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Pachymetric measurements using 3 different methods for cross linked keratoconic corneas

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Abstract

Purpose: To compare corneal thickness using US pachymeter, Scirus, and OCT, evaluate agreement between them as well as determine the variation of each machine in pre- and post- cross-linking of keratoconic eyes.

Patients and methods: In total, 50 eyes of 25 keratoconic patients before and after cross linking treatment were analyzed in this study. Pachymetry measurements were performed at center, inferior and superior temporal and nasal locations using US pachymeter, OCT and Scirus. One way ANOVA, intra-class correlation coefficient (ICC), coefficient of variation (COV) and Bland and Altman plots were used for statistical analysis.

Results: The mean corneal thicknesses measured at center, ST, and IT before cross-linking and at center, ST, SN and IN after cross-linking was statistically different between pachymeters. The highest correlation coefficient was found to be between US and OCT at IT (0.868) before cross-linking and between US and OCT at center (0.826) and ST (0.801) after cross-linking. Poorest agreement prior to cross-linking was between Scirus and OCT at SN (0.211), and after cross-linking between US and Scirus at IT (0.003), between US and OCT (0.099) as well as and between US and Scirus (ICC = 0.098) at IT. Variation was negligible for OCT and maximal for US pachymetry before and after cross-linking.

Conclusion: OCT displayed high agreement with ultrasound pachymeter and least variations in readings before and after cross-linking of keratoconous eyes. Hence it is recommended to use only OCT for pre-surgical evaluation and follow-up. However, replication of the results in larger sample size is needed.

Keywords: Pachymetry; Keratoconous; OCT; Scirus US pachymeter; Corneal thickness; cross linking;

Introduction

Keratoconus is a non-inflammatory corneal disease which is symptomized by ectasia of the central, paracentral, or mid peripheral region of the cornea causing thinning, scarring and unevenness in the surface of cornea [1,2]. It normally affects both the eyes. Keratoconus is the most common corneal dystrophy noted in people worldwide affecting about 50 to 230 per 100000 people [1]. In patients seeking kerato-refractive surgery measurement of corneal thickness before surgery is necessary for determining the disease progression and appropriate treatment regime as sequential measurements of corneal thickness helps in differentiating keratoconus from corneal thinning due to contact lens wear [3,4]. Postoperative measurement of corneal thickness is required for outcome measurements and follow-up [5]. Besides, corneal thickness is needed for determining the intraocular pressure accurately [6].

Numerous devices have been developed to measure the corneal thickness which includes ultrasound pachymetry, optical pachymetry, contact and noncontact specular microscope, scanning slit topography/pachymetry, corneal confocal microscopy, ultrasound biomicroscopy, partial coherence interferometry, and optical coherence tomography [7,8,9]. However, the ideal device should be able to measure the corneal thickness accurately and safely as the outcome and success rate of corneal cross-linking depends on the accuracy and precision of pachymetry measurements [10]. Ultrasound pachymetry is considered the gold standard for determining corneal thickness due to high degree of inter-observer and inter-instrument reproducibility [11]. Nonetheless, this device uses contact method for measuring corneal thickness which requires application of topical anesthesia and is unsafe due to many reasons such as direct contact of the probe with cornea increases the risk of tear film, incorrect and unrepeatable probe placement, and differences in pressure applied during measurement [9,12,13]. Additionally, with each contact of the probe, the machine can determine the corneal thickness at one single point only. Therefore, non-contact methods of measuring corneal thickness have gained popularity.

The Scirus uses a rotating Scheimpflug camera system and provides 3-dimensional scanning of the whole anterior segment of the eye, information regarding corneal pachymetry, anterior and posterior corneal topography, anterior chamber depth, volume, and angle and lens density [14]. The noncontact measuring process with the Scirus system takes 2s, performing 12– 50 single captures while rotating around the optical axis of the eye. As every slit image consists of 500 true elevation points, the Scirus system detect, in total, up to 2500 height values, which are processed to a 3-dimensional model of the entire anterior eye segment [15]. The slit-lamp OCT (SL-OCT) combines slit-lamp biomicroscopy and imag¬ing technology of OCT and it can also be used to measure the corneal thickness [8].

Pachymeters are highly expensive instruments. It incurs heavy expenses on the hospitals to keep more than one pachymeters for patient examination. Further, high variability in the outcome is expected when corneal thickness is measured with one pachymeter before cross-linking and with another after cross-linking. To address the issues related to variability in outcome most of the studies so far have assessed the agreement of non-contact pachymeters with ultrasound pachymeter (the gold standard) in pre- and post-treated keratoconic eyes [7,15,16]. However, along with above mentioned evaluation, assessing the agreement between pre- and post- cross-linking corneal thickness measurements by each non contact pachymeter is necessary as it gives the most accurate outcome of the treatment during follow-up, beside reducing the hospital expenses.

