Role of corneal epithelial thickness mapping in the evaluation of keratoconus

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ABSTRACT

Purpose: To investigate the corneal epithelial thickness profiles in patients with a confirmed diagnosis of stable and progressive keratoconus.
Setting: Studio Italiano di Oftalmologia, Rome, Italy.
Design: Observational study.
Methods: 86 patients with either stable (n = 52) or progressive (n = 34) keratoconus and 182 healthy controls were enrolled in the study. Disease progression was confirmed by repeated corneal topographies over 1 year follow-up before inclusion in the study. All subjects had full corneal and epithelial thickness mapping taken by spectral domain optical coherence tomography (SD-OCT). The full corneal mapping was investigated by evaluating the central corneal thickness, the thinnest point, the superonasal-inferotemporal thickness difference and the minimum-median thickness difference. The epithelial mapping was investigated by assessing the 2 mm central thickness, the inferior paracentral (2–5 mm) thickness, and the minimum-maximum thickness difference.
Results: No significant differences in full corneal mapping were found between stable and progressive keratoconic eyes. Of note, the inferior paracentral region of the corneal epithelium was significantly thinner in progressive ($50 \pm 3\,\mu m$) than stable ($53 \pm 4\,\mu m$) keratoconus (P < 0.001).
Conclusions: The SD-OCT corneal epithelial mapping was valuable for detecting local thickness changes in eyes with keratoconus. Monitoring the corneal epithelial changes across the inferior area in patients with keratoconus could be worthy for assessing disease progression.

1. Introduction

Keratoconus is the most frequent degenerative disease of the cornea [1,2]. It is widely known that the disease is strongly correlated with stromal and epithelial thickness changes [3]. The changes of corneal epithelium have been hypothesized to develop in the sub-clinical stage of keratoconus [4–7]; in addition, these alterations may occur even in eyes that do not show severe abnormalities with corneal topography [6,8].

Optical Coherence Tomography (OCT) and Very High-Frequency digital UltraSound (VHF-US) have been the most commonly used tools for mapping the corneal epithelial thickness. In general, OCT and VHF-US have been shown to be in close agreement for epithelial thickness measurement, although OCT usually provided a central epithelial thickness 2–5% thinner than VHF-US in two thirds of cases [9,10].

Studies on normal population have shown moderate or high variability of the central, paracentral, and midperipheral corneal epithelial thickness values in subjects of different age and gender [11]. Since significant differences in the corneal epithelial thickness profile have been found even in normal eyes, there is need of studies with large sample size in order to understand whether the investigation of corneal epithelial mapping could be valuable to detect pathologic changes in eyes with keratoconus and, most importantly, to assess disease progression. Although a number of studies [5,12–14] has shown the accuracy of OCT imaging in assessing the variations of epithelial thickness between patients with keratoconus and healthy subjects, there is no evidence on its role on monitoring diseases progression.

The scope of the present study was to investigate the full corneal and epithelial thickness profiles in subjects with a confirmed diagnosis of stable or progressive keratoconus; a large group of healthy subjects was enrolled in the study and data used as controls.

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2. Patients and methods

Patients with confirmed diagnosis of stable or progressive keratoconus and healthy subjects were invited to participate in this study. Diagnosis of keratoconus was made by two expert corneal specialists (M.L. and S.S.) by complete eye examination, which included corneal topography, corneal pachymetry, slit-lamp bio-microscopy and manifest refraction. Keratoconus was deemed to be progressive if there was an increase of at least 1 diopter (D) in maximum simulated keratometry value (K_{max}) derived by computerized Placido disk corneal topography over 1 year preceding the present study.

Exclusion criteria for this study included any ocular disorder except for keratoconus (e.g., cataract, glaucoma, herpetic keratitis, retinal diseases), previous corneal excimer or femtosecond laser or intra-ocular surgery, and corneal scarring or epithelial defects. Patients with K_{max} greater than 61 D were also excluded from the study. Participants who were pregnant or breastfeeding at the time of enrollment were also excluded. Contact lens wearers were instructed to discontinue their use for a minimum of 3 weeks before the eye examination [12–14]. After participating in this study, all the patients with progressive keratoconus underwent riboflavin/UV-A corneal cross-linking.

All subjects underwent complete eye examination, including corrected distance visual acuity (CDVA, logMAR units), combined Placido disk-Scheimpflug topography (Sirius, CSO, Scandicci, Italy) and corneal thickness mapping by AS-OCT (Optovue, Fremont, CA USA). Only the right eye for each subject was used for corneal mapping analysis. The investigated parameters were the central corneal thickness ("CCT"), the thinnest point, the superonasal-inferotemporal thickness difference ("SN-IT 2–5 mm") and the minimum-median thickness difference ("min-med"). The corneal epithelial mapping was investigated by assessing the central thickness (mean of epithelial thickness values across the 2 mm central area), the inferior paracentral thickness ("I 2–5 mm"; mean of thickness values recorded in the inferior epithelium 2–5 mm from the center), and the minimum-maximum thickness difference ("min-max"; difference between minimum and maximum epithelial thickness).

