

Unmasking keratoconus: effect of corneal epithelium in keratoconus

Desmascarando o queratocone: efeito do epitélio corneano no queratocone

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ABSTRACT

Objective: In the presence of an irregular cornea, like in keratoconus, the corneal epithelium has a different profile “masking” the irregular corneal stroma surface. Topographical analysis without considering the epithelium effect can result in an incorrect assessment, affecting the accuracy of any topography guided treatment. The aim of this study was to evaluate the “masking” effect of the corneal epithelium on corneal curvature in patients with keratoconus, comparing topographical findings before and after removal of the epithelium during Crosslinking (CXL). **Methods:** Five eyes of 5 patients with progressive keratoconus submitted to CXL according to the original Dresden protocol. Corneal topography was performed before and during the procedure (immediately after epithelium removal) and values of keratometry: K1, K2, mean K (Km), maximum corneal apical curvature (Kmax), corneal thickness and corneal astigmatism were evaluated. **Results:** The average values of corneal curvature before and after epithelial remove were: K1: 43.50±2.54D and 44.32±2.64D (p=0.080); K2 46.64±2.35D and 49.38±2.86D (p=0.043); Km 45.48±2.42D and 46.72±2.60D (p=0.042). It was also observed a significant increase in the maximum values of corneal apical curvature after removal of the epithelium (p=0.043). In central corneal thickness there was seen a reduction of 26.8µm from 524.8±33.0µm to 498.2±37.7µm (p = 0.042). **Conclusion:** There was seen a significant increase in the values of Kmax, K2 and mean K. This study demonstrates the masking effect of corneal epithelium on corneal curvature in keratoconus.

Keywords: Cornea; Keratoconus; Epithelium; Crosslinking; Corneal topography

RESUMO

Objetivo: Na presença de uma córnea irregular, como no queratocone, o epitélio tem uma distribuição diferente do normal, “mascarando” a irregularidade do estroma. A avaliação topográfica sem considerar o efeito do epitélio pode induzir uma avaliação menos correta, afetando a precisão de um eventual tratamento guiado por topografia. O objetivo deste trabalho é avaliar o efeito de “máscara” do epitélio no queratocone, comparando a tomografia corneana com e sem epitélio, no decurso da realização de Crosslinking (CXL). **Métodos:** Cinco olhos de 5 doentes com queratocone em progressão e submetidos a CXL segundo o protocolo original de Dresden. Foi realizada tomografia corneana antes e durante o procedimento (imediatamente após a remoção do epitélio), e avaliados os valores da queratometria - K1, K2, K médio (Km), ponto de máxima curvatura (Kmax), valores paquimétricos e astigmatismo corneano. **Resultados:** Os valores médios de curvatura corneanas antes e após a remoção do epitélio foram de: K1: 43,50 ± 2,54D e 44,32±2,64D (p=0,080); K2: 46,64±2,35D e 49,38±2,86D (p=0,043); Km: 45,48±2,42D e 46,72±2,60D (p=0,042). Observou-se igualmente um aumento estatisticamente significativo do valor de queratometria máxima após a remoção do epitélio (p=0,043). Na paquimetria central observou-se uma redução média de 26,8µm de 524,8±33,0µm para 498,2±37,7µm (p= 0,042). **Conclusão:** Observou-se um aumento estatisticamente significativo nos valores do Kmax, K2, e Kmédio após remoção do epitélio. Este estudo demonstra o efeito de “máscara” que o epitélio exerce no queratocone.