In this study, we evaluated the agreement between contact (ultrasound pachymeter) and non contact instruments (Sirius and OCT) in measuring corneal thickness at different locations in Keratoconous patients before and after corneal cross-linking and also evaluated the variability in readings by three devices before and after the treatment.

Materials and methods

This prospective study was conducted at Dar El-Oyoun Eye Laser Center, Oman to compare corneal thickness at different places of cornea by Scirus, (CSO), OCT (Optos) and, US pachymeter (Nidek) pre- and post corneal cross-linking. Written informed consent was obtained from all the subjects after the protocol and possible risks were explained; the consent form was approved by the Institutional Review Board and carried out according to the Declaration of Helsinki.

Patient Selection

In this study 25 patients (50 eyes) with Keraoconous were included who were suitable for the cross linking treatment. The diagnosis of keratoconus was made by analyzing the degree of distortion of the retinoscopic red reflex (scissoring reflex) and Sirius corneal maps.

Inclusion criteria: All keratoconus patients with corneal thickness more than 400 micron.

Exclusion criteria: The patients with corneal thickness less than 400 micron were excluded. Moreover, the patients with Vogt's striae, scaring, previous hydrops, refractive excimer cornea surgery, and corneal ring or previous cross linking treatment were also excluded.

Measurements of Corneal Thickness

Corneal thickness was measured by US pachymetry, anterior segment Optical coherence tomography (SL- OCT) and Corneal Topographer (Scirus) according to manufacturer's guidelines. To avoid the influence of probe application on corneal thickness measurements by other modalities, US pachymeter measurements were recorded in the last. For measurements with US pachymeter, eyes were anesthetized topically with 0.5% proparacaine/0.5% tetracaine after which the ultrasound probe was gently applied to the cornea center perpendicularly.

Corneal thickness was noted at five different locations: center, superior temporal (ST), inferior temporal (IT), superior nasal (SN), and inferior nasal (IN) repeated three times and average of the three reading was recorded. The corneal thickness at the geometric center was used as the reference for defining the rest of the locations through the x-y Cartesian coordinate grid embedded in the software of each instrument. Subjects were asked to blink before every scanning for an optically smooth tear film over the cornea, head was positioned in rest, and they were asked to fixate on the target on the center of the camera without blinking during the scan.

All images and pachymetric maps were analyzed after recording the measurements with the respective software of the respective machines. All the above measurements were repeated after six month postoperatively.

Statistical analysis

The Statistical Package for the Social Sciences Statistical Software, version 21 (SPSS Inc., Chicago, IL, USA), was used for the statistical analysis. Data was expressed as means ± standard deviations and 95% CI levels. One-way ANOVA followed by Tukey's post-hoc test was used to compare the means of the same measurements made by different instruments. The intra-class correlation coefficient (ICC) was calculated for each dataset to assess the reliability (ie degree of relatedness) between same measurements made by different instruments; a two-way mixed model for absolute agreement was used with statistical significance set at 95%. Bland and Altman analysis was used to demonstrate the agreement between measurements made with different tools and present them as 95% limits of agreement (LoA). Coefficient of variation was calculated for each instrument before and after treatment Levene's test of homogeneity of variance was used to analyze inter-modality reproducibility in corneal measurements. $P < 0.05$ was considered to be statistically significant.

Results

Comparison of corneal thickness at different locations

The mean values of corneal thickness at different locations (central, ST, IT, SN, and IN) obtained by the ultrasound pachymeter, Scirus, and OCT are shown in (Table 1). The differences between the devices are statistically significant for corneal thickness measured at center, ST, and IT before cross-linking ($p < 0.001$). Post cross-linking, the readings obtained for central, ST, SN and IN corneal thickness were statistically different between instruments ($p < 0.001$). Post-hoc analysis showed that considerable differences existed between different instruments for mean corneal thickness measured at different locations (Table 2).

Correlations between pachymetry methods

The correlation coefficients showing agreements between the corneal thickness measurements obtained with ultrasound pachymetry, Scirus, and the OCT system in the present study are

Table 1: Comparative analysis of US pachymeter, Scirus and OCT measurements in pre- and post cross-linked corneas

