Reproducibility and Repeatability of Central Corneal Thickness Measurement in Keratoconus Using the Rotating Scheimpflug Camera and Ultrasound Pachymetry

UGO DE SANCTIS, ALESSANDRO MISSOLUNGI, BERNARDO MUTANI, LORENZO RICHIARDI, AND FEDERICO M. GRIGNOLO

• PURPOSE: To assess repeatability, reproducibility, and agreement of rotating Scheimpflug camera (Pentacam Oculus, Wetzlar, Germany) and ultrasound pachymetry in measuring central thickness of keratoconic corneas.
• DESIGN: Method-comparison study.
• METHODS: In 33 patients with keratoconus (one eye per patient), two examiners each used both pachymetric methods to measure central corneal thickness (CCT); in the same session, measurements then were repeated by examiner 1 (A.M.). The difference between two examiners, and between first and second measurements by examiner 1, with both methods and the difference between the two pachymetric methods in measuring central thickness of keratoconic corneas were noted.
• RESULTS: With the rotating Scheimpflug camera, inter-examiner correlation was higher (intra-class correlation coefficient [ICC], 0.98 vs 0.76) and inter-examiner variability was lower (95% limits of agreement [95% LoA], -14.8 to 13.8 μm vs -18.0 to +49.5 μm) than with ultrasound pachymetry. Both methods showed close first- to second-measurement correlation (ICC, > 90), but the rotating Scheimpflug camera had lower variability (95% LoA, -14.5 to 14.2 μm vs -27.4 to 26.0 μm). Mean CCT was 478.9 ± 34.6 μm with the rotating Scheimpflug camera and 486.6 ± 30 μm with ultrasound pachymetry. Although the mean difference was small (−7.8 μm), the 95% LoA (−43.8 to 28.2 μm) showed that the difference between the two methods can be considerable.
• CONCLUSIONS: In keratoconic corneas, the rotating Scheimpflug camera provides measurements of central thickness that are more reproducible and repeatable than those obtained with ultrasound pachymetry. The rotating Scheimpflug camera seems to be suitable for disease staging and follow-up, when corneal thickness measurements may be repeated over time by different examiners. (Am J Ophthalmol 2007;144: 712–718. © 2007 by Elsevier Inc. All rights reserved.)

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Keratoconus is the most frequent corneal ectatic dystrophy and is characterized by progressive noninflammatory corneal thinning with well-described slit-lamp findings.1,2 In this disorder, corneal thickness measurement is used for diagnosis,3 staging,4 follow-up, and planning surgical procedures.4–8 Currently, the clinical method most widely used to measure corneal thickness is ultrasound pachymetry; this has the advantages of ease of use, portability, and low cost and has been shown to have a high degree of intra-examiner, inter-examiner, and inter-instrument reproducibility in normal corneas.9–12 However, major limitations of ultrasound pachymetry are the need for cornea–probe contact, as well as the variability of measurements caused by probe misalignment or centering and changes in the speed of sound in corneal tissues with different degrees of hydration.
These limitations have led to the introduction of several optical technologies that offer the advantages of a noncontact technique and objective determination of the center of the cornea10,13–18. Among these, the rotating Scheimpflug camera (Pentacam Oculus, Wetzlar, Germany) calculates thickness and curvature values for the entire cornea, determining its front and back surfaces. Recent studies have shown that, in normal corneas and in corneal grafts, this method provides central corneal thickness (CCT) measurements that are reproducible, repeatable, and comparable with those obtained with ultrasound pachymetry.19–23
In keratoconus, the corneal thinning and corneal shape irregularity may reduce the reproducibility and repeatability of the rotating Scheimpflug camera and ultrasound pachymetry. Moreover, in eyes with keratoconus, a low inter-examiner and intra-examiner variability in measuring corneal thickness is required: in clinical practice, this parameter frequently is remeasured in the same eye over time, and in some cases by different examiners, during disease monitoring. This study investigates and compares the inter-examiner reproducibility and the intra-examiner repeatability of the rotating Scheimpflug camera and ultrasound pachymetry in measuring the central thickness of keratoconic corneas; it also assesses agreement between the two pachymetric methods in these eyes.
METHODS

CCT was measured with the rotating Scheimpflug camera and ultrasound pachymetry in 33 patients with previously diagnosed keratoconus (22 men, 11 women; mean age ± standard deviation [SD], 37 ± six years; range, 19 to 61 years). The initial diagnosis of keratoconus was based on clinical slit-lamp findings and associated characteristic Placido-based topographic patterns. Slit-lamp findings included one or more of the following signs: stromal thinning, conical protrusion, Fleischer ring, Vogt striae, and anterior stromal scarring. The mean simulated keratometric values obtained with Placido-based topography ranged from 41.65 to 64.00 diopters (D; mean ± SD,
49.54 ± 5.86 D) and corneal astigmatism ranged from 1.40 to 9.60 D (mean ± SD, 3.73 ± 1.65 D). Eyes with previous acute corneal hydrops or a history of corneal surgery were excluded from the study.

