detect biomechanical changes after CXL (Spoerl et al., 2011; Terai et al., 2012).

Corneal biomechanics can be also assessed using the Corneal Visualization Scheimpflug Technology (Corvis-ST; Oculus Optikgeräte GmbH, Germany) which measures the corneal biomechanical response caused by a puff of air using deformation indices.

According to the published 1-year adjusted results of the present study, given the increase in the first applanation time, reduced second applanation time, reduced highest concavity time, and reduced deformation amplitude in the 5-min group, better stiffening was suggested with this protocol compared to the 10-min approach (Hashemi et al., 2017). Since these indices are

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https://doi.org/10.1016/j.jbiomech.2019.06.011
0021-9290/

IOP-dependent, which is a confounding factor in studying corneal biomechanics, other novel indices were needed.

Novel indices have been shown to have acceptable sensitivity and specificity in discriminating clinical conditions from the normal status (Ambrósio et al. (2011); Vinciguerra et al., 2016). Among these indices, four parameters including the deformation amplitude ratio at 1 mm (DA ratio-1 mm) and 2 mm (DA ratio-2 mm), integrated radius, and stiffness parameter at A1 (SP-A1) are more suitable for detecting biomechanical changes after CXL than other parameters (Vinciguerra et al., 2016). Definitions and calculation methods for these indices are described in the Supplement Fig. 1.

In this report, 2-year changes in corneal biomechanical parameters were evaluated with these indices to compare 5-min and 10-min CXL protocols. In addition, the relationship of these parameters with vision and topography indices was also examined.

2. Methods and material

The methodology of the study has been described previously (Hashemi et al., 2017). In brief, in this non-randomized trial, patients with progressive keratoconus were enrolled in two groups to undergo accelerated CXL either with the 5-min (18 mW/cm^2 for 5 min) or the 10-min (9 mW/cm^2 for 10 min) approaches. Inclusion criteria were the diagnosis of progressive keratoconus [$\geq$1.0 diopter (D) increase in maximum keratometry (Kmax) or manifest cylinder or manifest refraction spherical equivalent or $\geq$2 lines reduction in corrected distance visual acuity (CDVA) in the past 12 months], age between 15 and 35 years, Kmax < 55.0 D, and a minimum corneal thickness of 400.0 μm. Exclusion criteria were a history of any ocular surgery or disease.

The study protocol was approved by the Institutional Review Board of Tehran University of Medical Sciences. The study was conducted in adherence to the tenets of the Helsinki Declaration. Written informed consents were obtained from all participants.

2.1. Surgical technique

As mentioned previously (Hashemi et al., 2017), the procedures were done by two surgeons (H.H, R.Gh) with similar experiences and skill in CXL at two sites in Tehran, Iran; the 5-min group received treatment at Noor Eye Hospital and the 10-min group at Farabi Eye Hospital. In both groups, after removing the central 9-mm of the corneal epithelium, the lid speculum was removed. Then, 0.1% Riboflavin in 20% dextran drops (StreuliPharma, Uznach, Switzerland) was instilled onto the cornea every 3 min for 30 min. UV irradiation was done using the CCL 365 (PESCHKE Meditrade GmbH, Waldshut-Tiengen, Germany) in the 5-min group and LightLink-CXL (LightMed, San Clemente, CA, USA) in the 10-min group. These two devices are similar in terms of UV light wavelength, spot size, beam power and homogenizer, voltage, and light emission (continuous mode). After rinsing the corneal surface and placing a soft bandage contact lens, Levofloxacin was instilled. The post-CXL regimen included eye drop Levofloxacin, corticosteroid and artificial tears.

2.2. Pre and post-CXL examinations

Corvis-ST was used to measure dynamic corneal response (DCR) indices included DA ratio-1 mm, DA ratio-2 mm, integrated radius, and SP-A1. Also, radius of curvature, Ambrosio relational thickness (ARTh), and bIOP were extracted from Corvis ST. Uncorrected and corrected visual acuity (UDVA and CDVA) were evaluated by Snellen SC-2000 (Nidek Inc., Tokyo, Japan). All before and after tests were conducted by the same optometrist.