Measurement localization	Pre cross-linking (Mean ± SD µm)	P value	Post cross-linking (Mean ± SD µm)	P value
Central				
US Pachymeter	480.28 ± 17.25	0.003	424.42 ± 11.61	0.000
Scirus	468.28 ± 18.85		406.32 ± 18.18	
OCT	471.04 ± 18.03		421.70 ± 13.60	
ST				
US Pachymeter	511.38 ± 19.29	0.000	451.96 ± 14.77	0.000
Scirus	515.32 ± 29.44		432.14 ± 19.89	
OCT	496.82 ± 20.70		448.26 ± 16.87	
IT				
US Pachymeter	508.76 ± 17.38	0.002	444.42 ± 58.98	0.260
Scirus	522.76 ± 27.89		433.80 ± 17.87	
OCT	509.96 ± 16.59		444.04 ± 14.42	
SN				
US Pachymeter	503.50 ± 17.83	0.578	447.16 ± 13.71	0.000
Scirus	508.48 ± 24.18		429.88 ± 20.01	
OCT	501.96 ± 47.53		442.26 ± 15.33	
IN				
US Pachymeter	509.46 ± 17.63	0.273	454.64 ± 10.27	0.000
Scirus	503.28 ± 20.75		434.02 ± 19.51	
OCT	505.52 ± 19.50		449.34 ± 13.97	

Table 2: Post-hoc analysis of US Pachymeter, Scirus, and OCT for different measurements in pre- and post cross-linked corneas

Measurement localization	Pre cross-linking Mean diff.	P value	Post cross-linking Mean diff.	P value
Central				
US Pachymetervs. Scirus	12.0	0.003	18.1	0.000
Pachymeter vs. OCT	9.24	0.031	2.72	0.626
Scirusvs.OCT	-2.76	0.726	-15.38	0.000
ST				
US Pachymetervs. Scirus	-3.94	0.682	19.82	0.000
Pachymeter vs. OCT	14.56	0.007	3.7	0.535
Scirusvs.OCT	18.50	0.000	-16.12	0.000
IT				
US Pachymetervs. Scirus	-14.0	0.004	10.62	0.317
Pachymeter vs. OCT	-1.2	0.957	0.38	0.999
Scirusvs.OCT	12.8	0.009	-10.24	0.343
SN				
US Pachymetervs. Scirus	-4.98	0.724	17.28	0.000
Pachymeter vs. OCT	1.54	0.969	4.9	0.304
Scirusvs.OCT	6.52	0.575	-12.38	0.001
IN				
US Pachymetervs. Scirus	6.18	0.250	20.62	0.000
Pachymeter vs. OCT	3.94	0.566	5.3	0.187
Scirusvs.OCT	-2.24	0.832	-15.32	0.000

shown in (Table 3). The range of differences in the pachymetric measurement between instruments was not constant throughout cornea. There was a larger range of difference between devices in before and after treatment at different locations of cornea. The highest correlation coefficient was found to be between ultrasound and OCT in IT measurements (ICC = 0.868, *P*< 0.001) before treatment and between ultrasound and OCT in central (ICC = 0.826, *P* < 0.001) as well as ST (ICC = 0.801, *P* < 0.001) measurements after treatment. Poorest agreement prior to cross-linking was between Scirus and OCT in SN (ICC= 0.211 (*P* > 0.05), and after cross-linking between Ultrasound and Scirus in IT (ICC=0.003, *P* > 0.05), between Ultrasound and OCT in IT (ICC = 0.099, *P*> 0.05), and between Ultrasound and Scirus (ICC = 0.098, *P* > 0.05).

Table 3: Degree of correlation between measurements of two devices at different locations in pre- and post cross-linked corneas

Measurement localization	Pre cross-linking		Post cross-linking	
	ICC (r)	95% CI	ICC (r)	95% CI
Central				
US Pachymetervs. Scirus	0.683***	0.025 – 0.879	0.291***	-0.086 – 0.588
US Pachymeter vs. OCT	0.786***	0.128 – 0.925	0.826***	0.694 – 0.901
Scirusvs.OCT	0.771***	0.629 – 0.823	0.331***	-0.040 – 0.604
ST				
US Pachymetervs. Scirus	0.447***	0.197 – 0.643	0.347***	-0.087 – 0.650
US Pachymeter vs. OCT	0.513***	0.062 – 0.751	0.801***	0.653 – 0.886
Scirusvs.OCT	0.555***	0.049 – 0.788	0.334***	-0.013 – 0.593
IT				
US Pachymetervs. Scirus	0.490***	0.153 – 0.705	0.003	-0.270 – 0.277
US Pachymeter vs. OCT	0.868***	0.779 – 0.923	0.099	-0.188 – 0.368
Scirus vs.OCT	0.510***	0.190 – 0.714	0.355**	0.069 – 0.582
SN				
US Pachymetervs. Scirus	0.676***	0.489 – 0.803	0.226**	-0.054 – 0.478
US Pachymeter vs. OCT	0.326*	0.051 – 0.554	0.644***	0.421 – 0.788
Scirus vs.OCT	0.211	-0.068 – 0.460	0.276**	0.008 – 0.510
IN				
US Pachymeter vs. Scirus	0.795***	0.578 – 0.894	0.098	-0.077 – 0.303
Pachymeter vs. OCT	0.573***	0.355 – 0.732	0.693***	0.390 – 0.839
Scirus vs.OCT	0.612***	0.406 – 0.760	0.332***	-0.022 – 0.596

To demonstrate the differences between devices graphically, Bland-Altman plots of the differences in central, ST, IT SN, and IN corneal thickness between the ultrasound versus Scirus, Ultrasound versus OCT and Scirus versus OCT in pre and post treated keratoconous corneas were plotted against the mean value of both which are shown in (Figures 1-5).

