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an increase in ipsilateral fusional vergence, the finding was statistically significant for both distance and near vision (p = 0.01 and p = 0.02 respectively). By contrast, we did not succeed in demonstrating this fact in the group of patients with esophoria. In our study we also demonstrated a statistically significant differences in the values of the AC/A ratio

measured with the gradient and heterophoric method. The values determined by the gradient method are lower (3.0  $\pm 1.1$  pD/D versus 5.8  $\pm 0.9$  pD/D) than in the case of the heterophoric method. The difference is caused by the absence of a proximal convergence component in the case of determining the AC/A ratio by the heterophoric method.

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## DIAGNOSTIC IMPORTANCE OF OCT PACHYMETRY IN KERATOCONUS

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#### **SUMMARY**

**Purpose:** To evaluate the value of AS OCT pachymetry as a method capable of detecting early differences between keratoconus, latent keratoconus and corneal astigmatism based on measurements of the parameters of corneal epithelial thickness and total corneal thickness.

**Methods:** This study analyzed 162 eyes of 89 patients examined with a Zeiss Cirrus 500 Anterior Segment Premier Module. OCT Pachymetry maps were created in 97 eyes with keratoconus, 33 eyes with latent (forme fruste) keratoconus, and 32 eyes with regular corneal astigmatism (≥ 1.5 Dcyl). The parameters of epithelial thickness (central epithelial thickness in the 2 mm zone, paracentral epithelial thickness in the 2–5mm zone, minimal and maximal epithelial thickness) and total corneal thickness (S-I in the 2–5 mm zone, SN-IT in the 2–5 mm zone, minimal thickness, max-min thickness) were analyzed in all pachymetry maps.

**Results:** Statistically significant differences were determined in 3 parameters of epithelial thickness (paracentral epithelial thickness in the 2–5 mm zone, minimal epithelial thickness, maximal epithelial thickness) between group A and group B (p < 0.001), as well as between group A and group C (p < 0.001). Statistically significant differences were determined in 3 parameters of total corneal thickness (S-I in the 2–5 mm zone, SN-IT in the 2–5 mm zone, minimal thickness) between group A and group B (p < 0.001), between group A and group C (p < 0.001), as well as between group B and group C (p < 0.001).

**Conclusion:** AS OCT Pachymetry maps are a reliable method capable of detecting differences between keratoconus and corneal astigmatism based on the comparison of paracentral epithelial thickness in the 2–5 mm zone, minimum epithelial thickness, and maximum epithelial thickness. Furthermore, based on the evaluation of the parameters of total corneal thickness, it is a method capable of defining the differences between keratoconus, latent keratoconus and corneal astigmatism (S-I in the 2–5 mm zone, SN-IT in the 2–5 mm zone and minimum thickness). In the statistical analysis, the most reliable parameters appear to be: the difference between groups A, B and C in the parameters S-I in the 2–5 mm paracentral zone, SN-IT in the 2–5 mm paracentral zone and in the values of minimum corneal thickness.

Key words: anterior segment OCT, keratoconus, OCT pachymetry

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#### **INTRODUCTION**

Keratoconus is a bilateral, progressive, non-inflammatory disease of the cornea. Although the etiopathology of this disease remains unclear, a two-hit hypothesis has been described, according to which genetic predisposition as the first (endogenous) factor is followed by eye rubbing as the second (exogenous) factor [1]. Together they contribute to corneal thinning, an increase of corneal steepness and the subsequent onset of irregular astigmatism, accompanied by a deterioration of visual acuity [2]. Anterior segment optical coherence tomography (AS

OCT) is an imaging method enabling the creation of corneal pachymetry maps, which analyze total corneal thickness as well as corneal epithelial thickness [3–5].

It remains unclear as to which corneal changes in keratoconus occur first (corneal thickness, curvature of the anterior surface of the cornea or curvature of the posterior surface of the cornea). However, the capacity of the corneal epithelium to compensate through its restructuring for the majority of initial changes is generally known [6,7]. This concerns typical corneal thinning at the apex and thickening of the epithelium in the surrounding area of the base of corneal ectasia. Changes of corneal

topography subsequently appear over time, when the compensatory mechanisms of the corneal epithelium have been overpowered by the pathology [8]. However, determining early ectatic changes may be of fundamental significance, especially before planning laser corneal refractive surgery.