2.3. Statistical analysis

At baseline, there were 30 eyes in the 5-min group, and 25 eyes were treated with the 10-min CXL protocol. Of these, 23 eyes in each group completed the 2-year follow-up exams and others were lost to follow-up. After exclusion of low quality score tests, data from 16 eyes in the 5-min group and 21 eyes in the 10-min group were used in the analysis. Two-year changes were evaluated within each group using paired *t*-test. The sample size and analysis power were too low to compare changes of DCR parameters between two groups. So, pre-CXL parameters were compared between groups by covariate analysis. This analysis was repeated for post-CXL parameters. Since thickness and radius of curvature influence corneal biomechanics (Roberts et al. 2017), and ARTh is a more accurate index compared to point thickness (Ambrósio et al. 2011), pre-CXL radius of curvature and ARTh were adjusted as confounders. Also, to control the IOP effect, bIOP, which is a close estimate of the true IOP (Eliasy et al., 2018) was adjusted. Kolmogorov-Smirnov test showed normal distribution of pre- and post-CXL indices.

3. Results

Data from 16 eyes in the 5-min group and 21 eyes in the 10-min group were used in the analysis. The mean age of these patients at baseline was 21.7 ± 4.9 years in the 5-min group and 21.5 ± 5.2 years in the 10-min group (p = 0.895).

Table 1 summarizes a comparison of UDVA, CDVA, radius of curvature, ARTh, and bIOP between two groups. At baseline and 2-year follow up, none of them was statistically different between groups (all P > 0.05).

Assessment of changes in DCR parameters within each group and also pre-CXL parameters and post-CXL parameters comparisons is summarized in Table 2. Adjusted comparison analysis showed significant higher pre-CXL SP-A1 in 5-min group than the other group (73.19 ± 19.31 vs. 61.91 ± 14.22, P = 0.025). But at 2 year after CXL, all DCR parameters were similar between groups (all P > 0.05).

In the 5-min group, a statistically significant decrease was observed in integrated radius (1.12 ± 1.27, P = 0.003), and the increase in SP-A1 (7.11 ± 14.86, P = 0.075) was borderline significant. In the 10-min group, the decreases in DA ratio-2 mm (0.43 ± 0.58, P = 0.003) and integrated radius (1.89 ± 1.72, P < 0.001), and the increase in SP-A1 (7.67 ± 10.92, P = 0.004) were statistically significant.

The relationship of changes in DCR parameters with changes in UDVA, CDVA, radius of curvature, and ARTh, in each group is presented in Table 3. The strongest correlation was observed between changes in DCR parameters and changes in radius. Supplement Table A compares the other variables extracted from Corvis between the two groups.

4. Discussion

Dynamic corneal response parameters are indicators of corneal stiffness, as they represent a measure of resistance to deformation. It is expected that CXL can increase corneal stiffness through the creation of new covalent bonds in the stroma, and it is aimed to at least stop the further weakening of the cornea (Wollensak, 2006).

Table 1
Comparison of baseline and 2-year vision, topographic, and IOP indices between the 5-min (16 eyes) and 10-min (21 eyes) groups.

		5-min	10-min	P-value
UDVA (logMAR)	Baseline	0.62 ± 0.61	0.45 ± 0.39	0.321
	After 2 year	0.67 ± 0.67	0.46 ± 0.47	0.341
CDVA (logMAR)	Baseline	0.11 ± 0.17	0.11 ± 0.11	0.952
	After 2 year	0.13 ± 0.15	0.09 ± 0.09	0.442
Radius of curvature (mm)	Baseline	7.62 ± 0.33	7.56 ± 0.43	0.629
	After 2 year	7.67 ± 0.62	7.53 ± 0.34	0.399
ARTh (μm)	Baseline	316.91 ± 150.13	251.83 ± 61.24	0.119
	After 2 year	255.54 ± 91.19	241.09 ± 57.12	0.558
bIOP (mmHg)	Baseline	14.87 ± 2.25	15.09 ± 1.92	0.754
	After 2 year	15.43 ± 2.16	15.47 ± 2.09	0.960

ARTh: Ambrosio relational thickness, bIOP: biomechanically correct intraocular pressure.

Table 2
Two year changes (post CXL-pre CXL) in dynamic corneal response parameters in the 5-min and 10-min groups.