Intra-device and inter- device variations in pre- and post- treated corneal thickness

Coefficient of variation for each instrument before and after corneal cross-linking is shown in (Table 4). Complete agreement between pre- and post treated corneal thickness at five different locations was seen in OCT. Significant differences between corneal thickness at inferior temporal region only was seen in Scirus in between before and after cross-linking (p < 0.043). Ultrasound pachymetry showed maximum variations between pre- and post treated corneas at central, ST and IN locations.

(Table 5) shows the homogeneity in variances within different modalities for corneal measurements. At central location before surgery, all the three instruments had good agreement in the measurements (p = 0.872) which was absent post surgery (p < 0.001). Similarly, at SN and IN also, the measurements between instruments are in accord before surgery which is not present after surgery. All the three instruments were in agreement for corneal measurement after surgery at IT position. At ST position, all the three instruments differed considerably in corneal measurements in both time points (pre-and post-surgery).

Discussion

Accurate evaluation of corneal thickness is important to minimize the risk of post corneal cross-linking complications such as keratectasia [15]. Nonetheless, in real sense, the actual value of corneal thickness of eye is difficult to determine. Therefore, maintaining consistency with negligible variations in measurements before and after treatment is the prime

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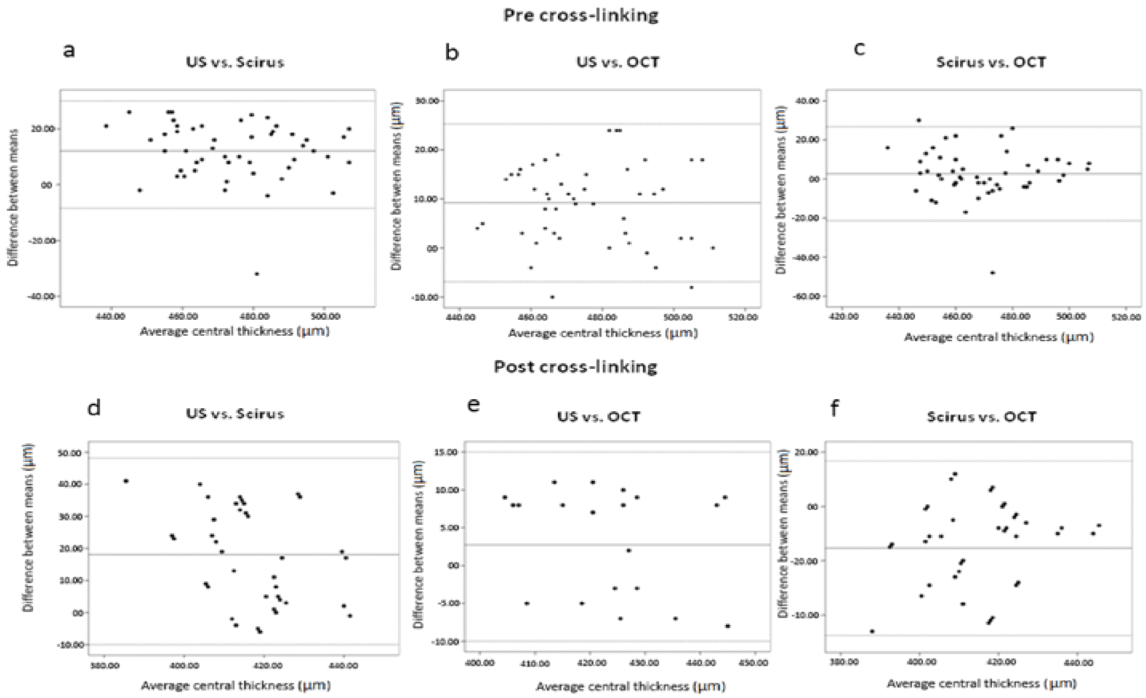

Figure 1: Bland–Altman plot with 95% limits of agreement (dotted lines) illustrates the difference in central corneal thickness measurements (y-axis) between values obtained by different instruments against the average central corneal thickness measurements of the two methods (x-axis) before and after cross-linking treatment.