The aim of this study was to evaluate the significance of AS OCT pachymetry as an examination method which is capable of identifying early differences in the parameters of corneal epithelial thickness and total corneal thickness between keratoconus, latent keratoconus and corneal astigmatism without ectatic changes.

#### **MATERIAL AND METHODS**

The examined cohort comprised a total of 162 eyes of 89 patients (27 women and 62 men) within the age range of 16 to 61 years. The patients were examined at UVEA Klinika s.r.o. in Martin in the period from June 2019 to June 2021. The auxiliary parameters in the observed group of eyes covered uncorrected visual acuity (UCVA) and corrected distance visual acuity (CDVA) examined on the optotype Topcon CC-100XP (Topcon Corporation), as well as keratometry (K<sub>1</sub> and K<sub>2</sub> as a designation of the least steep and steepest corneal meridian) corneal topography reading on Schwind Sirius (Schwind eye-tech-solutions), spherical equivalent (SE) and value of cylindrical refraction component (Cyl) according to measurement by the automatic refractokeratometer Nidek ARK-1 (Nidek Co.,

Ltd.). On the basis of these parameters, 3 groups of eyes were evaluated: A – 97 eyes with clinically manifest keratoconus (stage 1–3 according to the Amsler – Krumeich classification), B – 33 eyes with latent keratoconus (or more precisely "forme fruste", since these were second eyes of patients with manifest disease in the first eye, which was classified in group A), and C – 32 eyes with regular corneal astigmatism of  $\geq$  1.5 D. The auxiliary instrument Zeiss Cirrus 500 with the Anterior Segment Premier Module for corneal imaging was used to create corneal pachymetry maps, which were subsequently analyzed in the "Pachymetry" imaging mode.

The imaging mode entitled "Pachymetry" creates a complex corneal pachymetry map, which provides an analysis of total corneal thickness and also specifically of corneal epithelial thickness (Fig. 1,2,3). Scanning is performed under the conditions of optimal centering of the lens of the instrument to the center of the pupil, and approximation to the patient's corneal surface. The output is an analysis of the thickness of the cornea and of the corneal epithelium in the form of maps created by one central, eight paracentral and eight peripheral concentrically oriented segments. The analyzed parameters of corneal epithelial thickness were: median corneal epithelial thickness in central zone 2 mm, median corneal epithelial thickness in paracentral zone 2-5 mm, minimal corneal epithelial thickness and maximal corneal epithelial thickness. The analyzed parameters of total corneal thickness were: difference in corneal thickness between superior

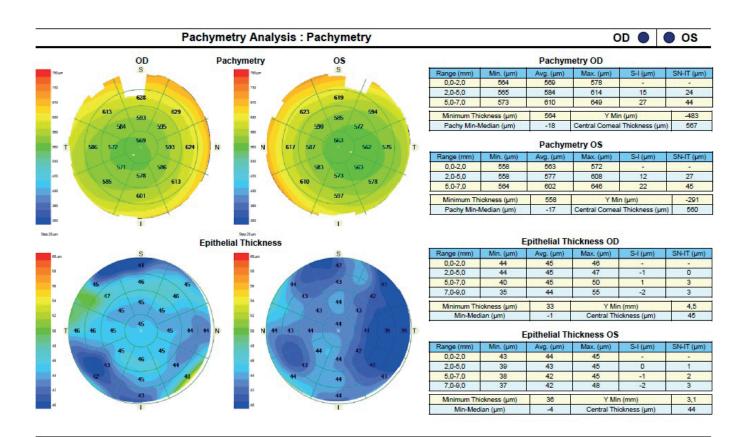


Figure 1. OCT Pachymetry of corneal astigmatism. Note the relatively homonymous distribution of corneal epithelial thickness

