TREATMENT OF KERATOCONUS WITH CORNEAL CROSS-LINKING – RESULTS AND COMPLICATIONS IN 2 YEARS FOLLOW-UP

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SUMMARY

TREATMENT OF KERATOCONUS WITH CORNEAL CROSS-LINKING – RESULTS AND COMPLICATIONS IN 2 YEARS FOLLOW-UP

Objective: The objective of the study was to assessment of changes of monitored parameters after CXL. Incidence of complications were assessed in the whole group and in groups of patients divided according to the selected criteria. Evaluated parameters were also relations between them and in time.
Methods: The 86 eyes of patients with progressive keratoconus who underwent CXL according to the Dresden protocol in the years 2007-2009 at the Ophthalmic clinic FN Brno Bohunice were included in this study.
Results: There was observed significant increase of BCVA (letters – before CXL 42,30 ± 10,35, 1st year after CXL (1Y) 44,68 ± 10,04, $p < 0,01$, 2nd year after CXL (2Y) 44,44 ± 10,57, $p < 0,01$) and SE (-5,95 ± 3,98D, -5,27 ± 3,84D, $p < 0,01$, -4,94 ± 3,68D, $p < 0,01$), and decrease of maximum curvature of the cornea (MAX – before CXL 50,39 ± 4,17D, 1Y 49,46 ± 4,13D, $p < 0,01$, 2Y 49,42 ± 4,14D, $p < 0,01$). Change of ultrasound CCT, polymegatisms, pleomorfisms and corneal endothelial cell density was not significant.
The value of MAX is the most important parameter in estimating the effect of CXL.
The highest incidence of corneal opacity after CXL was observed in the eyes of patients with III. stage of keratoconus over 40 years old, carrying hard contact lenses and with biomikroskopic symptom of keratoconus on the cornea.
We found that corneal thickness measurement with Orbscan II and the mesurement of IOP with noncontact method is incorrect by patients after CXL.
Conclusion: Corneal cross-linking of the cornea is safe and effective procedure of stopping the progression of keratoconus in 97% of eyes in the period up to 2 years after CXL.
Key words: keratoconus, corneal cross-linking, riboflavin, UVA irradiation, complications

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INTRODUCTION

Cross-linking occurs naturally in tissues in vivo via an enzymatic and non-enzymatic pathway, and in the case of the cornea is jointly responsible for increasing rigidity with advancing age. In patients with keratoconus, a progressive slowdown of the progression of ectasia is observed. The physiological process of natural cross-linking in the cornea takes place with the help of the enzyme lysyl oxidase, which transforms amino groups of amino-acids into aldehyde groups, which may then react amongst themselves, with the generation of a covalent bond (30, 25, 19).

It is precisely the observation that a halting of the progression of keratoconus takes place in patients with diabetes mellitus (28) that led Spoerl and Seiler (31) to conduct experiments at their Dresden eye clinic at the end of the last century. These trials were focused on seeking a suitable method for the induction of cross-linking in the cornea. They arrived at the finding that the most suitable method was to use riboflavin and UV-A radiation, which activates the natural lysyl oxidase pathway of cross-linking. Corneal cross-linking (CXL) realised by this method has a localised effect, a relatively short time of performance, and the cornea remains transparent after therapy (31). In tests on enucleated eyes, increased rigidity by an amount of 328% was confirmed (36). The precise mechanism of CXL is not completely clarified to date, nevertheless today several studies from around the world have demonstrated the success of the method in halting the progression of the disease, as well as in particular its safety (1, 3, 4, 6, 11, 14, 26, 33, 35, 37). Some authors now rank it in first place in the therapeutic procedure for patients aged up to 26 years with a progressive disorder (5).

The most common causes of complications of CXL include inappropriate selection of the patient (failure to detect contraindications, accompanying disease), improper technique of procedure, uncooperative patient, especially from the perspective of postoperative care and hygiene. Complications in connection with CXL are rare, and the published frequency is 1-10%, but in isolated cases they may lead to the necessity of corneal transplant (29, 18).

Complications in connection with corneal cross-linking have been described as sterile infiltrates and corneal opacities, endothelial failure, infection of the cornea, and complications of treatment may include its failure (14, 18, 19, 22).

METHOD AND COHORT OF PATIENTS

The observed cohort included 86 eyes of 68 patients with an average age of 27.70 ± 9.34 years (range 13-55 years),

who underwent corneal cross-linking with isotonic riboflavin solution and abrasion of the corneal epithelium according to the Dresden protocol (Riboflavin 0.1% + 20% Dextran T500, IROC UV-X 1000) at the department of ophthalmology at the Brno Bohunice University Hospital in the period from 2007 to November 2009.

The basic indication criteria were: diagnosis of keratoconus with demonstrated progression maximally over the period of the last year, minimum corneal thickness of more than 400 µm measured using an Orbscan II instrument, patient with sufficient best corrected vision with contact lens (CL) or glasses correction (24).

The minimum observation period was 6 months (range 6-24 months). The patients underwent a preoperative examination before the procedure. All patients (in case of minors their legal representative) were regularly informed of the risks of the procedure, its course and postoperative care, and signed an informed consent form.