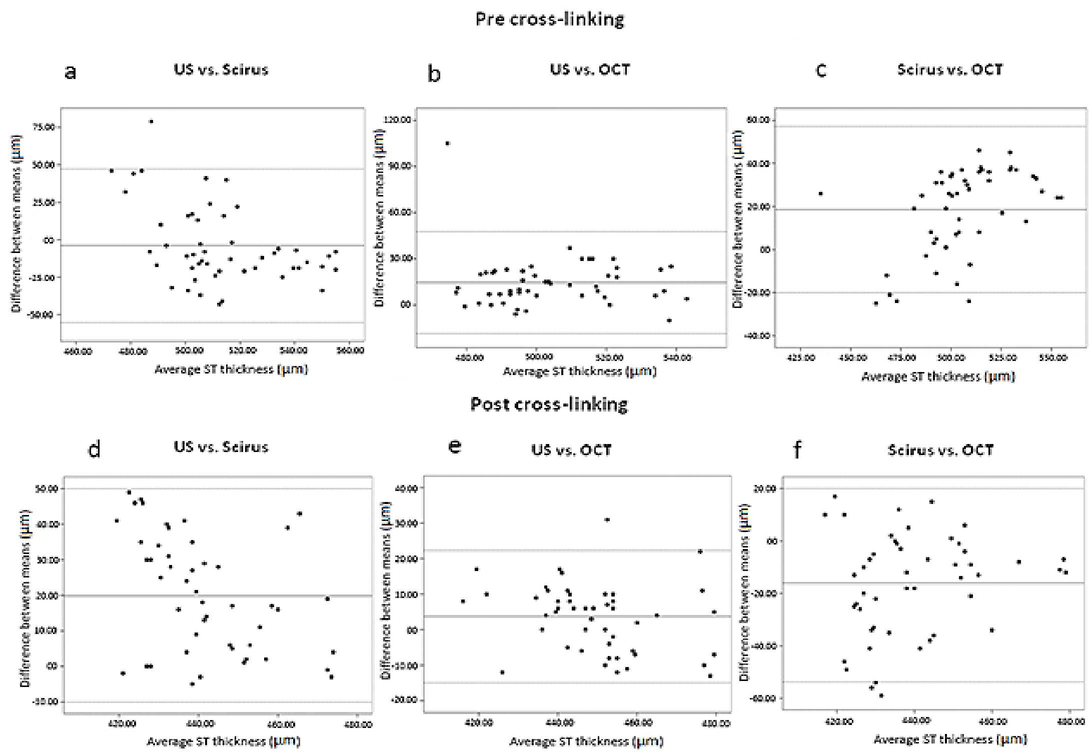

Figure 2: Bland–Altman plot with 95% limits of agreement (dotted lines) illustrates the difference in superior temporal (ST) corneal thickness measurements (y-axis) between values obtained by different instruments against the average corneal thickness measurements at ST of the two methods (x-axis) before and after cross-linking treatment.