The CCT in one eye of each patient was measured with both methods by two independent examiners (examiner 1 (A.M.) and 2 (B.M.)) in a single session; the examiners worked in random order according to a computer-generated random number table. During the same session, measurements then were repeated, with both methods, by examiner 1 alone five to 10 minutes later. For each method, differences between first measurements obtained by the two examiners were used to assess inter-examiner reproducibility; differences between the first and second

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**TABLE 2. Intra-examiner Repeatability of Rotating Scheimpflug Camera and of Ultrasound Pachymetry in Measuring Central Corneal Thickness of Keratoconic Eyes**

<table>
<thead>
<tr>
<th></th>
<th>Rotating Scheimpflug Camera</th>
<th>Ultrasound Pachymetry</th>
</tr>
</thead>
<tbody>
<tr>
<td>Intra-class correlation coefficient (95% confidence interval)</td>
<td>0.98 (0.95 to 0.99)</td>
<td>0.91 (0.82 to 0.95)</td>
</tr>
<tr>
<td>Repeatability coefficient</td>
<td>14.1</td>
<td>26.2</td>
</tr>
<tr>
<td>95% Limits of agreement (µm)</td>
<td>-14.5 to 14.2</td>
<td>-27.4 to 26.0</td>
</tr>
</tbody>
</table>

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**FIGURE 2.** Scatterplots demonstrating the differences between first and second measurements of CCT of individual keratoconic eyes (Top) with the rotating Scheimpflug camera and (Bottom) with ultrasound pachymetry, plotted against the mean values of first and of second measurements. The mean difference is represented by the dotted line and the 95% LoA are represented by the solid lines. The variability between first and second measurements, expressed by the width of the 95% LoA, was larger with ultrasound pachymetry than with the rotating Scheimpflug camera.
measurements obtained by examiner 1 were used to assess intraexaminer repeatability. To assess method agreement, the means of the two initial measurements (by examiner 1 and examiner 2) for each pachymetric method were calculated and compared.

All measurements were taken between 2 P.M. and 5 P.M., all subjects having been awake for at least two hours. The eye to be examined was chosen using a computer-generated random number table, except for patients who had previously undergone corneal surgery for keratoconus in one eye. For each eye, the rotating Scheimpflug camera was used for thickness measurement, followed by the ultrasound pachymeter Allergan-Humphrey 850 (Allergan-Humphrey, Dublin, California, USA), within a three- to 10-minute interval. Calibration of the Pentacam (software version 1.14) was checked by the manufacturer at the beginning of the study; a single measurement was obtained for each eye with this method. The patient was asked to open both eyes and to stare at the fixation target. The automatic release mode was used to reduce operator-dependent variables, and 25 single images were captured by the rotating camera for each eye within two seconds. The Pentacam pachymetric measurement at the pupil center was not read until after ultrasound pachymetry had been completed.

The Allergan-Humphrey 850 ultrasonic pachymeter has a transducer frequency of 12 MHz and a calibration velocity of 1640 ms\(^{-1}\) for normal human corneal thickness at 34.5 C. Calibration of the instrument was checked using its test block (529 \(\mu\)m; SD, 11 \(\mu\)m). The cornea was anesthetized with topical 0.4% oxybuprocaine hydrochloride. The seated patient was asked to look at a distant fixation target straight ahead. The probe of the pachymeter was aligned perpendicular and central to the pupil as precisely as possible, and the mean of five measurements was calculated.

**STATISTICAL ANALYSIS:** For each method, intra-class correlation coefficients (ICCs) were estimated to assess interexaminer reproducibility using a two-way random effects analysis of variance (ANOVA) and to assess intra-examiner repeatability using a one-way random effects ANOVA. Method-specific inter-examiner reproducibility and intra-examiner repeatability also were determined using the approach proposed by Bland and Altman, which includes calculation of the repeatability coefficients and the 95% limits of agreement (95% LoA).

Agreement between the two methods was assessed by calculating the 95% LoA, using the mean of the two examiners' first measurements. Linear regression was used to evaluate whether, after adjusting for mean CCT, simulated keratometry modified the mean difference between the two methods.

All analyses were carried out using the software package STATA version 8.0 (StataCorp LP, College Station, Texas, USA). The study had 80% power to detect statistically significant (\(\alpha = 0.05\), two-tails) differences between the two methods of at least 7.5 \(\mu\)m, considering an SD for the difference of 15 \(\mu\)m.