Descritores: Córnea; Ceratocone; Epitélio; Crosslinking; Topografia da córnea

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INTRODUCTION

Keratoconus is a bilateral, noninflammatory and slowly progressive corneal ectasia characterized by thinning and increased corneal curvature.⁽¹⁾ It is the most common corneal ectasia affecting one in every 2000 people.⁽²⁾ Changes in corneal collagen structures observed in keratoconus reduce corneal stability causing progressive protrusion typically with apex location in the lower temporal quadrant.^(2,3) Irregular astigmatism, progressive myopia, thinning of the cornea, and the onset of central leukomas are corneal changes induced by these corneal collagen alterations.^(1,4) Consequently, visual acuity and quality of vision deteriorate progressively and irreversibly.⁽¹⁾

Histopathological studies show that in the keratoconus all layers of the cornea are in some way altered, and a structural disorganization of lamellar collagen fibers are observed with significant and progressive alteration of their matrix.^(5,6) It is also demonstrated that the corneal epithelium in the apex is thinner in corneas with keratoconus when compared to normal corneas, with the corneal epithelium having a different profile and distribution in the presence of an irregular cornea in order to compensate or “mask” the stromal irregularity.^(1,7,8) In very advanced cases this decrease in thickness may even result in epithelial ruptures.^(7,8)

Some studies have already shown that the corneal epithelium has a different distribution in the presence of an irregular stroma surface.⁽⁸⁻¹¹⁾ Vinciguerra et al. demonstrated a “flattening” effect of the epithelium in patients with keratoconus.⁽¹¹⁾

Thus, evaluation of corneal progression and topography before considering refractive surgery without considering the effect of epithelium on CT and corneal topography may induce an incorrect evaluation affecting the accuracy of a possible keratorefractive treatment.⁽¹⁰⁾

The objective of the present study was to evaluate the effect of a “mask” of epithelium on keratoconus comparing corneal topography (Oculus Wavelight Pentacam® - Oculus, Wetzlar, Germany) with and without epithelium in the course of Crosslinking (CXL) in patients with progressive keratoconus.

METHODS

Retrospective study carried out in the Cornea section of the Ophthalmology Service of Centro Hospitalar do Porto, Portugal, between March and September 2016 with 5 eyes of 5 patients with progressive keratoconus subjected to CXL according to the original Dresden protocol. Exclusion criteria were the existence of corneal leukomas, previous corneal surgery, recent use of contact lenses (> 14 days), or connective tissue diseases. Patients or legal guardians provided an informed consent about the procedure and study, as well as permission to use clinical information for research purposes.

Keratoconus was diagnosed by an ophthalmologist making use of ophthalmologic examination and analysis of corneal topography (Orbscan IIz, Bausch & Lomb®) and corneal tomography (Oculus Wavelight Pentacam® - Oculus, Wetzlar, Germany). Of the selected cases, four were in stage I and one in stage II of the Amsler-Krumeich classification. The progression of keratoconus was defined by at least one of the following criteria in the last 12 months, and consecutive cases that met the following inclusion criteria were selected: increase of 1.00D or more in the keratometry value for the more curved axis, increase of 1.00D or more in the cylinder value for the

subjective refraction; increase of at least 0.50D in the spherical equivalent value for subjective refraction.

A corneal tomography was performed with the Scheimpflug camera system (Pentacam®-Oculus, Wetzlar, Germany) during the CXL procedure, all acquisitions performed by the same observer (L.O.) in the block before starting treatment and immediately after removal of the epithelium with a 9mm diameter automated Amoils® brush and centering on the pupillary axis. Only quality purchases (Qs - Quality Specifications) classified as “OK” were considered for analysis, and in 2 cases this classification was not obtained after the removal of the epithelium. The acquisitions not reaching this quality parameter were discarded, and the test was repeated until obtaining quality captations. The values of curvature of the central cornea - K1, K2, average K (Km), point of maximum curvature (Kmax), superior, inferior, nasal and temporal curvatures and pachymetry values 3mm from the center on anterior sagittal curvature maps, and anterior corneal astigmatism were evaluated, and demographic data was also collected.

The values are presented as average and standard deviation of the average. The normality of the results was evaluated by the Shapiro-Wilk test. The Wilcoxon test was used for paired samples for statistical analysis. Data was analysed with performed using the 23rd edition of SPSS software (IBM®, USA). A value of $P < 0.05$ was accepted as statistically significant.