The preoperative examination and all control examinations in the first month (1M), third month (3M), sixth month (6M), one year (1Y) and two years (2Y) after CXL consisted of an examination of uncorrected (UCVA) and best corrected visual acuity (BCVA) on ETDRS optotypes in both eyes, determination of the level of intraocular pressure by the noncontact method, biomicroscopic examination of the anterior segment and posterior segment of the eye also in mydriasis, automatic refractometry, determination of corneal thickness by ultrasound (instrument Nidek UP-1000), examination on an ORBSCAN II instrument with determination of keratometry values of curvature and pachymetry map of the cornea and confocal microscope (Confoscan 4 semicontact probe 40x).

A statistical analysis was performed using the program STATISTICA version 12.2 in the licence of Masaryk University and the program Microsoft Excel 2007. With regard to the unconfirmed normal distribution of the values, non-parametric methods were used for the data analysis (Wilcoxon pair test, Spearman's correlation coefficient). Statistical significance of the tests was evaluated on the level of $\alpha = 0.05$.

RESULTS

Evaluation of change of best corrected visual acuity (BCVA)

At a follow-up examination one month after CXL, a statistically significant deterioration of BCVA was determined, on average by more than 4 letters, thus by almost 1 row of ETDRS optotypes. Subsequently at 1 year and 2 years after CXL there is a significant improvement of BCVA in comparison with the preoperative state at both checks, on average by more than 2 letters. An improvement of BCVA was recorded in more than 50% of eyes (54% in the 1st year and 62% in the 2nd year), and has a rising trend over time. The number of eyes with deteriorated BCVA decreases over time (38% of eyes in the 1st year and 26% of eyes in the 2nd year).

The complete data is presented in table 1 and graph 1.

Evaluation of change in level of intraocular pressure (IOP)

At follow-up examinations up to the 6th month after CXL there was no significant change of intraocular pressure. At the examination after the first year a statistically significant reduction of intraocular pressure was recorded, and after the second year following CXL a highly significant reduction in the values of intraocular pressure measured by the noncontact method.

The complete data is presented in table 2.

Evaluation of change to corneal thickness (CCT) measured by ultrasound method and Orbscan II instrument

We measured pachymetry by two independent methods for the reason of more precise determination of the value, with regard to the strict indication limitation of the Dresden protocol. This condition was met by the eyes of all patients, only one patient had a corneal thickness of 395 µm measured on the Orbscan II instrument and 432 µm measured by the ultrasound method. Borderline statistics of a significant reduction of pachymetry values measured by the ultrasound method were measured at the 3M and 6M check. In the case of values of corneal thickness measured by the Orbscan II instrument, a statistically highly significant reduction was recorded at all checks following CXL.

The complete data is presented in tables 3 and 4, and in graphs 2 and 3.

Evaluation of change to maximum corneal curvature

Maximum corneal curvature increased significantly at the 1M check on average by 0.33D, and at all subsequent checks a significant reduction was recorded. At the 6M check the average reduction was by 0.75 D, at the 1Y check by 0.93D and the 2Y check on average by 0.96D. At the 1Y and 2Y checks a decrease of maximum corneal curvature was recorded in up to 83% of eyes. The maximum measured value in the group decreased by more than 3D at the 2Y check. The minimum measured value at the 2Y check decreased by 0.75D.

The complete data is presented in table 5 and graph 4

Evaluation of change in density, polymegathism, pleomorphism and endothelial cells

In all three observed parameters, no significant difference was recorded between the values before the procedure and at the individual checks.

Evaluation of mutual relationships of observed parameters over time before the procedure and 1 year and 2 years after CXL

We determined that the value of maximum corneal curvature is the most important parameter in estimating the effect of CXL, because it best correlates with the other observed parameters within the framework of the individual checks and also between the checks.

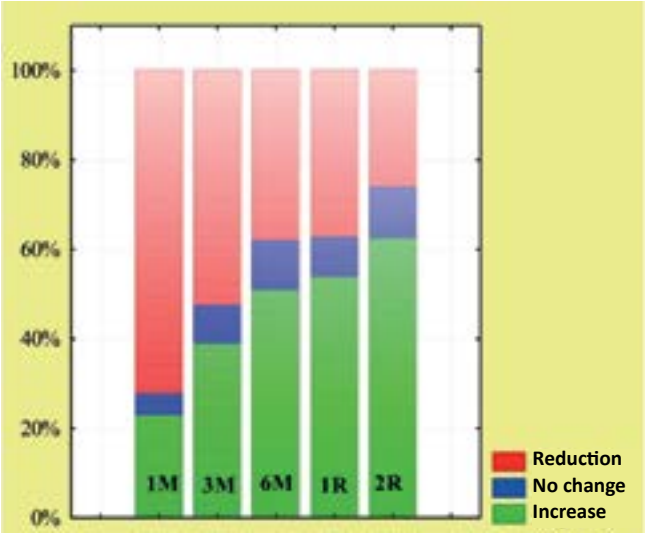
Evaluation of incidence of corneal opacity in entire cohort and groups of eyes of patients

Corneal opacity after CXL is characterised by appearance 2-3 weeks after CXL, localised in the anterior 2/3 of the corneal stroma, where cross-linking of collagen took place. It is of fine to medium density, sometimes vertical, parallel lines are visible in the stroma, reminiscent of Vogt's striae, which however recede with the disappearance of the opacity (19, 22, 23, 24).