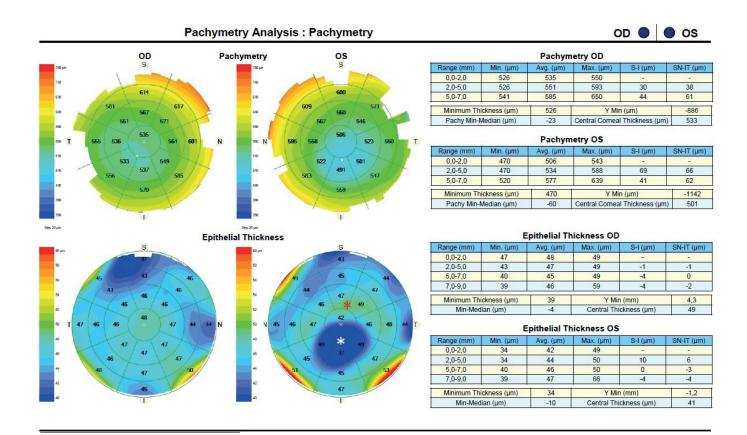
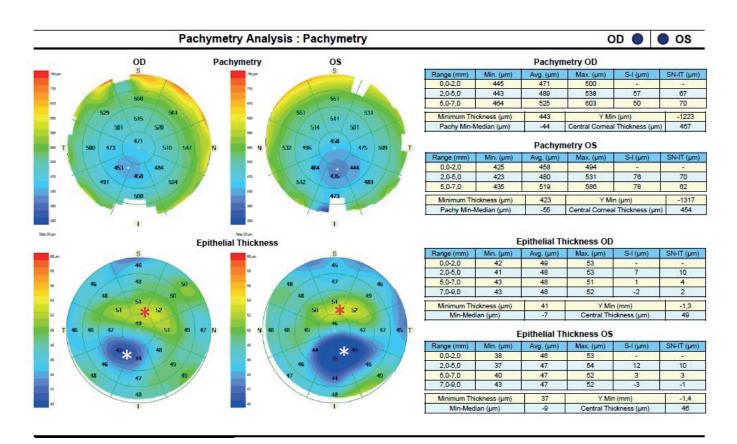


Figure 2. OCT Pachymetry of keratoconus – latent in the right eye and clinically manifested in the left eye. White asterisk epithelial thinning on the apex of the cone, red asterisk – epithelial thickening near the base of the cone



**Figure 3.** OCT Pachymetry of keratoconus – clinically manifested in both eyes. White asterisk – epithelial thinning on the apex of the cone, red asterisk – epithelial thickening near the base of the cone

and inferior quadrant in 2–5 mm paracentral zone (designation S-I), difference in corneal thickness between nasal and inferior quadrant in 2–5 mm paracentral zone (designation SN-IT), minimal corneal thickness and difference between maximal and minimal corneal thickness (max-min).

The statistical software SYSTAT (Systat Software Inc.) was used. For the statistical analysis of the corneal pachymetry maps, non-Gauss distribution of the observed cohort of eyes was initially confirmed with the aid of a Shapiro-Wilk test. On this basis, we continued to work with nonparametric tests in the statistical analysis. For the evaluation of the presence of differences between the groups (A – keratoconus, B – latent keratoconus, C – astigmatism) we used a Kruskal-Wallisov test. In the case that we determined a statistically significant difference, as a post hoc test we used the Dwass-Steele-Critch-low-Flinger test. We also tested groups A, B and C for the presence of differences in age and sex (Kruskal-Wallisov test, Chi-squared test).

#### **RESULTS**

No difference was found between the 3 evaluated groups (A, B, C) in terms of the sex (p = 0.927) or age of the observed patients (p = 0.946). The auxiliary parameters analyzed in the cohort are presented in Table 1.

Through a detailed evaluation of the parameters of corneal epithelial thickness we determined the following results. Median corneal epithelial thickness in the central 2 mm zone of the cornea in group A was 45.0 µm, in group B 48.0 μm and in group C 50.5 μm. We evaluated the difference between groups A and B (p < 0.001) and between groups A and C (p < 0.001) as statistically significant, but not between groups B and C (p = 0.143) (Fig. 4). Median corneal epithelial thickness in the paracentral 2–5 mm zone of the cornea in group A was 47.0 μm, in group B 47.0 μm and in group C 49.0 μm. We evaluated the differences in the values between the individual groups as insignificant (p = 0.097), and as a result no post hoc test was conducted for this parameter (Fig. 5). Median minimal corneal epithelial thickness in group A was 33.0 µm, in group B 35.0 µm and in group C 35.0 µm. We evaluated the difference between groups A and

B (p < 0.001) and between groups A and C (p < 0.001) as statistically significant, but not between groups B and C (p = 0.576) (Fig. 6). Median maximal corneal epithelial thickness in group A was  $60.0 \, \mu m$ , in group B  $58.0 \, \mu m$  and in group C  $56.0 \, \mu m$ . In this parameter also, we evaluated the difference between groups A and B (p < 0.001) and between groups A and C (p < 0.001) as statistically significant, but not between groups B and C (p = 0.714) (Fig. 7).