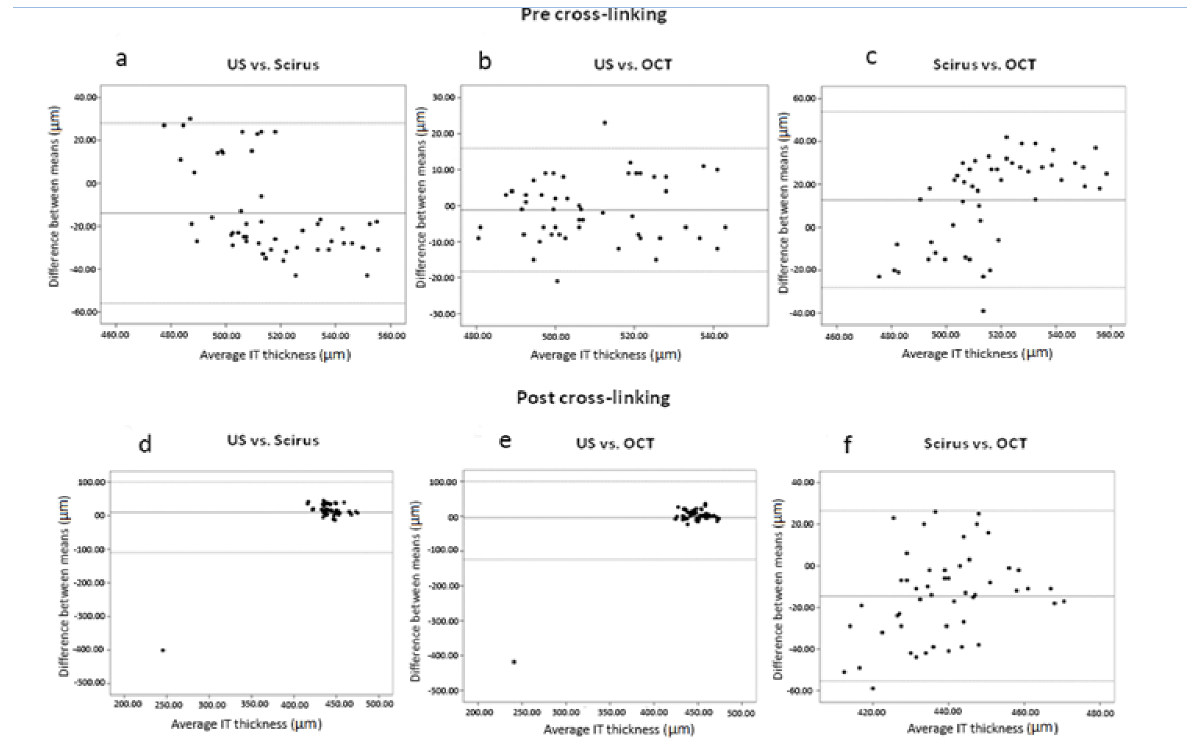

Figure 3: Bland–Altman plot with 95% limits of agreement (dotted lines) illustrates the difference in inferior temporal (IT) corneal thickness measurements (y-axis) between values obtained by different instruments against the average corneal thickness measurements at IT of the two methods (x-axis) before and after cross-linking treatment.

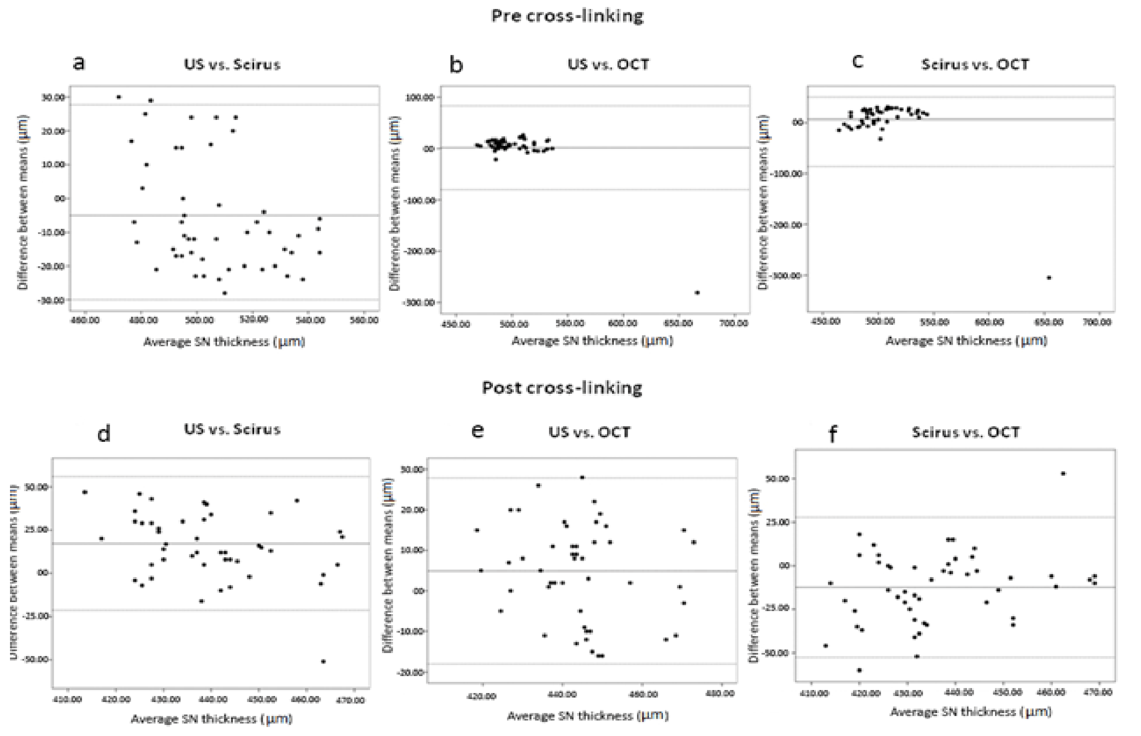

Figure 4: Bland–Altman plot with 95% limits of agreement (dotted lines) illustrates the difference in superior nasal (SN) corneal thickness measurements (y-axis) between values obtained by different instruments against the average corneal thickness measurements at SN of the two methods (x-axis) before and after cross-linking treatment.