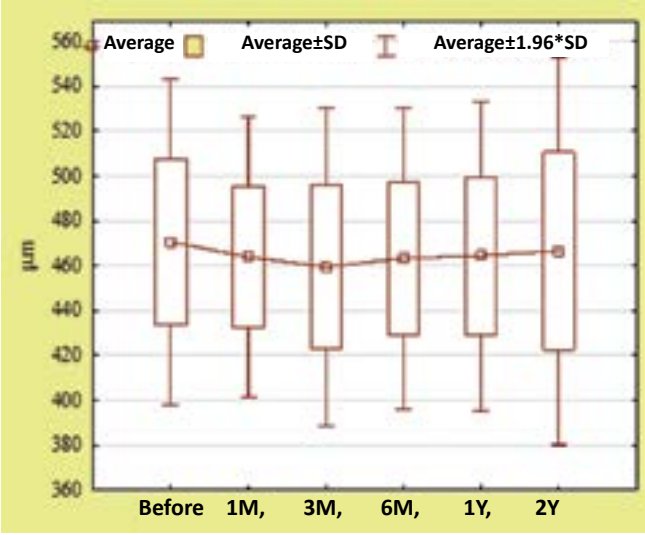
Table 1 Evaluation of change of best corrected visual acuity (BCVA) at individual checks (number of letters)

BCVA (number of letters)	Number of eyes	Average	Standard deviation	Range of values	Increase Number (%)	Stabilisation Number (%)	Reduction Number (%)	p - value
Before	83	42,30	10,35	15 – 59	-	-	-	-
1M	84	38,10	11,70	5 – 59	19 (23 %)	4 (5 %)	60 (72 %)	**p < 0,01**
3M	82	41,57	10,69	13 – 59	31 (39 %)	7 (11 %)	42 (52 %)	0,32
6M	81	43,63	10,11	16 – 61	40 (51 %)	9 (11 %)	30 (39 %)	0,09
1Y	82	44,68	10,04	15 – 60	43 (54 %)	7 (9 %)	30 (38 %)	**p < 0,01**
2Y	62	44,44	10,57	10 - 63	38 (62 %)	7 (11 %)	16 (26 %)	**p < 0,05**

(**before** - value before procedure, **1M** – 1 month after procedure, **3M** – 3 months after procedure, **6M** – 6 months after procedure, **1Y** – 1 year after procedure, **2Y** – 2 years after procedure).
Note: for some patients it was not possible to determine BCVA with glasses lenses reliably due to high astigmatism in the case of an advanced pathology, the change of the value and significance was determined from pair values.



Graph 1 Change of best corrected visual acuity



Graph 2 Corneal thickness measured by ultrasound method (µm)

In the cohort of eyes there is a visible trend of decrease in the incidence of opacity over time. The maximum incidence was in the 3rd month after CXL. Persistence of opacity after 2 years was recorded in 27% of eyes.

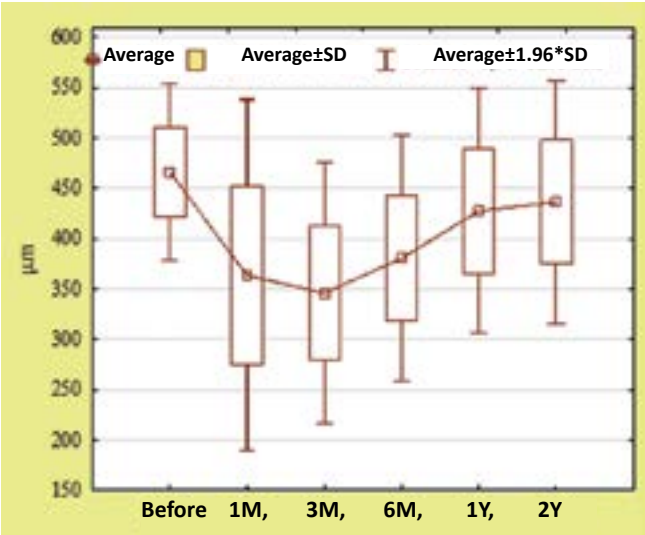
For a more detailed analysis, we divided the cohort of eyes into groups according to selected patient characteristics. The criteria for division and the abbreviations used for the groups are presented in tables 6 and 7.

Upon a comparison of the groups of eyes divided according to sex (F, M) and according to the presence of atopy in

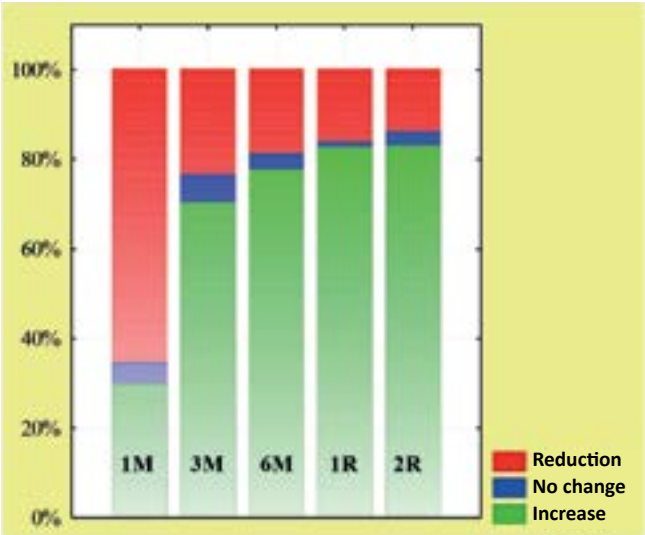
Table 2 Evaluation of change to level of intraocular pressure (IOP) by noncontact method at individual checks (torr)

IOP (torr)	Number of eyes	Average	Standard deviation	Range of values	p - value
pred	81	11,63	2,93	6 - 19	-
1M	79	11,91	2,71	7 - 18	0,16
3M	79	11,2	2,52	7 - 18	0,55
6M	78	11,09	2,55	7 - 16	0,11
1R	77	10,92	2,33	7 - 17	**p < 0,05**
2R	62	10,76	2,61	7 - 17	**p < 0,01**

(**before** - value before procedure, **1M** – 1 month after procedure, **3M** – 3 months after procedure, **6M** – 6 months after procedure, **1Y** – 1 year after procedure, **2Y** – 2 years after procedure).
Note: for some patients it was not possible to determine IOP by noncontact tonometry due to advanced keratoconus, the change of the value and significance was determined from pair values.