RESULTS

The average age of our sample was 17.8 ± 5.6 years (11-25), with all patients being male.

The average values of corneal curvature before and after removal of the epithelium can be seen in table 1, where a statistically significant increase after removal of the epithelium in the average value of K2 from $46.64 \pm 2.35D$ (46.1 - 51.8) to $49.38 \pm 2.86D$ (47.3-54.4) - $p=0.043$; as well as in Km: $45.48 \pm 2.42D$ (43.9-49.7) to $46.72 \pm 2.60D$ (45.1-54.4) - $p=0.042$. A statistically significant increase of 4.06D in the value of the maximum keratometry point (Kmax) after removal of the epithelium was also observed: $55.76 \pm 4.40D$ (52.6-62.7) to $59.82 \pm 4.14D$ (55.1-64.7) - $p=0.043$. These differences were observed in all patients evaluated.

The analysis of the differences in keratometric values of K1 and superior, inferior, nasal and temporal curvatures at 3mm from the corneal center showed no statistically significant differences after removal of the epithelium (Table 1).

Regarding pachymetric changes - table 2 - central pachymetry showed an average reduction of central corneal pachymetry of $29.8\mu m$, from $528.0 \pm 38.5\mu m$ with epithelium to $498.2 \pm 37.7\mu m$ - $p=0.043$ after removal of the epithelium. The analysis of pachymetric alterations in the different quadrants - superior and inferior temporal and superior and inferior nasal, 3mm from the center of the cornea - showed statistical difference in all of them except in the inferior temporal quadrant (Table 2).

No complications, in particular infectious, were observed during the exams and procedures. A quality capture of all patients was possible for export and treatment.

The corneal tomography curvature maps show a significant increase in the keratometric values in the more curved meridian in the central 3mm, point of maximum curvature and average curvature without epithelium with non-significant reduction of the pachymetry value in the apex zone (inferior temporal) after the removal of the epithelium. (Figures 1A and B).

Table 1
Corneal keratometry values with and without epithelium

Kinetic Measurements	Cornea with epithelium			Cornea without epithelium			Average difference with Epithelium and without epithelium	P-value
	Average SD	Min	Max	Average SD	Min	Max		
K1, D	43.50 ± 2.54	41.4	47.7	44.32 ± 2.65	42.1	48.3	0.86	0.080
K2, D	46.64 ± 2.35	46.1	51.8	49.38 ± 2.86	47.3	54.4	2.74	0.043
Km, D	45.48 ± 2.42	43.9	49.7	46.72 ± 2.60	45.1	51.2	1.24	0.042
Kmax, D	55.76 ± 4.40	52.6	62.7	59.82 ± 4.13	55.1	64.7	4.06	0.043
K superior, D	40.42 ± 3.55	35.0	44.2	42.54 ± 3.98	36.7	47.6	2.12	0.080
K inferior, D	54.6 ± 4.42	50.6	61.6	55.62 ± 4.82	49.9	63.0	1.02	0.416
K nasal, D	42.44 ± 3.8	439.0	47.1	42.6 ± 6.22	36.2	51.9	0.16	0.893
K temporal, D	46.72 ± 2.2	243.4	48.8	49.12 ± 4.46	41.8	54.0	2.4	0.080
Anterior corneal astigmatism, D	4.12 ± 1.10	2.4	5.4	5.08 ± 1.89	2.2	6.5	0.96	0.080

Table 2
Values of corneal pachymetry with and without epithelium obtained by tomography

Pachymetric Measurements (µm)	Pre-CXL with epithelium			Pré-CXL without epithelium			Average difference with and without epithelium	P-value
	Average SD	Min	Max	Average SD	Min	Max		
Central	524.8 ± 33.0	488	568	498.2 ± 37.7	462	544	26.8	0.042
Temporal inferior	504.8 ± 33.0	459	535	488.4 ± 33.7	457	531	16.4	0.138
Temporal superior	541.1 ± 39.4	498	582	511.0 ± 37.4	468	552	30.4	0.043
Nasal inferior	527.2 ± 36.0	43.2	62.7	496.6 ± 34.0	457	541	30.6	0.043
Nasal superior	555.0 ± 40.8	35.0	44.2	515.0 ± 39.7	475	561	40	0.043