**RESULTS**

- **REPRODUCIBILITY AND REPEATABILITY OF THE TWO METHODS IN CENTRAL THICKNESS MEASUREMENT OF KERATOCONIC CORNEAS:** Analysis of inter-examiner reproducibility revealed a lower ICC, higher coefficient of

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FIGURE 3. Scatterplot demonstrating the differences between the rotating Scheimpflug camera and ultrasound pachymetry in measuring CCT of individual keratoconic eyes, plotted against the mean value obtained with the two methods. The mean difference is represented by the dotted line and the 95% LoA are represented by the solid lines. The size of each dot is proportional to the square of the corresponding mean keratometric value. Note the large difference (> 60 \(\mu\)m) in the three eyes with mean keratometric readings (K) of more than 60 diopters.
repeatability, and wider 95% LoA for ultrasound pachymetry compared with the rotating Scheimpflug camera (Table 1). The 95% LoA for ultrasound pachymetry indicated that differences between examiners could be of clinical significance with this method. Figure 1 graphically illustrates the difference between the two examiners in measuring each individual CCT, with both methods, through Bland-Altman plots.

Intra-examiner repeatability was found to be high with both methods (ICC, > 90), although the ICC value was statistically significantly lower for ultrasound pachymetry. Consistently, the repeatability coefficient for this method was approximately twice that of the rotating Scheimpflug camera, and the 95% LoA were larger (Table 2). Individual differences between first and second measurements with each method are presented graphically in form of Bland-Altman plots (Figure 2).

**AGREEMENT BETWEEN THE TWO METHODS IN CENTRAL THICKNESS MEASUREMENT OF KERATOCONIC CORNEAS:** Mean ± SD CCT was 478.9 ± 34.6 μm (range, 396 to 544 μm) with the rotating Scheimpflug camera and 486.6 ± 30 μm (range, 428 to 550 μm) with ultrasound pachymetry. The CCT measurement was systematically lower with the rotating Scheimpflug camera than with ultrasound pachymetry, with a mean difference of −7.8 μm (95% confidence interval [CI], −14.0 to −1.6). The 95% LoA between the two methods were −43.8 to 28.2 μm. However, part of this disagreement was because of the three eyes with mean simulated keratometric values of more than 60 D (Figure 3). When these eyes were excluded from the analysis, the mean difference between the two methods dropped to −4.3 μm (95% CI, −9.3 to 0.7 μm) and the 95% LoA to −32.4 to 23.8 μm. Linear regression analysis revealed a positive quadratic association between mean simulated keratometry and mean difference between the two methods (P < .01), implying that the difference between them increases with increased mean keratometric values.

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**DISCUSSION**

IN KERATOCONUS, THE CORNEA TENDS TO BE THINNER than normal, both centrally and peripherally, so that reproducible, repeatable, and accurate measurements of corneal thickness are required for the diagnosis, staging, and follow-up of this disease, as well as for planning surgical procedures. Although a comprehensive clinical examination plus Placido-based topography have good accuracy in detecting keratoconus, measurement of corneal thickness may provide useful corroborative evidence for diagnosis.

Because the degree and progression of corneal thinning in keratoconus vary widely among patients, measurement of corneal thickness is repeated over time to stage and monitor the disease. This measurement also is required to plan surgical procedures, including penetrating keratoplasty, deep anterior lamellar keratoplasty, and intrastromal corneal ring segment implantation.

This study assessed inter-examiner reproducibility and intra-examiner repeatability of the rotating Scheimpflug camera and ultrasound pachymetry in measuring central thickness of keratoconic corneas. Although the rotating Scheimpflug camera can measure the thickness of the entire cornea, for the purposes of this study, it might have biased comparison of the two methods if thickness was measured at sites, such as the cone apex, previously located with topographic analysis. Apart from those with a history of acute hydrops, the study included keratoconic eyes with a wide range of disease severity: mean simulated keratometric values and corneal astigmatism ranged, respectively, from 41.65 to 64.00 D and from 1.40 to 9.60 D. In this series of patients, the rotating Scheimpflug camera provided measurements of CCT that were highly reproducible and repeatable. Ultrasound pachymetry was less reproducible and repeatable; importantly, when using ultrasound pachymetry in keratoconic corneas, different examiners and measurements repeated by the same examiner may record thickness values that are clinically different.

With the rotating Scheimpflug camera, inter-examiner reproducibility and intra-examiner repeatability were similar. With this method, the center of the cornea is detected automatically and measurement alignment is not examiner related, so that reproducibility and repeatability of this method depend mainly on the patient’s correct gaze. Moreover, with this method, reproducibility and repeatability in measuring the central thickness of keratoconic corneas did not change with disease severity; they were also similar to those measurements observed in normal corneas and slightly better than those in corneal grafts.