Through a detailed evaluation of the parameters of total corneal thickness we then determined the following results. The median value of the difference of total corneal thickness in the superior (S) and inferior (I) segment in the paracentral 2–5 mm zone in group A was 55.0 µm, in group B 37.0 μm and in group C 20.0 μm. We evaluated the difference between groups A and B (p < 0.001) and between groups A and C (p < 0.001) as statistically significant, and in this case the difference between groups B and C (p = 0.003) was also statistically significant. By far the most pronounced dispersion of values was recorded in group A, which corresponds to the typically diverse asymmetry in corneal thickness in keratoconus (Fig. 8). The median value of the difference of total corneal thickness in the superior nasal (SN) and inferior temporal (IT) segment in the paracentral 2–5 mm zone in group A was 59.0  $\mu$ m, in group B 45.0  $\mu$ m and in group C 32.0  $\mu$ m. We evaluated the difference between groups A and B (p < 0.001) between groups A and C (p < 0.001), and in this case also between groups B and C (p < 0.001) as statistically significant. Similarly, as in the case of previous S-I parameter, in the SN-IT parameter also, the largest dispersion of values was determined in group A (Fig. 9). The median value of minimal total corneal thickness in group A was 446.0 µm, in group B 498.0 µm and in group C 536.5 µm. In this parameter we determined a statistically significant difference between groups A and B (p < 0.001), between groups A and C (p < 0.001) and between groups B and C (p < 0.001) (Fig. 10). The median value of the difference of maximal and minimal total corneal thickness in group A was 163.0 µm, in group B 135.0 μm and in group C 116.5 μm. In this parameter we determined a statistically significant difference between groups A and B (p < 0.001), and between groups A and C (p < 0.001). The difference between groups B and C was borderline significant (p = 0.057) (Fig. 11). Within

**Table 1.** Characteristics of the group of eyes – data are presented as median (interquartile range)

Auxiliary parameters										
	UDVA CDVA		K <sub>1</sub> (D)	K <sub>2</sub> (D)	SE (D)	Cyl (D)				
A	0.2	0.7	44.75	47.25	-4.43	-2.62				
	(0.05–0.6)	(0.45–0.9)	(43.50–48.12)	(46.12–52.,62)	(-7.53 až -2.,12)	(-4.62 až -1.62)				
В	0.7	1.0	43.0	44.87	-0.75	-1.5				
	(0.2–1.0)	(0.8–1.0)	(42.0–44.25)	(43.75–46.75)	(-3.0 až 0)	(-3.0 až -0.5)				
С	0.35	1.0	43.0	44.87	0.9	-2.25				
	(0.08–0.5)	(0.8–1.0)	(41.75–44.0)	(44.25–46.62)	(-3.62 až -0.81)	(-3.5 až -1.5)				

A – keratoconus, B – latent keratoconus, C – astigmatism, UDVA – uncorrected distance visual acuity, CDVA – best corrected distance visual acuity, K<sub>2</sub> – keratometry in the flattest and steepest corneal meridian, SE – spherical equivalent, D – diopters

the framework of the statistical analysis, on the basis of the results the most reliable parameters appear to be the following: difference between groups A, B and C in the parameters of S-I in the 2–5 mm paracentral zone, in SN-IT in the 2–5 mm paracentral zone and in the values of minimal corneal thickness (Table 2,3).

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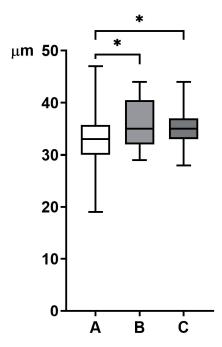
**Graph 1**. Corneal epithelial thickness in the 2mm central zone of the cornea. Results are presented as median (interquartile range). Asterisk – differences between groups were statistically significant

A – keratoconus, B – latent keratoconus, C – astigmatism, \* – p < 0,001

A

В

C

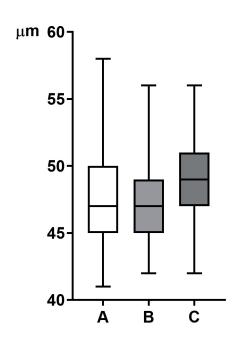


**Graph 3.** Minimum corneal epithelial thickness. Results are presented as median (interquartile range). Asterisk – differences between groups were statistically significant