2.1. Statistical analysis

Statistical analysis was performed using SPSS (ver. 17, IBM Corp, NY). All data were reported as the mean ± standard deviation. Normal data distribution was tested by using the one-sample Kolmogorov-Smirnov test. The Tukey-Kramer test was used to compare data between groups.

The relationship between the corneal thickness mapping data and baseline parameters (age, gender, progression of keratoconus) in either group was assessed using Pearson's correlation analysis.

Sample size calculation was performed to detect differences of 15 μm and 2 μm between the average corneal (CCT) and epithelial (central) thickness for keratoconus and controls respectively, at a significance level of 5% and a power of 82%, assuming a standard deviation of 10%. The minimum sample size of this study was 260 participants/eyes (allocation ratio of 1:2).

3. Results

The study included 268 eyes of 268 subjects. Eighty-six (86) eyes of 86 patients had a diagnosis of keratoconus; 52 of these had stable keratoconus and 34 had progressive keratoconus. One hundred eighty two (182) healthy eyes of 182 subjects were enrolled as controls (Table 1).

All the full corneal thickness values, such as CCT, thinnest point, SN-IT 2–5 mm and min-med values were significantly thinner (P < 0.001) in keratoconus (either stable or progressive) than controls. No significant differences in full corneal thickness profiles were found between stable and progressive keratoconic eyes. Corneal full thickness

mapping data are summarized in Table 2.

The central epithelial thickness and the min-max value were significantly thinner (P < 0.001) in keratoconic eyes than controls; in addition, the paracentral inferior epithelial thickness was significantly thinner (P < 0.001) in progressive keratoconus than both stable keratoconus and controls. Corneal epithelial thickness mapping data are summarized in Table 3.

Males had thicker CCT than females both in controls (P = 0.03) and keratoconic eyes (P = 0.04); there were no significant gender-related differences in any other full corneal thickness parameter. The central epithelial thickness was thicker in males than females both in keratoconic (P < 0.001) and control eyes (P < 0.001); the paracentral inferior epithelial thickness was significant thicker in males than females in controls (P < 0.01) and keratoconus (P < 0.01).

The CCT (R = 0.18; P = 0.02) and thinnest point (R = 0.15; P = 0.04) moderately correlated with aging in healthy eyes; the paracentral inferior epithelial thickness (R = 0.26; P = 0.02) moderately correlated with aging in keratoconus.

4. Discussion

This study investigated the full corneal and epithelial thickness mapping in patients with a confirmed diagnosis of stable and progressive keratoconus. A large cohort of healthy controls was enrolled in order to have a powerful control dataset of epithelial mapping values, which was lacking in the scientific literature.

Contact lens wearers (24% of the overall population) were recommended to discontinue their use for a period of at least 3 weeks before eye examination in order to minimize methodology bias. In previous studies [12–14], a 2-week contact lens free period has been shown to be adequate for the cornea to stabilize.

The analysis of full corneal thickness provided significant differences in the measurements of CCT, thinnest point, SN-IT 2–5 mm values and minimum-median difference values between controls and keratoconus, as expected. The full thickness corneal mapping did not discriminate between eyes with stable or progressive keratoconus. On the other hand, the corneal epithelial mapping analysis provided valuable information about the local differences between eyes with stable and progressive keratoconus. The paracentral (2–5 mm) inferior epithelial thickness was significantly thinner in progressive than stable keratoconus.

In keratoconus, the epithelium usually thins over areas of relative increases in curvature and thickens over areas of relative flattening; the focal epithelial thinning at the apex of the cone, which is in general located across the paracentral inferior area, is likely a direct response to anterior curvature and profile [13,15]. Zhou et al. [3] have investigated the epithelial and stromal thickness distributions in eyes with keratoconus and healthy astigmatic eyes; they have found that the epithelial and stromal thicknesses were distributed similarly and the epithelium seemed to compensate for stromal anterior surface shape irregularities. The same findings have been found in several studies using different imaging instruments [6,13,15–18]. Catalan et al. [19] have compared the OCT corneal epithelial maps between 22 eyes with unilateral keratoconus (i.e., the fellow eyes did not show any clinical sign of keratoconus) and 104 healthy eyes. The authors have found significant differences in the minimum epithelial thickness between keratoconus and control eyes. Schallhorn et al. [20] have compared the OCT epithelial maps of keratoconus with those taken in forme fruste keratoconus, contact lens wearers and healthy eyes, showing a typical pattern of inferior thinning and superior thickening in keratoconus only. Similarly, Li et al. [5] have evaluated the corneal epithelial mapping in eyes with subclinical keratoconus (n = 50) showing a typical inferotemporal thinning and a relative superior-nasal thickening in comparison with controls (n = 150). Reinstein et al. [21] have shown – by using VHF-US – a doughnut pattern of the corneal epithelial profile, which was slightly decentered infero-temporally, in keratoconus. The

Table 1

Participants' demographics and characteristics. Data are given as M ± SD.

