Biomechanical Responses of Healthy and Keratoconic Corneas Measured Using a Noncontact Scheimpflug-Based Tonometer

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Purpose. To examine the repeatability of measurements obtained using a noncontact Scheimpflug-based tonometer (Corvis ST) and investigate potential differences in these parameters between healthy and keratoconic (KC) corneas.

Methods. Forty-five keratoconic eyes and 103 healthy eyes were examined using biomicroscopy, corneal tomography and the Corvis ST (CST).

Results. Intraocular pressure and central corneal thickness (CCT) were highly repeatable (intraclass correlation [ICC] > 0.70, coefficient of variation [CV] < 0.20). Deformation amplitude (DA) and applanation-1 time (A1T) were fairly repeatable (ICC > 0.60, CV < 0.20). There was no association between DA and age, sex, or ethnicity in healthy eyes. There was a greater mean DA in the KC group compared with 46 age-matched healthy eyes (KC 1.37 $\pm$ 0.21 mm, healthy 1.05 $\pm$ 0.11 mm, $P < 0.001$). Multivariate analysis showed DA in KC was predicted by IOP, CCT, and the steepest simulated keratometry value ($P = 0.03$, $P = 0.03$, $P = 0.001$ respectively, $R^2 = 0.75$). A subgroup analysis of healthy and KC eyes with comparable mean CCT and IOP highlighted a statistically significant difference in mean DA (KC 1.25 $\pm$ 0.08 mm, thin healthy 1.13 $\pm$ 0.09 mm, $P = 0.006$). Receiver-operating characteristic analysis showed an area under the curve of 0.77 (95% CI 0.61–0.93, $P = 0.006$) but no ideal cutoff value for DA.

Conclusions. Key parameters assessed by the CST are repeatable. Keratoconus is associated with greater DA than in healthy eyes, even when controlled for CCT and IOP. Deformation amplitude may be a useful adjunct in keratoconus assessment and monitoring, but cannot solely discriminate between healthy and keratoconic corneas.

Keywords: Corvis ST, corneal deformation amplitude, non-contact tonometry, diagnostic instruments, repeatability

The human cornea is considered to have both viscous and elastic properties.[1,2] It deforms in response to a given force in a biphasic fashion and some energy is dissipated during this process, increasing the time taken to assume its original contour.[3]

Describing corneal biomechanical properties has been of particular interest due to the myriad potential clinical applications, such as the assessment and monitoring of corneal diseases and glaucoma[4,5] and in the assessment for corneal refractive surgery.[6,7] Keratoconus (KC) is an ectatic disease of the cornea in which there is progressive thinning and protrusion of the corneal apex. Currently, the diagnosis of KC is based on a combination of clinical and corneal tomographic features.[8,9] In its earliest stages, KC is often difficult to diagnose (forme fruste, or subclinical KC), yet it is crucial to identify in patients considering corneal refractive surgery.[7,9–11]

In vivo confocal microscopy studies have demonstrated cellular changes, such as reduced keratocyte density even in the early stages of KC.[12–14] Changes in the biomechanical properties of the cornea may therefore be detectable before the tomographic and clinical signs of KC become apparent.

Few studies have explored the biomechanical behavior of healthy and keratoconic corneas in vitro and such studies have been limited by nonphysiological IOP and hydration states.[15,16] Until recently, only one device, the Ocular Response Analyzer (ORA; Reichert Ophthalmic Instruments, Buffalo, NY, USA), has been widely commercially available to estimate the biomechanical properties of the human cornea in vivo.

The Corvis ST (CST; Oculus, Wetzlar, Germany) is a new noncontact tonometer that emits a pulse of air, which applanates and indents the cornea. It differs from the ORA in that it uses an ultra–high speed Scheimpflug camera (4330 frames/s) to record in vivo cross-sectional images of the cornea during and after application of the air pulse. The aims of this study were to determine the repeatability of parameters measured by the CST, determine the mean and range for these parameters in healthy corneas, examine possible variation with age, sex, and ethnicity in healthy corneas, and to investigate how these parameters are affected in KC.