Graph 3 Corneal thickness measured by Orbstacn II instrument (µm)



Graph 4 Change of maximum corneal curvature

Table 3 Evaluation of change of corneal thickness (CCT) measured by ultrasound method on patients at individual checks (µm)

UZV CCT (µm)	Number of eyes	Average	Standard deviation	Range of values	p - value
pred	84	470,62	37,00	417 – 580	-
1M	78	463,81	31,72	401 - 555	0,26
3M	75	459,43	36,26	354 – 538	**p < 0,05**
6M	78	463,29	34,24	370 – 540	**p < 0,05**
1R	80	464,41	35,13	380 – 550	0,13
2R	49	466,53	44,09	402 - 600	0,23

(**before** - value before procedure, **1M** – 1 month after procedure, **3M** – 3 months after procedure, **6M** – 6 months after procedure, **1Y** – 1 year after procedure, **2Y** – 2 years after procedure).
Note: for some patients it was not possible to determine the value for technical reasons, significance was determined from pair values.

Table 5 Evaluation of change of maximum corneal curvature (MAX) at individual checks (D)

MAX (D)	Number of eyes	Average	Standard deviation	Range of values	Increase Number (%)	Stabilisation Number (%)	Reduction Number (%)	p - value
pred	84	50,39	4,17	43,20 – 63,10	-	-	-	-
1M	81	50,72	4,13	43,50 – 62,80	24 (30 %)	4 (5 %)	53 (65 %)	**p < 0,01**
3M	81	49,92	4,09	42,70 – 62,20	57 (70 %)	5 (6 %)	19 (23 %)	**p < 0,01**
6M	80	49,64	4,07	41,80 – 60,10	62 (78 %)	3 (4 %)	15 (16 %)	**p < 0,01**
1R	80	49,46	4,13	42,90 – 60,20	66 (83 %)	1 (1 %)	13 (16 %)	**p < 0,01**
2R	58	49,42	4,14	42,50 – 59,90	48 (83 %)	2 (3 %)	8 (14 %)	**p < 0,01**

(**before** - value before procedure, **1M** – 1 month after procedure, **3M** – 3 months after procedure, **6M** – 6 months after procedure, **1Y** – 1 year after procedure, **2Y** – 2 years after procedure).
Note: for some patients it was not possible to conduct en examination using an Orbscan II instrument due to advanced keratoconus, significance was determined from pair values.

their anamnesis (AY, AN), the incidence of corneal opacity is comparable at the individual checks. In groups divided according to age, the incidence of corneal opacity is comparable at the stage of 1-6 months, at 1Y it is lowest in the group of eyes of patients aged between 30-39 years (A3), but at 2Y there is a visible trend of increasing corneal opacity with increasing age. In groups divided according to stage of keratoconus according to the Amsler-Krumeich scale (AK1-AK3), in groups AK1 and AK2 the incidence of opacity decreases over time, whereas no decrease was recorded in group AK3. In groups of eyes divided according to wearing of hard contact lenses (CLY, CLN), there is a lower incidence

of corneal opacity at the checks up to 6 months in the group of eyes of patients wearing CL. At 6 months the incidence equals out, and at the 1Y and 2Y check eyes of patients who do not wear hard contact lenses have a lower incidence of corneal opacity. In the comparison of groups of eyes with a biomicroscopic symptom of keratoconus on the cornea (KY, KN), there was a lower incidence of corneal opacity in the group without manifestations of keratoconus at all the checks.

The complete data is presented in table 6 and graph 5.

Evaluation of incidence of corneal opacity according to success of CXL

Upon the division of eyes according to the success of CXL (P, S, R1, R2), corneal opacity was present in the 1st month in 100% of eyes in groups P and R2. After 1 year and 2 years the highest incidence was in group R2, where the highest regression of maximum corneal curvature was recorded.

Of two patients in whom progression (P) of the disease was recorded according to Koller (14), both had corneal opacity in the first month after CXL, in the 3rd month it was present only in one patient. Subsequently no opacity was recorded in this group up to the 2-year check, which may indicate that the cross-linking of collagen in this group was not sufficient in order to halt the progression of the disease.

With regard to the results, corneal opacity following CXL need not be strictly defined as a complication of therapy, but probably concerns a morphological correlative of realised cross-linking of collagen of the corneal stroma, which is more pronounced in more advanced stages of keratoconus.