	CXL protocol	Pre op	After 2 years	2-year change	P-value**
DA ratio-1 mm (unitless)	5-min	1.66 ± 0.06	1.68 ± 0.08	0.02 ± 0.10	*0.446*
	10-min	1.67 ± 0.04	1.66 ± 0.06	−0.01 ± 0.07	*0.576*
	*P-value**	*0.658*	*0.695*		
DA ratio-2 mm (unitless)	5-min	4.95 ± 0.51	4.92 ± 0.56	−0.04 ± 0.64	*0.825*
	10-min	5.33 ± 0.50	4.90 ± 0.40	−0.43 ± 0.58	*0.003*
	*P-value**	*0.285*	*0.815*		
Integrated radius (1/mm)	5-min	10.37 ± 1.81	9.25 ± 1.55	−1.12 ± 1.27	*0.003*
	10-min	11.31 ± 1.47	9.42 ± 0.80	−1.89 ± 1.72	*<0.001*
	*P-value**	*0.891*	*0.765*		
SP-A1 (mmHg/mm)	5 min	73.19 ± 19.31	80.30 ± 19.49	7.11 ± 14.86	*0.075*
	10 min	61.91 ± 14.22	69.58 ± 10.25	7.67 ± 10.92	*0.004*
	*P-value**	*0.025*	*0.501*		

DA ratio-2 mm: Deformation Amplitude Ratio at 2 mm; DA ratio-1 mm: Deformation Amplitude Ratio at 1 mm; SP-A1: stiffness parameters at A1.
* P-value of comparisons of pre-CXL and also post-CXL parameters between groups.
** P-value of 2-year change in each CXL protocols.

Table 3
Pearson correlation coefficient (P-value) between changes in dynamic corneal response indices and vision and topography changes after corneal cross linking (n = 37 eyes).

		ΔUDVA	ΔCDVA	ΔRadius of curvature	ΔARTh
5-min group	ΔDA ratio-1 mm (unitless)	0.125 (P = 0.658)	0.191 P = 0.495)	−0.462 (P = 0.072)	−0.428 (P = 0.059)
	ΔDA ratio-2 mm (unitless)	0.490 (P = 0.064)	0.486 (P = 0.067)	−0.792 (P < 0.001)	−0.415 (P = 0.110)
	ΔIntegrated radius (mm)	0.272 (P = 0.326)	0.351 (P = 0.199)	−0.783 (P < 0.001)	−0.433 (P = 0.094)
	ΔSP-A1 (mmHg/mm)	−0.312 (P = 0.258)	−0.252 (P = 0.365)	0.461 (P = 0.072)	0.417 (P = 0.108)
10-min group	ΔDA ratio-1 mm (unitless)	0.607 (P = 0.004)	0.165 (P = 0.474)	−0.731 (P < 0.001)	−0.232 (P = 0.312)
	ΔDA ratio-2 mm (unitless)	0.479 (P = 0.028)	0.100 (P = 0.665)	−0.781 (P < 0.001)	−0.232 (P = 0.312)
	ΔIntegrated radius (mm)	0.454 (P = 0.039)	0.123 (P = 0.596)	−0.733 (P < 0.001)	−0.185 (P = 0.421)
	ΔSP-A1 (mmHg/mm)	−0.178 (P = 0.441)	−0.058 (P = 0.802)	0.620 (P = 0.003)	−0.271 (P = 0.235)

DA ratio-2 mm: Deformation Amplitude Ratio at 2 mm; DA ratio-1 mm: Deformation Amplitude Ratio at 1 mm; SP-A1: stiffness parameters at A1; UDVA: uncorrected distance visual acuity; CDVA: corrected distance visual acuity; ARTh: Ambrosio relational thickness.

Corneal stiffness is influenced by corneal tomography (thickness and curvature), and consequently, the IOP and the stress-strain behavior of corneal tissue (Roberts et al. 2017). We observed increased corneal thickness, corneal curvature, and bIOP in both groups 2 years after CXL; this can be indicative of increased stiffness. The material behavior of the cornea can hardly be measured in vivo, but SP-A1 has been able to limit this bias to some extent and represents the overall corneal stiffening. In addition, we controlled for all three variables (thickness, radius of curvature, and bIOP) in the analyses to compare the strengthening effect of CXL between the two protocols. The standard deviation (SD) of 2-year change in DCRs were relatively large compared to the mean. This can be due to variations in the efficacy of CXL in the studied population or due to variation in measurements in post-CXL follow-up examinations. To our knowledge, there are no published studies addressing the repeatability of these indices after CXL. Given that a more indices showed evidence of corneal strengthening in the 10-min group, we could assume better biomechanical strengthening is achieved with this protocol. Especially the pre-CXL SP-A1 was better in 5-min group but 2 years after CXL SP-A1 became similar in both groups because of better improvement in 10-min group.