Methods

Patients

Patients with KC, diagnosed by fellowship trained corneal specialists, were recruited from anterior segment clinics at

Applanation 1	Abbreviation	Description
Time	A1T	Time at first flattening of the cornea
Length	A1L	Length of flattened cornea at applanation 1
Velocity	A1V	Inward velocity of the cornea at applanation 1

Highest Concavity	Abbreviation	Description
Deformation Amplitude	DA	Corneal displacement at highest concavity
Radius of Curvature	RoC	Radius of curvature of the cornea at highest concavity
Peak Distance	PD	Distance between the corneal peaks at maximal concavity

Applanation 2	Abbreviation	Description
Time	A2T	Time at second flattening of the cornea
Length	A2L	Length of flattened cornea at applanation 2
Velocity	A2V	Outward velocity of the cornea at applanation 2

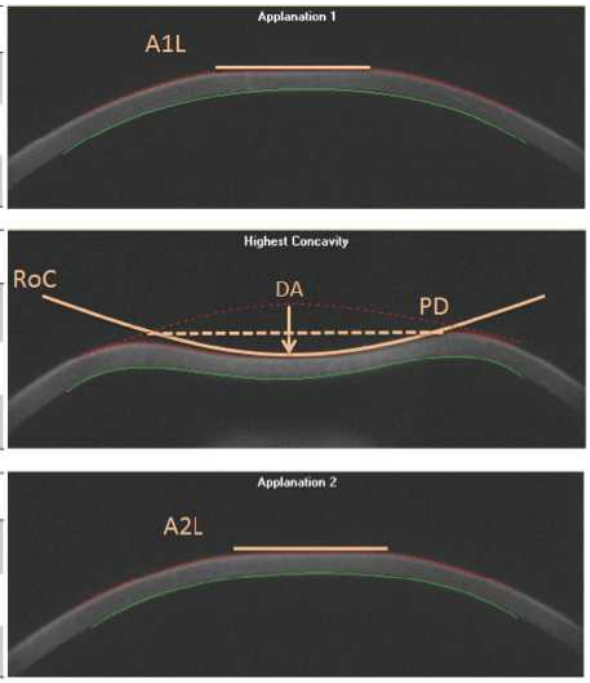


Figure 1. Parameters of the Corvis ST.

Greenlane Clinical Centre, Auckland District Health Board, New Zealand. Inclusion criteria were based on the Rabinowitz definition of KC relating to topographic features of the anterior corneal surface:

- An increased area of corneal power surrounded by concentric areas of decreasing power;
- Inferior-superior power asymmetry; and
- Skewing of the steepest radial axes above and below the horizontal meridian, with less than or equal to 150° between arms.[8]

Subjects with healthy corneas were recruited from staff and relatives of patients at the same center. Inclusion criteria for healthy subjects were absence of the abovementioned characteristics of KC, and posterior elevation on pentacam less than 29 μm.[17] Patients were excluded from both groups if there was a history of any previous ocular or periocular trauma or surgery, contact lens wear, or any ocular or systemic condition that may affect the eye. Keratoconus severity was classified based on the Collaborative Longitudinal Evaluation of Keratoconus (CLEK) study criteria using the steepest keratometry value (Kmax): mild Kmax less than 45 diopters (D), moderate Kmax 45 to 52 D, and severe Kmax greater than 52 D.[18]

Demographic data including age, sex, and ethnicity were collected. Only one eye from each subject was examined. In the control group, only left eyes were examined. In the KC group the left eye was examined unless it had evidence of previous corneal hydrops or had undergone corneal transplantation, in which case, the right eye was examined.

Institutional review board/ethics committee approval was obtained. Informed consent was also obtained from each patient after explanation of the nature and possible consequences of the study prior to participation. This study adhered to the tenets of the Declaration of Helsinki.

Examinations

All examinations were performed by a single experienced investigator (NQA). All eyes were examined by slit-lamp biomicroscopy to determine the health status of the cornea. Tomography was performed on the Pentacam HR (Oculus) to obtain measurements of the corneal thickness and simulated keratometry values.

Corvis ST (CST, software version V2.x; Oculus, Wetzlar, Germany) examination was performed by positioning the patient onto the chin-rest and forehead band, and centering the instrument on the cornea using the four red alignment markers on the computer screen. Once centered, the instrument automatically emits a focused puff of air at a pressure of 60 mm Hg from a nozzle, which was 3.05 mm in diameter aimed at the cornea at a distance of 11 mm. The Schiempflug camera was angled at 45°, and captured a video sequence that included: the first inward applanation or flattening of the cornea, deformation at maximal concavity, and the second outward applanation or flattening as the cornea resumes its original contour. Intraocular pressure measurements by the CST were derived from measurement of time to first applanation of the cornea, the same principle used by noncontact tonometry. This study used the CST software version V2.x which did not include the "Corneal Compensated IOP" parameter provided by subsequent software updates. The parameters measured by the CST, their definitions, and abbreviations are shown in Figure 1. In our analyses, we used IOP measurements and CCT measurements taken by the CST, and Kmax measurements taken by the Pentacam HR (Oculus).

A subset of healthy subjects underwent repeatability testing, which comprised of three sets of measurements. The first set consisted of three successive CST examinations 2 to 5 minutes apart. The second set consisted of CST examinations at 4 different times of day: between 8 and 10 AM, 10:30 AM and 12 PM, 1 and 2:30 PM, and 3 and 5:30 PM. The third set consisted of three CST examinations, each taken at the same time of day, on three different days.