In a previous study, in which the same examiners used the rotating Scheimpflug camera in normal corneas and corneal grafts, the 95% LoA between examiners were −17.8 to 12.6 μm and −18.2 to 21.8 μm, respectively, and the 95% LoA of repeated measurements were −12.5 to 12.5 μm and −12.8 to 22.2 μm. It thus seems that reproducibility and repeatability of this method are not be influenced by the steep curvature and irregular prolate shape of keratoconic corneas.

With ultrasound pachymetry, the difference between examiners was larger than that between repeated measurements by the same examiner. Ultrasound pachymetry is a manual technique, and thus its accuracy may be examiner dependent, because it relies on corneal touch technique, correct centering and perpendicular alignment of the probe. For these reasons, even when performed in normal corneas by expert examiners, inter-examiner reproducibility is slightly lower than intra-examiner repeatability.

However, the intervals of 95% LoA indicated that both inter-examiner reproducibility and intra-examiner repeatability of ultrasound pachymetry in keratoconic corneas were lower than in normal corneas, and even than in...
Typically, keratoconic corneas have more irregular and variable thickness values than normal corneas. Thus, slight but systematic differences between examiners in centering and aligning the probe may cause wider inter-examiner variability than that reported in normal corneas. In keratoconic eyes, the distorted corneal shape can make it difficult to determine the real position of the pupil center. For these reasons, when measurements are repeated, as occurred here, intra-examiner repeatability even of expert examiners may be lower than that in normal corneas. Moreover, in this study, we took the average value of five readings with ultrasound pachymetry, but only one reading with the rotating Scheimpflug camera. Averaging readings tends to increase reproducibility and repeatability. When a single ultrasound pachymetry reading was considered, and when the central three values (after eliminating the highest and lowest values) were taken, inter-examiner variability increased, with 95% LoA intervals of $-22.8$ to $59.5$ $\mu$m and $-21$ to $58.4$ $\mu$m, respectively.

The measurements provided by the rotating Scheimpflug camera were lower than those obtained with ultrasound pachymetry. Although the mean difference ($-7.8$ $\mu$m) vs ultrasound pachymetry may be judged not to be clinically significant, the variability expressed by the 95% LoA values (from $-43.8$ to $28.2$ $\mu$m) suggests that large differences between these two pachymetric methods can occur and that they cannot be used interchangeably for clinical or research purposes. In particular, the discrepancy between the two methods seemed to increase with increased mean keratometric values; in the three eyes with mean keratometric values of more than 60 D, CCT values produced by the rotating Scheimpflug camera were 30 to $60$ $\mu$m thinner than those of ultrasound pachymetry. Although this finding is limited to a small number of cases, it is in agreement with the study by Ucakhan and associates, who reported that the discrepancy between rotating Scheimpflug camera and ultrasound pachymetry was higher in eyes with severe disease (mean keratometric reading, $>55$ D; mean difference, $-27$ $\mu$m) than in those with mild disease (mean keratometric reading, $<47$ D; mean difference, $-3.3$ $\mu$m).

The tendency of the rotating Scheimpflug camera to underestimate slightly the measurement of CCT vs ultrasound pachymetry also was reported in normal corneas, where the mean difference between the two methods ranged from $-5$ to $-9$ $\mu$m. These instruments use different methods to generate corneal thickness data (optical vs ultrasound), and detailed information on the algorithms that the Pentacam rotating Scheimpflug camera uses to generate these data is not available. The discrepancy between rotating Scheimpflug camera and ultrasound pachymetry in measuring the CCT may be increased further by some histopathologic features affecting the optical properties and hydration of corneal tissue in keratoconic corneas; irregular and elongated epithelial cells, altered organization of collagen fibers, clear stromal spaces, and endothelial irregularity have been observed in these corneas. It can be speculated that these alterations may influence the path of light rays and the velocity of ultrasound, leading to a distorted measurement. However, further studies are needed to clarify this issue.

In conclusion, in keratoconic corneas, the rotating Scheimpflug camera provides measurements of central thickness that are more reproducible and repeatable than those obtained with ultrasound pachymetry; this makes the rotating Scheimpflug camera suitable for disease staging and follow-up, when measurements are repeated over time, in some cases by different examiners. In comparison with ultrasound pachymetry, on average the rotating Scheimpflug camera slightly underestimates the central thickness of keratoconic corneas; particularly in eyes with a severe stage of the disease, large differences between the two methods are possible.

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REFERENCES


Biosketch

Ugo de Sanctis, MD, PhD, completed the post-graduate training in Ophthalmology, and the post-graduation doctorate in Cornea and External Diseases at the Ophthalmology Institute, University of Turin, Italy. Currently, Dr de Sanctis is an Assistant Professor of Ophthalmology and Head of Cornea Service at the Medical University of Turin. His clinical and research interests include corneal and external diseases, refractive surgery, and cataract surgery.