In the published 1-year report of the present study (Hashemi et al., 2017), changes in time-A1, time -A2, highest concavity time, and deformation amplitude (DA) were in favor of greater change with the 5-min protocol after adjusting for pachymetry. At two years after CXL (Table B), mean time-A2 had a significant increase with CXL in only the 5-min group, and other parameters had non-significant changes in groups. Unfortunately, the 1-year novel stiffness parameters were not available to evaluate the trend of changes.

In Supplement Table A, some indices including HC time (in the 5-min group), time-A1 and Def Amp (in the 10-min group) had a descending trend during the first year and an improving trend during the second year after CXL. There can be different explanations for this observation: (1) These indices are slow to change after CXL, and they cannot detect the 1-year effect of CXL. (2) These indices are dependent on IOP and/or corneal thickness, and they are not suitable for assessing the effects of CXL. (3) The repeatability of the measurements of these indices. In normal samples, the SD of repeated measures was 0.04 for DA (Lopes et al., 2017), 0.21 for time-A1, 0.43 for time A2, 0.09 for DA, and 0.47 for HC time (Miki et al., 2017). Given the variation in treatment response, the SDs, as observed in this study, are expected to be larger. To our knowledge, no study has assessed the SD of repeated measures after CXL. However, since the SDs are large relative to the means, the difference between first- and second-year trends can be partly attributed to measurement repeatability. Prospective repeatability studies after CXL are needed to elaborate on this issue.

Should the hypothesis of focal biomechanical weakening in the cone be true, CXL should have a better effect in the cone area (Scarcelli et al., 2014). Therefore, changes in corneal biomechanical properties should be detectable before changes in radius of curvature, corneal thickness, and vision. We demonstrated the increase of radius correlated to increase of corneal stiffness. This is in agreement with a recent report (Steinberg et al., 2016) demonstrating that the DCRs are able to show early changes in biomechanics after CXL and are measurable before corneal shape modifications take place.

As presented in Tables 1 and 3, despite corneal stiffening, there was no change in the radius of curvature or vision in either group. The effect of CXL on these parameters is controversial and improvement (Henriquez et al., 2011), decline (Razmjoo et al., 2013) and stability of indices (Hashemi et al., 2015) were reported. Therefore, CXL efficacy cannot be judged based on these indices alone. In the current study, although change of radius was similar between groups, changes in corneal biomechanical indices were different.

Limitation of this study are small sample size and the non-random assignment to groups. Another limitation is unavailability of 1-year data on the 4 novel DCR indices which lack the limitations of previous indices and would allow us to draw a closer comparison between first- and second-year results.

In conclusion, both the 5-min and 10-min CXL protocols lead to changes that are indicative of strengthened corneal biomechanics. DA ratio-2 mm, integrated radius, and SP-A1 are able to demonstrate this effect better than others, especially in the 10-min group. Changes in these indices can be detected before change in clinical and topographical indices. We need to augment evidence on the biomechanical changes of CXL (Randleman et al., 2017); such metrics can serve as suitable parameters to test novel modifications in CXL techniques. We anticipate a fast evolution of CXL protocols to demonstrate its efficacy in the clinical setting.

Acknowledgment

We thank Sven Reisdorf and Stefanie Berger who extracted the data and Dr Shiva Mehravaran who assisted us in the preparation of the manuscript.

Declaration of Competing Interest

Dr. Riccardo and Paolo Vinciguerra, Dr. Ambrosio and Dr. Roberts are consultants for Oculus Optikgeräte GmbH, Germany. None of the remaining authors have any financial disclosures.

Funding

None.

Appendix A. Supplementary material

Supplementary data to this article can be found online at https://doi.org/10.1016/j.jbiomech.2019.06.011.

References

Ambrósio Jr., R., Caiado, A.L., Guerra, F.P., Louzada, R., Sinha, R.A., Luz, A., Dupps, W. J., Belin, M.W., 2011. Novel pachymetric parameters based on corneal tomography for diagnosing keratoconus. J. Refract. Surg. 27, 753–758.