Statistical Analyses

Statistical analysis was performed with input from an experienced biostatistician using Microsoft Office Excel

Table 1. Repeatability of the Corvis ST Parameters for Three Measurements Taken 2 to 5 Minutes Apart

Parameter	Precision	Repeatability	CV	Individual ICC (95% CI)	Average ICC (95% CI)	*P* Value
IOP	1.6 mm Hg	2.3 mm Hg	6.10%	0.73 (0.53 to 0.87)	0.89 (0.77 to 0.95)	<0.001
CCT	19 μm	27 μm	1.83%	0.86 (0.73 to 0.94)	0.95 (0.89 to 0.980	<0.001
DA	0.08 mm	0.11 mm	3.64%	0.63 (0.39 to 0.82)	0.83 (0.66 to 0.93)	<0.001
RoC	0.48 mm	0.66 mm	3.23%	0.17 (−0.85 to 0.79)	0.38 (−0.31 to 0.73)	0.1
PD	0.56 mm	0.79 mm	15.90%	0.07 (−0.16 to 0.37)	0.41 (−0.18 to 0.74)	0.29
A1T	0.20 ms	0.28 ms	1.40%	0.73 (0.53 to 0.87)	0.89 (0.769 to 0.951)	<0.001
A1L	0.56 mm	0.79 mm	15.90%	0.20 (−0.06 to 0.50)	0.41 (−0.18 to 0.74)	0.07
A1V	0.05 $msec^{-1}$	0.08 $msec^{-1}$	17.10%	0.21 (−0.03 to 0.51)	0.45 (−0.10 to 0.76)	0.05
A2T	0.52 ms	0.77 ms	1.20%	0.53 (0.28 to 0.75)	0.77 (0.54 to 0.90)	<0.001
A2L	0.95 mm	1.34 mm	26.30%	−0.16 (−0.32 to 0.10)	−0.73 (−2.72 to 0.26)	0.9
A2V	0.08 $msec^{-1}$	0.11 $msec^{-1}$	10.95%	0.33 (0.05 to 0.61)	0.60 (0.15 to 0.83)	0.009
T HC	0.73 ms	1.04 ms	2.20%	0.24 (−0.3 to 0.53)	0.48 (−0.1 to 0.78)	0.04

RoC, radius of curvature; PD, peak distance; A1L, applanation 1 length; A1V, applanation 1 velocity; A2L, applanation 2 length; A2T, applanation 2 time; A2V, applanation 2 velocity; T HC, time to highest concavity.

(Microsoft, Redmond, WA, USA) and SPSS software version 19.0 (SPSS, IBM, Chicago, IL, USA). We performed multivariate analyses for factors predictive of deformation amplitude separately in healthy and keratoconic patients as there were significant interactions between group status and the predictive variables. Additionally, the coefficient of the relationship between each predictive variable and DA was different in the healthy and KC groups, thus making a combined analysis invalid.

Repeatability. The Bland-Altman methodology for measurement error was used to test the repeatability of all CST parameters.[19] The mean within-subject standard deviation (SD) S_w was calculated. Precision (Pr) was calculated using the formula $1.96 \times S_w$ and repeatability (R) was calculated using the formula $2.77 \times S_w$. The coefficient of variation (CV) was calculated by the dividing the mean of within-subject means by S_w. Intraclass correlation (ICC) was calculated using ANOVA. An ICC of more than 0.70, and a CV less than 0.20 were considered highly repeatable, an ICC of more than 0.60 and CV less than 0.20 were considered fairly repeatable.

Characteristics of Healthy Corneas. Descriptive statistical testing was conducted on baseline characteristics. The Shapiro-Wilk test was conducted to determine if age, CCT, IOP, Kmax, and DA, were normally distributed. A two-tailed Pearson's correlation coefficient was calculated for age and DA in this group. The independent *t*-test was conducted to determine any differences in DA between males and females, and one-way ANOVA test to determine differences in mean DA between ethnicities.

Characteristics of Keratoconic Corneas in Comparison With Healthy Corneas. For keratoconic eyes, descriptive statistical testing was conducted on baseline characteristics, the Shapiro-Wilk test was conducted to determine if IOP, CCT, Kmax, and DA were normally distributed.[20] An age-matched subgroup of healthy eyes was selected for comparative analyses. Baseline characteristics of these two groups were tested for significant differences: continuous data with independent *t*-tests, categorical data with χ^2 tests. The mean and SD of each CST parameter were compared between KC and healthy groups by independent *t*-tests. For each parameter where a statistically significant difference was demonstrated between healthy and KC eyes, multiple linear regression analyses were performed to determine predictors of that parameter in each group.