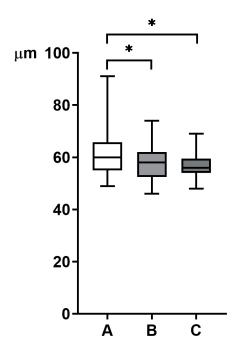
A – keratoconus, B – latent keratoconus, C – astigmatism, \* – p < 0,001

#### **DISCUSSION**

AS OCT enables visualization of the cornea and its pathological states, including keratoconus. Since this bilateral ectatic corneal pathology usually afflicts young patients and poses a considerable risk of a seve-



**Graph 2.** Corneal epithelial thickness in the 2–5mm paracentral zone of the cornea. Results are presented as median (interquartile range) A – keratoconus, B – latent keratoconus, C – astigmatism



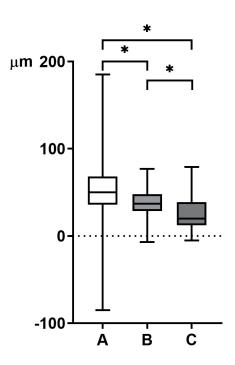
**Graph 4.** Maximum corneal epithelial thickness. Results are presented as median (interquartile range). Asterisk – differences between groups were statistically significant

 $A-keratoconus, B-latent\ keratoconus, C-astigmatism,\ ^*-p<0,001$ 

re and irreversible impact on the patient's visual acuity, the need for diagnostic options in its initial stages is highly imperative.

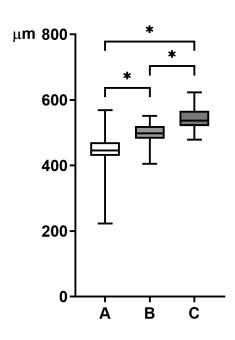
In this study, using AS OCT pachymetry we determined statistically significant differences in the majority of the analyzed parameters of corneal epithelial

thickness (thickness in the 2 mm central zone, minimal thickness, maximal thickness). This concerned differences between the analyzed groups of eyes with keratoconus and latent keratoconus, as well as between keratoconus and astigmatism. However, significant differences in corneal epithelial thickness were not



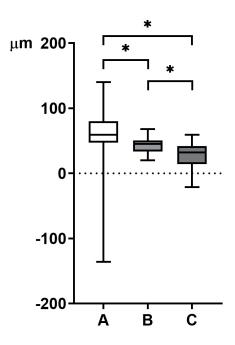
**Graph 5.** Difference in total corneal thickness in the superior (S) and inferior (I) segments in the 2 – 5mm paracentral zone. Results are presented as median (interquartile range). Asterisk – differences between groups were statistically significant

A-keratoconus, B-latent keratoconus, C-astigmatism, \*-p < 0.001



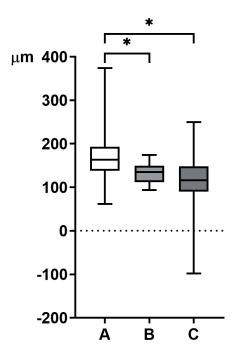
**Graph 7.** Minimum total corneal thickness. Results are presented as median (interquartile range). Asterisk – differences between groups were statistically significant

A – keratoconus, B – latent keratoconus, C – astigmatism, \* – p < 0,001



**Graph 6.** Difference in total corneal thickness in the superior nasal (SN) and inferior temporal (IT) segments in the 2–5mm paracentral zone. Results are presented as median (interquartile range); asterisk = differences between groups were statistically significant

A – keratoconus, B – latent keratoconus, C – astigmatism, \* – p < 0.001



**Graph 8.** Difference between the maximum and minimum total corneal thickness. Results are presented as median (interquartile range). Asterisk – differences between groups were statistically significant A - keratoconus, B - latent keratoconus, C - astigmatism, \*- p < 0,001

confirmed between latent keratoconus and astigmatism. We therefore did not demonstrate the capability of AS OCT to differentiate between the latent form of the disease and astigmatism on the basis of the parameters of corneal epithelial thickness. In this point AS OCT does not appear to be a beneficial imaging method in potentially detecting the latent pathology, merely a method capable of differentiating clinically manifest keratoconus from its latent form and from corneal astigmatism without ectatic changes. In the parameters of total corneal thickness, AS OCT demonstrated significant differences in all 4 analyzed parameters (S-I, SN-IT, min, max-min). This concerned differences between the analyzed groups with keratoconus and latent keratoconus, between keratoconus and astigmatism, but also between latent keratoconus and astigmatism. In this point AS OCT is capable of registering parameters differentiating clinically manifest keratoconus from the latent form of the disease, and is also capable of differentiating latent keratoconus from corneal astigmatism without ectatic changes. Although medium-severe and advanced forms of ectatic corneal pathology can be diagnosed relatively easily and reliably by means of a clinical examination of the patient and corneal topography, it may not always