Table 6 Incidence of corneal opacity in entire cohort (EC) and in groups of eyes of patients at individual checks after CXL (%)

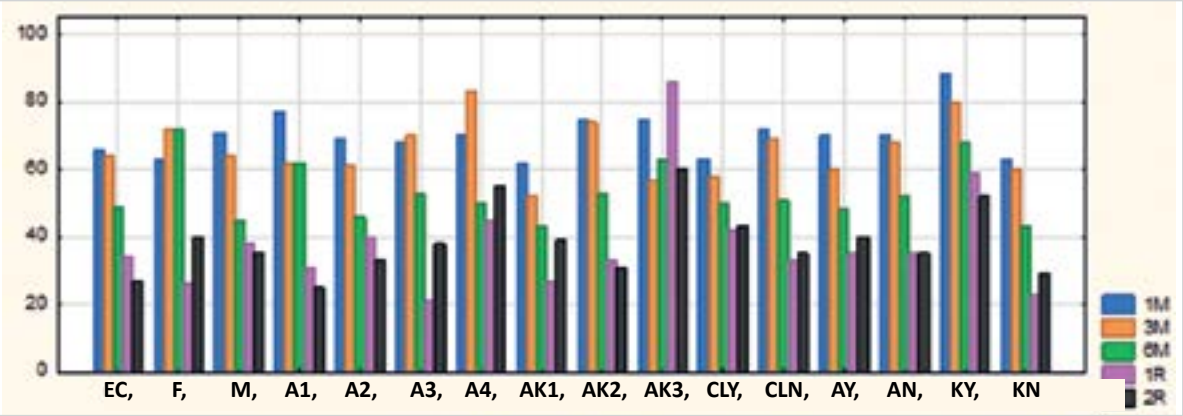
Zákal rohovky	Designation in graph	1M	3M	6M	1Y	2Y
Corneal opacity	CS	66 %	64 %	49 %	34 %	27 %
Entire cohort	F	63 %	72 %	72 %	26 %	40 %
Women	M	71 %	64 %	45 %	38 %	35 %
Men	V1	77 %	62 %	62 %	31 %	25 %
Age up to 19 years	V2	69 %	61 %	46 %	40 %	33 %
Age 20-29 years	V3	68 %	70 %	53 %	21 %	38 %
Age 30-39 years	V4	70 %	83 %	50 %	45 %	55 %
Age over 40 years	AK1	62 %	52 %	43 %	27 %	39 %
Stage I of Amsler-Krumeich scale	AK2	75 %	74 %	53 %	33 %	31 %
Stage II of Amsler-Krumeich scale	AK3	75 %	57 %	63 %	86 %	60 %
Stage III of Amsler-Krumeich scale	KCA	63 %	58 %	50 %	42 %	43 %
Wears CL	KCN	72 %	69 %	51 %	33 %	35 %
Does not wear CL	AA	70 %	60 %	48 %	35 %	40 %
Atopy present	AN	70 %	68 %	52 %	35 %	35 %
Atopy absent	KA	88 %	80 %	68 %	59 %	52 %

(**1M** – 1 month after procedure, **3M** – 3 months after procedure, **6M** – 6 months after procedure, **1Y** – 1 year after procedure, **2Y** – 2 years after procedure).

Table 7 Incidence of corneal opacity in groups of eyes of patients divided according to success of CXL at individual checks

Corneal opacity	Designation in graph	1M	3M	6M	1Y	2Y
Progression	P	100 %	50 %	0 %	0 %	0 %
Stabilisation	S	52 %	69 %	78 %	30 %	26 %
Regression up to 1.9D	R1	68 %	54 %	40 %	36 %	40 %
Regression more than 2D	R2	100 %	100 %	45 %	36 %	45 %

(**1M** – 1 month after procedure, **3M** – 3 months after procedure, **6M** – 6 months after procedure, **1Y** – 1 year after procedure, **2Y** – 2 years after procedure).



Graph 5 Incidence of corneal opacity in entire cohort and in groups of eyes of patients at individual checks after CXL (%)
(**1M** – 1 month after procedure, **3M** – 3 months after procedure, **6M** – 6 months after procedure, **1Y** – 1 year after procedure, **2Y** – 2 years after procedure).
(Note: graphic illustration table 6)

It persists even after 2 years in 36% of patients with successfully realised CXL, thus in patients with stabilised or reduced maximum corneal curvature.

The complete data is presented in table 7 and graph 6.

Evaluation of groups of eyes of patients divided according to presence of corneal opacity 2 years after CXL

Upon the division of eyes into 2 groups on the basis of the presence of corneal opacity 2 years after CXL, no statistically significant differences were recorded in BCVA, SE or MAX between the groups. Upon a comparison in the representation of age groups (A1-A4), in the group with the presence of corneal opacity the lowest number of eyes was recorded in patients aged up to 19 years (13%), and in the group without the presence of corneal opacity after 2Y the lowest number of eyes was in patients aged over 40 years (13%). In the group where no corneal opacity was present after 2Y, patients who had not had biomicroscopic symptoms of keratoconus before CXL predominated (75%), in contrast with the group with corneal opacity, where the proportion of eyes with and without symptoms of keratoconus was 48% v 52% respectively, thus almost equal (table 8).

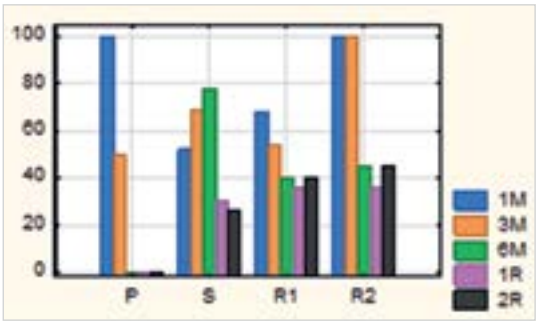
From the results it ensues that the greatest risk of persistence of corneal opacity 2 years after CXL is in patients aged over 40 years with biomicrocsopic symptoms of keratoconus.