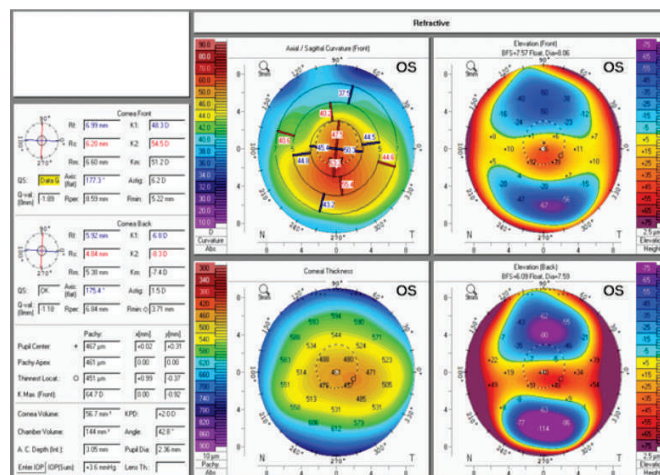
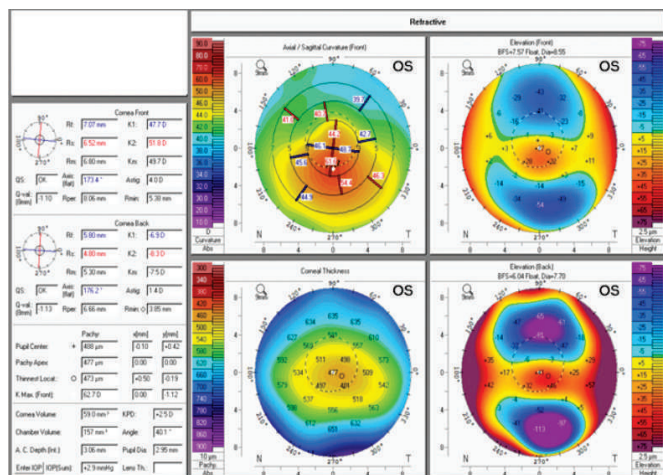


Figure 1 A: Corneal tomography (Pentacam) with epithelium

Figure 1 B: Corneal tomography (Pentacam) without epithelium

Figure 1. Corneal tomography (Pentacam) with (A) and without (B) epithelium. Increase keratometric values without epithelium and reduction of the central pachymetry value after removal of the epithelium are observed; the central thinning of the epithelium tries to compensate for true stromal ectasia.

DISCUSSION

In the present study, 5 eyes with progressive keratoconus were submitted to CXL according to the original Dresden protocol. After removal of the corneal epithelium in eyes with keratoconus, an increase in the average curvature (Km), apical curvature, (Kmax) and K2 was detected. Pachymetric alterations were also observed showing smaller epithelial thicknesses in the localization of the apex in the keratoconus eyes evaluated.

The epithelium affects the refractive power of the cornea, and consequently contributes to the total refractive power of the eye.⁽¹⁰⁾ This refractive effect is produced by the difference in refractive indexes between the tear film and air and the index between the epithelium and the stroma (1.40 vs 1.377).⁽¹²⁾ Vogt et al. described in 1921 the mask effect of the corneal epithelium in patients with irregular stroma surfaces.⁽¹³⁾ This compensatory mechanism was observed in all situations in which there is irregularity of the stromal surface (i.e.: patients with irregular

astigmatism, patients undergoing radiological keratotomies, leukomas or trauma).^(10, 11, 13)