Groups	Age (years)	CDVA (LogMAR)	Manifest Refraction (D)	K_{max} (D)
Keratoconus (all patients; n = 86; 25F and 61M)	38 ± 13	0.14 ± 0.15	− 4.25 ± 2.82	54.89 ± 6.01
Stable keratoconus (n = 52; 17F and 35M)	42 ± 14	0.14 ± 0.16	− 4.62 ± 2.88	54.98 ± 6.96
Progressive keratoconus (n = 34; 8F and 26M)	32 ± 8	0.14 ± 0.14	− 3.37 ± 2.41	55.00 ± 4.01
Controls (n = 182; 110F and 72M)	37 ± 10	0.00 ± 0.01	− 3.12 ± 2.18	42.44 ± 1.20 (K_{sim})

M = male; F = female.
CDVA = corrected distance visual acuity.
K_{max} = maximum simulated keratometry.
K_{sim} = simulated keratometry.
D = diopter.

Table 2

Corneal thickness values (µm; M ± SD) in stable and progressive keratoconus and control eyes.

	Stable Keratoconus	Progressive Keratoconus	Controls
Central corneal thickness (CCT)	479 ± 46[*]	477 ± 41[*]	532 ± 31
Thinnest point	456 ± 43[*]	444 ± 46[*]	523 ± 31
Superonasal-inferotemporal thickness difference (SN-IT 2–5 mm)	54 ± 25[*]	62 ± 27[*]	27 ± 13
Minimum-median thickness difference (min-med)	− 53 ± 40[*]	− 58 ± 19[*]	− 23 ± 7

* (P < 0.001) Tukey-Kramer between study group and control group.

Table 3

Corneal epithelial thickness values (µm; M ± SD) in stable and progressive keratoconus and controls.

	Stable Keratoconus	Progressive Keratoconus	Controls
Central (2 mm)	51 ± 5[*]	50 ± 4[*]	53 ± 3
Min-Max	− 17 ± 8	− 18 ± 6	− 7 ± 2
I (2–5 mm)	53 ± 4	50 ± 3[*,#]	54 ± 3

Central = 2 mm central epithelial thickness.
Min-max = minimum-maximum thickness.
I 2–5 mm = inferior epithelial thickness across 2–5 mm from the center.
* (P < 0.05) Tukey-Kramer between study group and control group.
# (P < 0.05) Tukey-Kramer between stable and progressive keratoconus groups.

authors have shown that the epithelium at the thinnest point is on average 7 µm thinner than at the corneal vertex and that the minimum epithelial thickness is thinner in eyes with steeper keratometry.

Histopathology studies in keratoconus have revealed that the epithelial thickness is highly variable across the central and peripheral regions, ranging from 13 µm to 92 µm (averagely 42 µm) and from 30 µm to 91 µm (averagely 55 µm), respectively [22,23]. At the cone apex, the usual 4–6 layers of epithelial cells vary substantially and may increase to up to 10–15 layers of epithelial cells, with the earliest detectable changes that occurs in the epithelial basal layer [24]. Rocha et al. [25] have evaluated, by using OCT, the epithelial thickness profile in 120 eyes showing that the average epithelial thickness at the highest topography point was significantly thinner in eyes with keratoconus and corneal ectasia than in control eyes. This study demonstrated that the paracentral inferior epithelial thickness is significantly thinner in progressive than stable keratoconus and would be suitable as clinical parameter for monitoring disease progression in combination with CCT and K_{max} [26,27].

Aging and gender differences in corneal epithelial thickness profile in healthy subjects have been confirmed in this study. In general, the corneal epithelial thickness is thicker in men than in women and the epithelial thickness across the central regions of the cornea is moderately correlated with aging [11,28]. It was of note in this study to highlight that the paracentral inferior epithelial thickness was moderately correlated with aging in patients with keratoconus. It is likely that aging may be inversely correlated with severity of disease progression, as hypothesized in previous studies [29,30].

In conclusion, the present data would be valuable in widening the understanding on the clinical importance of corneal epithelial mapping in keratoconus providing specific information on greater thinning of the inferior corneal region in eyes with progressive than stable keratoconus; this notion may be worthy for monitoring disease progression when used in combination with already known biomarkers of disease progression, such as CCT and K_{max}. Studies with large sample size, as the present one, and longitudinal design would be valuable for improving the diagnostic power of indices constructed from corneal epithelial thickness maps [5,6,17,31–33].

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Conflict of interest

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