Evaluation of incidence of other complications of CXL in cohort of patients

No incidence of sterile infiltrates of the cornea or endothelial failure of the cornea was recorded in the observed cohort of patient eyes.

Progression of the disease, defined according to Koller (13) as increase of the maximum curvature of the cornea by more than 1D, was recorded in two patients (3%).

Development of keratitis with corneal infiltrate occurred in one patient (1%) 13 months after CXL. Haemophilus influenzae was detected microbiologically from a swab from the conjunctival sac, and the presence of the HSV1 virus was confirmed (PCR method). Over the course of 5 days the condition of the cornea was rectified following the application



Graph 6 Incidence of corneal opacity in groups of eyes of patients divided according to success of CXK at individual checks (%)
(**1M** – 1 month after procedure, **3M** – 3 months after procedure, **6M** – 6 months after procedure, **1Y** – 1 year after procedure, **2Y** – 2 years after procedure).
(Note: graphic illustration table 7)

of local antibiotic therapy (levofloxacin 5 mg/ml solution 5x daily, ofloxacin ung 3.0 mg in 1g of grease before sleeping), but due to positive HSV1 the patient was also treated with antivirotics (anciclovir 0.15% 4x daily) for a period of 3 weeks. After infection there was no change of BCVA, SE or increase in the keratometric values in the patient, and the cornea remained without opacity.

Spontaneous detachment of the corneal epithelium within the scope of abrasion of the epithelium following CXL has not been published to date. We recorded this in one patient (1%) 1 month after CXL. After bandaging with contact lens, local therapy with artificial tears and preventive local antibiotic treatment (ofloxacin ung 3.0/ml solution 5x daily), the cornea was re-epithelialised and the condition did not repeat itself. One year after the procedure the patient underwent CXL also in the other eye, in which this complication was not repeated and healing took place without complications.

DISCUSSION

Wollensak et al. (37) recorded a significant improvement in

Table 8 Characteristics and comparison of certain observed parameters of patient eyes divided according to presence of corneal opacity 2 years after CXL

	Corneal opacity	No corneal opacity	p
Number of eyes	23	40	-
Women / men %	26 / 74	23 / 78	-
A1/A2/A3/A4 %	13 / 35 / 26 / 26	23 / 40 / 25 / 13	-
AK1/AK2/AK3 %	39 / 48 / 13	35 / 60 / 5	-
CLY/CLN %	74 / 26	80 / 20	-
AY/AN %	26 / 74	23 / 78	
KY/KN %	48 / 52	25 / 75	-
Age (average ± SD)	31.35 ± 11.83	27.03 ± 8.89	0.13
BCVA (average ± SD)	44.61 ± 9.56	44.33 ± 11.24	0.90
SE (average ± SD)	-5.09 ± 4.22	-4.90 ± 3.44	1.00
MAX (average ± SD)	49.48 ± 4.89	49.39 ± 3.76	0.85

(groups: A1 – age up to 19 years, A2 – age 20-29 years, A3 – age 30-39 years, A4 – age over 40 years, AK1, AK2, AK3 – keratoconus stages 1, 2 and 3 according to Amsler-Krumeich scale, CLN – hard contact lenses not worn, CLY – hard contact lenses worn, AY – with atopy, AN – without atopy, KY – with symptom of keratoconus on cornea, KN – no symptom of keratoconus on cornea; BCVA – best corrected visual acuity, SE – spherical equivalent, MAX – maximum corneal curvature).

BCVA 1Y after CXL in 65% of eyes, on average by 1.26 rows. Raiskup et al. (26) described a significant improvement at 1Y in 73.1% of eyes on average by 0.08 LogMAR and at 2Y a significant improvement in 81% of eyes on average by 0.09 LogMAR. Agrawal et al. (1) recorded an improvement by more than 1 row of Snellen's optotypes in 54% of eyes 1 year after CXL, and stabilisation of BCVA in 28% of eyes. Vinciguerra et al. (33) recorded an improvement in BCVA by 0.14 ± 0.08 LogMAR at 1Y after CXL. Caporossi et al. (4) describe an improvement in BCVA at 1Y by 1.34 ± 1.13 and at 2Y after CXL by 1.93 ± 1.04 rows on Snellen's optotypes. Asri et al. (3) in a cohort of 142 eyes describe an improvement in BCVA in 40.0% of eyes at 1Y, stabilisation in 47.6% and deterioration in 12% of eyes. Hersh et al. (11) recorded a significant improvement in BCVA in the entire cohort 1Y after CXL from 0.35 ± 0.24 LogMAR (20/45) to 0.23 ± 0.21 LogMAR (20/34), in 21.1% of eyes they described an improvement in BCVA by more than 2 rows of Snellen's optotypes and in 1.4% of eyes a loss of more than 2 rows.

In our cohort of patient eyes we confirmed the previous results of published studies and after 1M recorded a significant decrease of BCVA, and subsequently a significant improvement 1Y and 2Y after CXL, with the number of eyes with improvement increasing with time after the procedure. A comparison of the results is presented in table 9.