It was also demonstrated that epithelial thickness plays an important role in refractive surgery and in its outcome.^(7, 14) Irregular distributions of the epithelium may induce an unpredictable surgical result due to the regularization effect of the epithelium on stromal irregularities.⁽¹⁴⁾ Simon et al. made keratometric measurements on 10 human eyes of an eye bank before and after removal of the epithelium demonstrating the change in refractive power of the cornea and in the axis of astigmatism after desepithelialization.⁽¹⁵⁾ It has been proposed that this epithelial mask effect should also be considered in the diagnosis and classification of patients with keratoconus.^(11, 12)

In summary, an increase in corneal curvature was observed after removal of the epithelium in corneas with keratoconus. Besides, the epithelial thicknesses of the points with the highest keratometric indexes were lower than in the flatter points. All of these signals show the effect of the epithelium in the total refractive power of the cornea, thus playing a regularizing role of stromal ectasia to induce a mask effect on the real curvature of the cornea in patients with keratoconus. Intraoperative keratometric changes of the epithelium were already analyzed in vivo by Vinciguerra et al.⁽¹¹⁾ in 28 eyes with progressive keratoconus submitted to CXL with topographic analysis before and after removal of the epithelium. As in our study, they observed an increase in the apical keratometric index (Kmax) and in keratometry of the more curved meridian (K2) after removal of the epithelium. These results were then proposed as signs of the mask effect and regularization of the corneal epithelium in the irregularities of the corneal stroma.⁽¹¹⁾

In our study, besides the previously mentioned results, the keratometric changes of the superior, inferior, nasal and temporal quadrants were also evaluated. Some studies have been carried out with the objective of detecting epithelial features or patterns in keratoconus corneas, and currently the corneal epithelium thickness and distribution profiles are proposed as useful in the diagnosis of keratoconus.^(16, 17) High frequency ultrasonography already showed that the smallest epithelial thickness was found in the inferior temporal quadrant in 74% of cases.⁽¹⁴⁾ In our study, the thickness of the epithelium measured in the inferior temporal quadrant - cone location in all patients in the study - was 16.4 μm , which was lower than in the other quadrants.

The mask effect of the epithelium is thus supported by the statistically significant differences between the pachymetry and keratometry values before and after epithelial removal observed in the present study, in which pachymetric and keratometric alterations were analyzed at 3mm from the cornea center, a location including the cone apex in all patients.

Despite the results reported, there are some limitations in our study. First, the small number of patients evaluated. Secondly, the thickness of the epithelium evaluated was obtained with a subtraction technique which is not a direct measure such as optical coherence tomography (OCT) or ultra-high frequency ultrasonography. Finally, the calculation of the thickness of the epithelium by this technique has limitations, presenting lower values in the present study than those already described in corneas with keratoconus.⁽¹⁷⁾ Thus, there may be changes in the pachymetry due to the process of hydration and dehydration of the cornea before and after removal of the epithelium. One of the possible uses of this knowledge and evaluation is in treatments guided by topography to treat the true corneal topography, and despite the greater difficulty of capture in the examination, all the patients of the present study showed quality capture for export and treatment.

However, in spite of these limitations we believe that the present study provides valid information on the effect of corneal epithelium in corneal topography in patients with keratoconus corroborating what is previously described in the literature. Thus, the authors recommend this approach to all patients with progressive keratoconus who do not have contraindication to CXL.

CONCLUSION

The present study showed topographic and pachymetric variations in patients with keratoconus, showing that the corneal epithelium affects the corneal curvature with a regularization effect of the stroma irregularity masking its true value and severity. With the removal of the epithelium, a statistically significant increase was observed in the values of Kmax, K2 and Kmedian. These differences may be partly explained by the nonuniform distribution of the epithelium in the irregular corneas, being thinner in the more curved areas of the stroma, with higher keratometric indexes, namely in the apex as demonstrated. As a result, the epithelium behaves as a regularizing factor of true stromal ectasia.

Thus, the evaluation of corneal tomography without considering the effect of the epithelium may induce a poor diagnosis, evaluation and classification of keratoconus, affecting the accuracy of any topographic treatment or even hiding the progression of the disease.

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