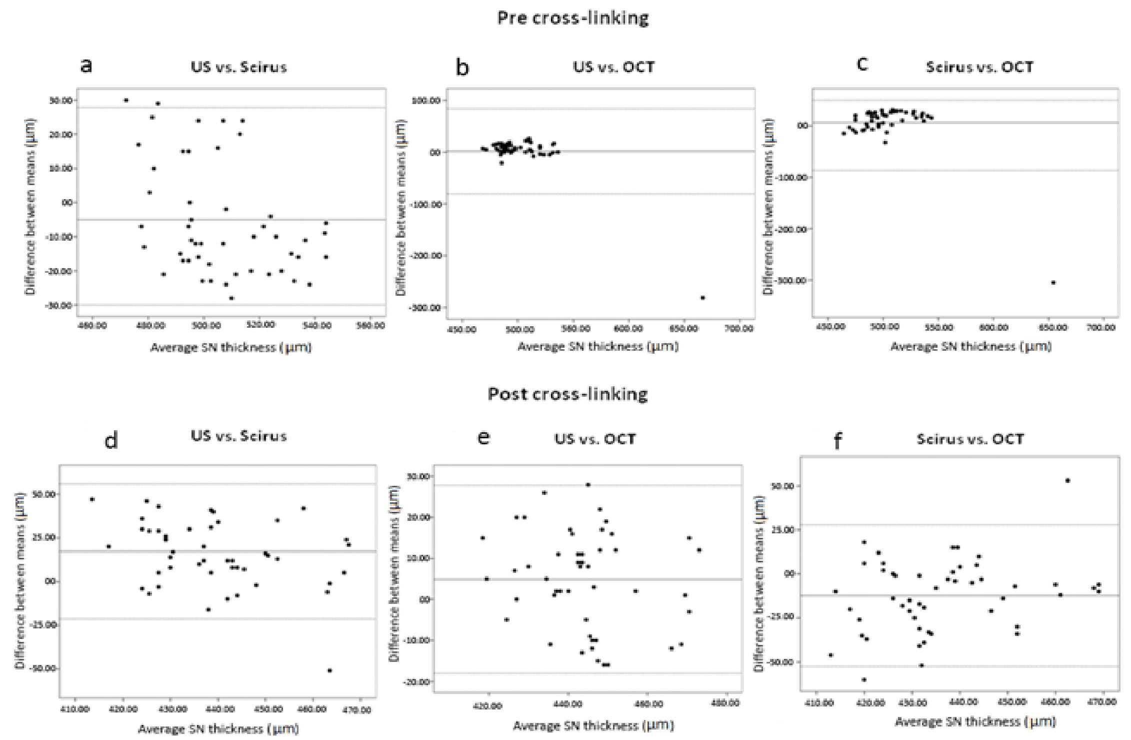

Figure 5: Bland–Altman plot with 95% limits of agreement (dotted lines) illustrates the difference in inferior nasal (IN) corneal thickness measurements (y-axis) between values obtained by different instruments against the average corneal thickness measurements at IN of the two methods (x-axis) before and after cross-linking treatment.

Table 4: Coefficient of variation of different pachymeters before and after cross-linking

Measurement localizations	Pre cross-linking (mean ± SD)	Post cross-linking (mean ± SD)	F- value	p-value
		Central		
US Pachymeter	0.030 ± 0.018	0.021 ± 0.016	6.322	0.014
Scirus	0.033 ± 0.022	0.036 ± 0.024	0.566	0.454
OCT	0.031 ± 0.021	0.025 ± 0.019	2.094	0.151
		ST		
US Pachymeter	0.032 ± 0.018	0.023 ± 0.022	4.464	0.037
Scirus	0.044 ± 0.035	0.037 ± 0.025	1.216	0.273
OCT	0.030 ± 0.028	0.029 ± 0.023	0.033	0.856
		IT		
US Pachymeter	0.029 ± 0.017	0.044 ± 0.124	0.748	0.389
Scirus	0.044 ± 0.029	0.033 ± 0.024	4.208	0.043
OCT	0.026 ± 0.018	0.026 ± 0.018	0.000	0.987
		SN		
US Pachymeter	0.030 ± 0.017	0.023 ± 0.019	3.134	0.080
Scirus	0.039 ± 0.026	0.036 ± 0.028	0.215	0.644
OCT	0.044 ± 0.083	0.027 ± 0.021	2.006	0.160
		IN		
US Pachymeter	0.030 ± 0.018	0.021 ± 0.016	16.735	0.000
Scirus	0.030 ± 0.018	0.021 ± 0.016	0.252	0.616
OCT	0.030 ± 0.018	0.021 ± 0.016	1.396	0.240