Wollensak et al. (37) and Raiskup et al. (26) also did not record a statistically significant difference between the IOP values before and 1Y after CXL. Coskuseven et al. (6) recorded a statistically significant increase of IOP at 9 months after CXL, from 9 ± 2 torr to 11 ± 2 torr. WittigSilva et al. (35) described a significant decrease of IOP measured by Goldmann applanation tonometry by 1.50 ± 0.44 torr 3 years after CXL. There was a decrease in thus measured IOP also at 1Y and 2Y, but this was not statistically significant. Upon measurement of IOP by a Tono-Pen instrument they did not record any significant change up to 3 years after CXL.

In the observed cohort of eyes there was no change in IOP up to 6 months after CXL, but after 1Y a significant decrease in the values was recorded and at 2Y a highly significant decrease in IOP values was recorded by the noncontact method.

From the results of the published studies it ensues that measurement of intraocular pressure by various methods and its evaluation in patients following CXL may be problematic, not only due to reduced corneal thickness in the case of keratoconus, but also due to the change to the rigidity of the cornea. In future it shall be necessary to conduct a complex study evaluating the various methods of measuring intraocular pressure in this group of patients, in order to prevent damage to the optic nerve by glaucoma disease. As a result, at present it is essential to ensure thorough and regular observation of the optic nerve in patients following CXL, because according to the results to date, screening measurement of intraocular pressure is imprecise.

Caporossi et al. (4) describe the measurement of lower pachymetry values after CXL using an Orbscan II instrument in comparison with measurement of corneal thickness by ultrasound, confocal microscopy or with OCT. Upon measurement of corneal thickness by the ultrasound method and confocal microscopy, they did not record significant differences between the values before CXL and the values at the individual follow-up examinations. Agrawal et al. (1) also did not record any significant change of corneal thickness measured by the ultrasound method at 1Y after CXL. Uopn evaluating the change of corneal thickness measured by ultrasound, Wittig-Silva et al. (36) did not record a significant change at the 1Y, 2Y and 3Y checks, and upon measurement using the Orbscan instrument recorded a significant decrease at all checks up to 3 years after CXL.

In our cohort of eyes we also measured corneal thickness by two independent methods. A borderline statistically sig-

Table 9 Comparison of change of best corrected visual acuity (BCVA) in entire cohort of patients after CXL with results presented in the literature.

1st author (year of publication)	Observation period	Change of BCVA (entire cohort)	% of eyes with improvement (average value)
Wollensak G. et al. (37) (2003)	1Y	-	65% (+ 1.26 rows)
Raiskup F. et al. (26) (2008)	1Y	-	73.1% (+ 0.08 LogMAR)
	2Y	-	81% (+0.09 LogMAR)
Agrawal V. et al. (1) (2009)	1Y	-	54% by more than one row
Vinciguerra P. et al. (33) (2009)	1Y	Improvement by 0.14 ± 0.08 LogMAR	-
Caporossi A. et al. (4) (2010)	1Y	Improvement by 1.34 ± 1.13 rows	-
	2Y	Improvement by 1.93 ± 1.04 rows	-
Asri D. et al. (3) (2011)	1Y	-	40.0%
Hersh P.S. Et al. (11) (2011)	1Y	Improvement from 0.35 ± 0.25 LogMAR to 0.23 ± 0.21 LogMAR	21.1% of eyes improvement by more than 2 rows of Snellen's optotypes
Wittig-Silva C. et al. (35) (2014)	1Y	Improvement by 0.09 ± 0.03 LogMAR	-
	2Y	Improvement by 0.09 ± 0.03 LogMAR	-
	3Y	Improvement by 0.09 ± 0.03 LogMAR	-
Strmeňová E. et al. (2014)	1Y	2.38 letters of ETDRS optotypes (corresponds to 0.048 LogMAR)	54%
	2Y	2.14 letters of ETDRS optotypes (corresponds to 0.043 LogMAR)	62%

nificant reduction in the pachymetry value measured by the ultrasound method was at the 3M and 6M check. At further checks there was no difference between the preoperative value and the value at the check. Upon measurement of corneal thickness using an Orbscan II instrument we recorded a significant reduction at all the follow-up examinations. At the 1M check the minimum value was 4 µm and at 3M 29 µm, which cannot be a realistic pachymetry value.

From our current and already published results (34), as well as the observations of other authors (1, 4, 34), it ensues that after CXL it is not suitable to determine corneal thickness using an Orbscan II instrument due to the imprecision of measurement.

Wollensak et al. (37) describe regression of the value of maximum corneal curvature at 1Y after CXL in 70% of eyes on average by 2.01D, they recorded stabilisation in 22% and progression in 5% of eyes. Raiskup et al. (26) recorded a reduction of maximum corneal curvature in 54% of eyes at the 2Y check by an average of 1.91D, and stabilisation (± 0.5D) in 35% of eyes. Agrawal et al. (1) recorded a reduction of maximum corneal curvature in 54% of eyes by an average of 2.47D and stabilisation in 38% of eyes. Koller et al. (14) observed flattening of the cornea following CXL in a study of 155 eyes, defined as a reduction of maximum corneal curvature by 1D. In 13% of eyes they recorded flattening of the cornea by more than 2D, in 60.3% of eyes MAX did not change and in 2% of eyes they recorded progression. Asri et al. (3) described a decrease of MAX by more than 2D in 21.3% of eyes. Hersh et al. (11) recorded a decrease of MAX by more than 2D in 31% of eyes and an increase by more than 2D in 4.2% of eyes. Wittig-Silva et al. (35) demonstrated a significant decrease of maximum corneal curvature at 1Y by 0.72 ± 0.15D, at 2Y by -0.96 ± 0.16 and at 3Y -1.03 ± 0.19D.