Eliasy, A., Chen, K.J., Vinciguerra, R., Maklad, O., Vinciguerra, P., Ambrósio Jr., R., Roberts, C.J., Elsheikh, A., 2018. Ex-vivo experimental validation of biomechanically-corrected intraocular pressure measurements on human eyes using the CorVis ST. Exp. Eye Res. 175, 98–102.

Hashemi, H., Asgari, S., Mehravaran, S., Miraftab, M., Ghaffari, R., Fotouhi, A., 2017. Corneal biomechanics after accelerated cross-linking: comparison between 18 and 9 mW/cm^2 protocols. J. Refract. Surg. 33, 558–562.

Hashemi, H., Miraftab, M., Seyedian, M.A., Hafezi, F., Bahrmandy, H., Heidarian, S., Amanzadeh, K., Nikbin, H., Fotouhi, A., Asgari, S., 2015. Long-term results of an accelerated corneal cross-linking protocol (18 mW/cm(2)) for the treatment of progressive keratoconus. Am. J. Ophthalmol. 160, 1164–1170.

Henriquez, M.A., Izquierdo Jr., L., Bernilla, C., Zakrzewski, P.A., Mannis, M., 2011. Riboflavin/ultraviolet A corneal collagen cross-linking for the treatment of keratoconus: visual outcomes and Scheimpflug analysis. Cornea 30, 281–286.

Lopes, B.T., Roberts, C.J., Elsheikh, A., Vinciguerra, R., Vinciguerra, P., Reisdorf, S., Berger, S., Koprowski, R., Ambrósio Jr., R., 2017. Repeatability and reproducibility of intraocular pressure and dynamic corneal response parameters assessed by the corvis ST. J. Ophthalmol. 2017, 8515742.

Miki, A., Maeda, N., Asai, T., Ikuno, Y., Nishida, K., 2017. Measurement repeatability of the dynamic Scheimpflug analyzer. Jpn. J. Ophthalmol. 61, 433–440.

Randleman, J.B., Santhiago, M.R., Kymionis, G.D., Hafezi, F., 2017. Corneal Cross-Linking (CXL): standardizing terminology and protocol nomenclature. J. Refract. Surg. 33, 727–729.

Razmjoo, H., Nasrollahi, A.P., Salam, H., Karbasi, N., Najarzadegan, M.R., 2013. Topographic corneal changes after collagen cross-linking in patients with corneal keratoconus. J. Res. Med. Sci. 18, 882–886.

Roberts, C.J., Mahmoud, A.M., Bons, J.P., Hossain, A., Elsheikh, A., Vinciguerra, R., Vinciguerra, P., Ambrósio Jr., R., 2017. Introduction of two novel stiffness parameters and interpretation of air puff-induced biomechanical deformation parameters with a dynamic Scheimpflug analyzer. J. Refract. Surg. 33, 266–273.

Scarcelli, G., Besner, S., Pineda, R., Yun, S.H., 2014. Biomechanical characterization of keratoconus corneas ex vivo with Brillouin microscopy. Invest. Ophthalmol. Vis. Sci. 55, 4490–4495.

Spoerl, E., Terai, N., Scholz, F., Raiskup, F., Pillunat, L.E., 2011. Detection of biomechanical changes after corneal cross-linking using ocular response analyzer software. J. Refract. Surg. 27, 452–457.

Terai, N., Raiskup, F., Haustein, M., Pillunat, L.E., Spoerl, E., 2012. Identification of biomechanical properties of the cornea: the ocular response analyzer. Curr. Eye Res. 37, 553–562.

Vinciguerra, R., Elsheikh, A., Roberts, C.J., Ambrósio Jr., R., Kang, D.S., Lopes, B.T., Morenghi, E., Azzolini, C., Vinciguerra, P., 2016. Influence of pachymetry and intraocular pressure on dynamic corneal response parameters in healthy patients. J. Refract. Surg. 32, 550–561.

Steinberg, J., Frings, A., Mousli, A., Casagrande, M.K., Druchkiv, V., Katz, T., Linke, S.J., 2016. New Scheimpflug dynamic in vivo curve analyses to characterize biomechanical changes of the cornea after cross-linking for progressive keratoconus. J. Refract. Surg. 32, 34–39.

Wollensak, G., 2006. Crosslinking treatment of progressive keratoconus: new hope. Curr. Opin. Ophthalmol. 17, 356–360.