be possible to differentiate the initial stages or latent form of keratoconus from a normal (healthy) cornea. In such a case, AS OCT represents a supplementary examination which is capable of detecting keratoconus before its clinical manifestation, which we consider to be the main benefit of using this examination method in clinical practice [9].

Elhennawi et al. used AS OCT for diagnosing keratoconus in patients before refractive surgery of the cornea. They analyzed a cohort of 40 eyes with myopic astigmatism, focusing on the 5 mm central zone of the cornea. In accordance with the results of our study, they determined that the values of minimal corneal thickness, the difference between corneal thickness in the inferior quadrant (I-S) and between thickness in the inferior temporal and superior nasal quadrant (IT-SN) may be good indicators in the diagnosis of keratoconus [10].

Ostadian et al. evaluated a cohort of 63 eyes before refractive surgery of the cornea, in which 24 eyes had a normal topographic finding with myopia of less than -6 D and astigmatism of less than -4 D, 17 had a topographic finding corresponding to latent keratoconus and 22 eyes had early form of manifest keratoconus. By contrast with our study, they used AS OCT only to evaluate corneal epithelial thickness and not the parameter

Tabulka 2. Statistical analysis – epithelial thickness

	Epithelial thickness in central 2 mm zone (μm)		Epithelial thickness in paracentral 2–5 mm zone (μm)			Minimum epithelial thickness (μm)			Maximum epithelial thickness (μm)			
	Average	SD (+/-)	Median	Average	SD (+/-)	Median	Average	SD (+/-)	Median	Average	SD (+/-)	Median
Α	45.2	5.23	45	47.	3.48	47	32.9	4.28	33	61.8	8.63	60
В	48.5	4.52	48	47.7	3.48	47	36	4.17	35	57.5	6.1	58
C	50.3	3.25	50.5	48.9	3.28	49	35.3	3.5	35	56.8	4.53	56
A vs. B	p < 0.001			-			p < 0.001			p < 0.001		
B vs. C	p = 0.143			-			p = 0.576			p = 0.714		
A vs. C	p < 0.001			-			p < 0.001			p < 0.001		

A – keratoconus, B – latent keratoconus, C – astigmatism, SD – standard deviation

**Tabulka 3.** Statistical analysis – total corneal thickness

	Difference in total corneal thickness between superior (S) and inferior (I) segment in 2–5 mm zone (µm)		superopasal (SN) and			Minimum total corneal thickness (μm)			Difference between maximum and minimum total corneal thickness (µm)			
	Average	SD (+/-)	Median	Average	SD (+/-)	Median	Average	SD (+/-)	Median	Average	SD (+/-)	Median
Α	56.4	17.92	55	60.6	34.62	59	443.7	53.74	446	169.7	50.04	163
В	38.4	17.67	37	43.5	12.66	45	497.3	30.50	498	132.3	21.78	135
C	25	18.92	20	30.2	17.05	32	543.4	33.81	536,5	115.6	54.73	116.5
A vs. B	p < 0.001		p < 0.001			p < 0.001			p < 0.001			
B vs. C	p = 0.003			p < 0.001			p < 0.001			p = 0.057		
A vs. C	p < 0.001			p < 0.001			p < 0.001			p < 0.001		

A – keratoconus, B – latent keratoconus, C – astigmatism, SD – standard deviation

of total corneal thickness. In their study they evaluated epithelial thickness on the basis of different selected parameters ("uniformity index" of the epithelium of the superior and nasal segment, difference between epithelial thickness in the superior nasal and inferior temporal segment – SN-IT, difference between epithelial thickness in the temporal and nasal segment – T-N) than those evaluated in our study. They determined that in cases of latent keratoconus the values were increased in the inferior and temporal quadrant. In addition to this, in the group of eyes with latent keratoconus they confirmed a significantly smaller difference in epithelial thickness between the superior and inferior, as well as between the superior nasal and inferior temporal quadrant (S-I and SN-IT) in comparison with the group of eyes with manifest keratoconus. In the analyzed group of eyes with manifest keratoconus, the area with minimal corneal epithelial thickness was significantly smaller in comparison with the groups with a normal and latent topographical finding. Epithelial thickness increases in order to cover the area of thinning of the stroma, and as a result indexes such as S-I and SN-IT, which reflect the asymmetry of the corneal epithelium as well as increased epithelial thickness in the inferior part of the cornea, may be of significance in helping diagnose the latent form or early stages of manifest keratoconus [11].