In our cohort of patient eyes we recorded a significant increase at 1M after CXL and subsequently from 6M onwards there was a significant reduction of maximum corneal curvature.

Our results confirm the findings of authors of already published studies, namely that stabilisation or a decrease of keratometric values takes place in the majority of eyes after CXL. A comparison of results is presented in table 10.

Wollensak et al. (37) after 1Y, Caporossi et al. (4) up to 2Y, Coskunseven et al. (6) after 9M and Wittig-Silva et al. (35) up to 3Y after CXL did not record a significant change in the density of endothelial cells, similarly to the situation in our cohort of eyes. In our cohort there was also no significant change in density or change of polymegathism and pleomorphism of endothelial cells up to 2 years after CXL.

Raiskup et al. (26), in a 6-year observation of 241 eyes of 130

Table 10 Comparison of change of maximum corneal curvature (MAX) in patients after CXL with results presented in the literature

1st author	Observation period	Change of MAX (entire cohort)	% of eyes with reduction (average value)	% of eyes with stabilisation (definition)	% of eyes with increase
Wollensak G. et al. (37 (2003)	1Y	-	70% (2.01D)	22%	5%
Raiskup F. et al. (26) (2008)	2Y	-	54% (1.91D)	35% (± 0.5D)	-
Agrawal V. et al. (1) (2009)	1Y	-	54% (2.74D)	38%	-
Caparossi A. et al. (4) (2010)	2Y	2.12 ± 0.65D	-	-	-
Koller T. et al. (14) (2011)	-	-	13% by more than 2D	60.3% (± 1D)	2% by more than 1D
Asri D. et al. (3) (2011)	1Y	-	By more than 2D in 21.3% of eyes	-	-
Hersh P.S. Et al. (11) (2011)	1Y	Significant reduction	By more than 2D in 31.0% of eyes	-	Increase by more than 2D in 4.2% of eyes
Wittig-Silva C. et al. (35) (2014)	1Y	-0.72 ± 0.15D	-	-	-
	2Y	-0.96 ± 0.16	-	-	-
	3Y	-1.03 ± 0.19D	-	-	-
Strmeňová E. et al. (2014)	1Y	0.93D	83%	1% (no difference)	16%
	2Y	0.96D	83%	3% (no difference)	14%

patients due to progressive keratoconus, did not record complications such as endothelial failure or cataract. They describe failure of treatment in 2 patients with neurodermatitis, and the progression of the disease was at the time of deterioration of the clinical picture of neurodermatitis. In both patients CXL was subsequently repeated without complications. Caporossi et al. (4) in an observation of patients 2 years after CXL describe stromal edema in 70% of patients up to 30 days after CXL, temporary corneal opacity in 9.8% of cases. They did not record any other complications up to two years after the procedure. Vinciguerra et al. (33) did not record any serious local or general complications in connection with CXL. 12.7% of eyes had corneal opacity at 1 month after CXL. Wittig-Silva (35), in a cohort of 46 eyes, recorded edema of the corneal stroma with small infiltrate in one eye at 1 month after CXL, and in one eye 2 days after CXL they recorded subepithelial infiltrate with signs of inflammation in the anterior chamber of the eye. Koller et al. (13), in a cohort of 105 eyes, determined that upon adjustment of the indication criteria for patients younger than 35 years with maximum corneal curvature up to 58D there is a reduction in the number of complications defined as loss of best corrected visual acuity and increase of maximum corneal curvature to less than 1%. They recorded sterile infiltrates in 7.6% of eyes and stromal scarring in 2.9% of eyes, but they did not succeed in identifying a predictive parameter of these complications. Vinciguerra et al. (32), Arora et al. (2) and Kankariya et al. (12) did not record any serious complications in paediatric patients who underwent CXL according to the Dresden protocol. The majority of published serious complications in patients after CXL are in the form of case reports (7, 9, 15, 16, 20, 21, 27, 30, 38, 39).

In our cohort of 86 eyes we did not record any serious complications up to 2Y after CXL. After 2 years 27% of eyes had corneal opacity, in two eyes (3%) we recorded progression of the disease after 2Y, thus an increase in the value of maximum corneal curvature by 1D. Our results are comparable with the published studies.

ZÁVER

V sledovanom súbore očí sme zaznamenali 1R a 2R po CXL signifikantné zvýšenie BCVA a zníženie všetkých keratometrických hodnôt, čím sme potvrdili úspešnosť CXL ako liečby progresívneho keratokónusu. Zmena pachymetrie meranej ultrazvukovou metódou, polymegatizmu, pleomorfizmu a hustoty endotelových buniek rohovky nebola v 1R a v 2R signifikantná. Zistili sme, že meranie hrúbky rohovky prístrojom Orbscan II a IOT bezkontaktnou metódou je nepresné u pacientov po CXL.

Najvyšší výskyt zákalu rohovky po 2 rokoch po CXL bol zaznamenaný u očí pacientov s III. štádiom keratokónusu podľa Amsler – Krumeichovej klasifikácie, starších ako 40 rokov, ktorí nosili tvrdú kontaktnú šošovku pred výkonom a mali biomikroskopické príznaky keratokónusu na rohovke.

Na základe našich výsledkov sme potvrdili, že CXL je účinný a bezpečný zákrok bez významných nežiadúcich účinkov lokálnych, alebo celkových.

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