Table 5: Inter-modality repeatability of corneal measurements

Corneal locations	Levene statistic	P value
Pre - Central	0.137	0.872
Post - Central	6.190	0.003
Pre - ST	4.260	0.016
Post - ST	3.623	0.029
Pre - IT	9.883	0.000
Post - IT	0.746	0.476
Pre - SN	0.965	0.383
Post - SN	3.392	0.036
Pre - IN	0.519	0.596
Post - IN	10.273	0.000

requirement from any pachymeters. Previous studies have compared agreement of non contact pachymeters such as Scirus, OCT, OrbscanII, etc, with US pachymeter (the gold standard) in terms of correlation and variations in measurement of corneal thickness between devices in normal, keratoconous suspected and in post LASIK treated corneas [6,7,11,12,15]. In the current study, along with evaluating agreement between contact and non contact pachymeters in pre and post cross-linked corneas at different locations, we also examined the agreement between pre and post treatment measurements by each individual device. This research enabled us to pinpoint one particular device for pre treatment assessment as well as post treatment follow-up in kertoconous eyes as pachymeters are highly expensive machines.

At five different locations where corneal thickness was measured, OCT showed maximum agreement with US pachymeter before and after cross-linking. Although corneal thickness measured centrally (mean difference, 9.24) and supratemporally (mean difference, 14.56) by OCT was lower in keratoconic eyes earlier to cross-linking when compared to US ($p < 0.05$), they were in complete agreement after the treatment. In accordance with our results, previous researches have also shown that OCT underestimated the central corneal thickness compared to US [15,16,17]. The maximum difference between US and OCT was 5.3 which are lower than what other studies have reported. With OCT, Ho et al [15] found the mean corneal thickness to be 11.64 ± 12.87 μm lower than with US pachymetry in post-LASIK eyes ($P < 0.01$). Ponce and colleagues found a decrease of 7.5 ± 1.4 μm in mean in pachymetry measurements with OCT which was comparable with keratoconus-suspect or post-LASIK eyes ($P > 0.05$). [16] Contrary to our other studies, Li et al [18], have noted higher mean value of corneal thickness for OCT than US. Higher US pachymetry values are attributed to corneal edema produced by anesthetic drops. [18] OCT pachymetry values tend to be accurate due to improved centration and perpendicularity. [18,19] On the other hand, tear-film displacement caused by US pachymetry may lead to lower pachymetry values than those obtained by OCT. [15,18,19] Research by other two groups, Li et al [20] and Beutelspacher et al [8], showed no significant dif¬ference between these two modalities. This high correlation of measurement values between the two modalities may be due to the similar mechanism of the two instruments – both use reflection of waves to determine the anterior and pos¬terior boundaries of the cornea. [21] Moreover, many studies consider OCT to be superior over other tomographic devices, such as Orbscan II (Bausch & Lomb), because it obtains corneal-thickness profiles in less time and is not influenced by stromal reflections or haze. [15,18,19] In addition to diagnosis, OCT systems can screen and monitor the progression of the keratoconus [22].

In this study, mean corneal thickness obtained by Scirus machine was variable at different locations in pre-surgical keratoconic eyes while they were remarkably less than both US and OCT in post surgical eyes. Our results were in agreement with the results of Ho et al [15], Ponce et al [16] and with the recent meta-analysis. All the three studies showed that Scirus considerably underestimated the corneal thickness at center in post-LASIK eyes compared to OCT and US [15,17,23]. Even in normal eyes, Lackner et al [24], showed Scirus readings to be slightly smaller (9.8 mm) than US. Contrarily, Ackay et al [22], reported that Scirus measurements were higher than OCT at different locations except at center in keratoconous eyes. In normal eyes, good correlation between US and Scirus readings were noted. [25] In our study although correlation between US and Scirus was good before cross-linking, it reduced greatly in post treated eyes and became non-significant at IT and IN locations.

Although few studies have analyzed the inter and intra operator variability of the pachymeters, none so far have focused on variations between measurements taken by same machine before and after surgery. [26,27] in the present study, variation in OCT measurements before and after cross-linking at different locations was comparable. Scirus significantly differed in pre and post readings at IT. Interestingly, US considerably varied in readings at three different locations before and after cross-linking. This poses a question on using of US pachymeter for follow-up after corneal cross-linking.

The two major limitations of the study include lack of the intra- and inter observer reproducibility of the measurements obtained with each instrument and small sample size.

Conclusion

In conclusion, OCT readings were in high agreement with US and were least variable in pre and post cross-linked keratoconic eyes. Hence, OCT should be recommended for diagnosis and pre cross-linking assessment of keratoconous eye as well as for follow-up. This will lead to reduction in post cross-linking complications as well as decrease in expenses of having and maintaining more than one pachymeters. Nonetheless, replication of the results of the present study in large sample size is warranted.

Disclosure

The author reports no conflicts of interest in this work.

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