Li et al. in their study compared the results of an analysis of corneal epithelial thickness with the aid of AS OCT in a cohort of 145 normal eyes and 35 eyes with keratoconus. Unlike our cohort of analyzed eyes, they did not include a group with the latent form of the disease, and this study also did not deal with the parameters of total corneal thickness. They did not determine any significant difference between the values of epithelial thickness in the central and superior part of the cornea, but epithelial thickness in the inferior part of the cornea was significantly lower in eyes with keratoconus in comparison with the group of normal eyes. Their results partially concur with ours, since similarly to in our study, minimal corneal epithelial thickness, the asymmetry index S-I and the difference between minimal and maximal corneal epithelial thickness (min-max) were significantly lower in eyes with keratoconus. Mean corneal epithelial thickness was lower in the inferior temporal quadrant and higher in the superior nasal quadrant in the group of eyes with keratoconus in comparison with normal eyes [3].

Qin et al. compared a cohort of 133 normal eyes and 84 eyes with keratoconus with the aid of AS OCT pachymetry maps, focusing only on the parameter of total corneal thickness. In contrast with our study, this publication also did not deal with a group of eyes with the latent form of the pathology. With regard to the study methodology, they evaluated parameters partially concordant with the parameters selected for analysis in our study: minimal corneal thickness, difference between minimal and medium corneal thickness (calculated from values in the 5 mm central zone of the

cornea), difference of corneal thickness in the superior and inferior octant (S-I), difference of corneal thickness in the superior nasal and inferior temporal octant (SN-IT) and vertical position of the thinnest point of the cornea ( $Y_{\rm min}$ ). In accordance with the results of our study, they determined statistically significant differences between the group of eyes with keratoconus and the group of eyes with astigmatism in the parameters of S-I, SN-IT and minimal total corneal thickness (p < 0.001). They also deduced a formula according to which it is possible to use the obtained variables in order to determine a reliable diagnosis of keratoconus: 0.543 × min + 0.541 × (S-I) – 0.886 × (SN-IT) + 0.886 × (min-med) + 0.0198 ×  $Y_{\rm min}$  [12].

A study conducted by Sella et al. evaluated the reproducibility and repeatability of pachymetry examination of the cornea and corneal epithelium with the aid of AS OCT in a cohort of 12 normal eyes and 48 eyes with a variable finding influencing the state of the cornea (dry eye syndrome, long-term use of soft or hard contact lenses for correcting refractive error, and keratoconus). Similarly, to in our study, corneal pachymetry maps in this study also were divided into the 2 mm central zone and the 2–5 mm paracentral zone, and in addition the 5-6 mm peripheral zone. The paracentral and peripheral zones were analyzed in 8 sectors (temporal, superior temporal, superior, superior nasal, nasal, inferior nasal, inferior and inferior temporal), which led to a total evaluation of 17 parameters of the map. However, the subgroup of eyes with keratoconus in this study included only 12 eyes, including one eye after perforating keratoplasty and one eye after implantation of intrastromal corneal segments; none of the eyes had undergone CXL. The study evaluated AS OCT as a universally reliable method of corneal examination in patients with keratoconus, and the results of total pachymetry and epithelial pachymetry as repeatable and reproducible for the entire spectrum of severity of the pathology. As in our study, in this publication also a statistically significant difference was determined in minimal and maximal epithelial thickness between the group of healthy eyes and the group of eyes with keratoconus (p < 0.001). In the conclusion, the study noted that in eyes with keratoconus there was a greater requirement for manual correction of the image (17.9 %), since automatic segmentation by the instrument may be imprecise in the case of irregular corneas. Nevertheless, repeatability and reliability remained high [13].

#### **CONCLUSION**

Pachymetry maps are a demonstrably reliable method for identifying differences between keratoconus and latent keratoconus, between keratoconus and astigmatism, but also – and within this context this is the most important factor – between latent keratoconus and corneal astigmatism. The results of the study indicate that