Another subgroup analysis was conducted to compare KC eyes with higher corneal thickness (≥460 μm) and healthy eyes with thin corneas (≤510 μm). *P* values of 0.05 or less were considered to be statistically significant.

Results

Repeatability

In a subgroup of 22 healthy eyes, three examinations taken 2 to 5 minutes apart on the same day demonstrated high repeatability in IOP, CCT, and applanation-1 time (A1T), while DA was fairly repeatable. For the examinations performed at four different times of day IOP, CCT, and A1T were highly repeatable; DA was fairly repeatable. For examinations taken at the same time of day on 3 different days IOP and CCT were highly repeatable, while DA and A1T were fairly repeatable. Therefore, IOP and CCT were all highly repeatable, DA and A1T were fairly repeatable when taken a few minutes apart, at different times of day and on different days.

The precision, repeatability, CV, and ICC are summarized for three measurements taken 2 to 5 minutes apart (Table 1), four measurements taken at different times of day (Table 2), and three measurements taken on three different days (Table 3).

Characteristics of Healthy Corneas

One hundred three healthy subjects were recruited with a mean age of 38.6 ± 14.2 years, lower quartile 26 years, upper quartile 49 years, and range 13 to 73 years. There was a larger proportion of females 63% (65) than males 37% (34). The majority of patients were of European ethnicity (52.4%, $n = 56$), followed by Indian subcontinent (14.6%, $n = 15$), Asian (13.6%, $n = 14$), Pacific Island nations (8.7%, $n = 9$), Middle Eastern (5.8%, $n = 6$), Maori (2.9%, $n = 3$), and African descent (2.9%, $n = 3$). Values for all CST parameters measured in healthy eyes are shown in Table 4. Deformation amplitude, CCT, Kmax, IOP, and age were normally distributed (Shapiro-Wilk test, $P \geq 0.05$). There was no correlation of DA with age ($r = 0.129$, $P = 0.20$), and no statistically significant difference in DA between male and female groups ($P = 0.32$) or between different ethnic groups (one way ANOVA, $P = 0.19$).

Characteristics of Keratoconic Corneas in Comparison With a Subgroup of Healthy Corneas

Forty-five patients with KC were recruited. According to CLEK study KC severity criteria,[18] one patient (2%) had mild disease, 18 (40%) had moderate disease, and 26 (58%) had severe disease. Deformation amplitude, IOP, CCT were normally distributed (Shapiro-Wilk test, $P \geq 0.05$), however age was not. A subgroup of 46 age-matched patients, with healthy corneas, was selected to compare with the KC group. The

Table 2. Repeatability of the Corvis ST Parameters Measured at Four Different Times of Day

Parameter	Precision	Repeatability	CV	Intraclass Correlation (95% CI)	*P* Value
IOP	1.83 mm Hg	2.59 mm Hg	6.72%	0.76 (0.60 to 0.88)	<0.001
CCT	8 μm	11 μm	7.41%	0.98 (0.96 to 0.99)	<0.001
DA	0.09 mm	0.12 mm	4.18%	0.64 (0.45 to 0.81)	<0.001
RoC	1.38 mm	1.95 mm	9.47%	0.44 (0.23 to 0.66)	<0.001
PD	1.05 mm	1.48 mm	10.89%	0.53 (0.32 to 0.73)	<0.001
A1T	0.21 ms	0.29 ms	1.46%	0.78 (0.63 to 0.89)	<0.001
A1L	0.54 mm	0.77 mm	15.56%	0.14 (−0.04 to 0.40)	0.07
A1V	0.05 $msec^{-1}$	0.08 $msec^{-1}$	3.69%	0.07 (−0.08 to 0.32)	0.19
A2T	0.47 ms	0.66 ms	10.63%	−0.05 (−1.02 to 0.27)	0.55
A2L	0.82 mm	1.17 mm	2.20%	0.16 (−0.02 to 0.42)	0.04
A2V	0.08 $msec^{-1}$	0.11 $msec^{-1}$	12.18%	0.35 (0.13 to 0.59)	<0.001
T HC	1.04	1.48	3.15%	0.15 (−0.03 to 0.40)	0.05

Table 3. Repeatability of the Corvis ST Parameters, for Measurements Taken at the Same Time of Day, on Different Days

Parameter	Precision	Repeatability	CV	Intraclass Correlation (95% CI)	*P* Value
IOP	1.92 mm Hg	2.72 mm Hg	7.17%	0.76 (0.59 to 0.88)	<0.001
CCT	11 μm	15 μm	1.07%	0.998 (0.996 to 0.999)	<0.001
DA	0.09 mm	0.12 mm	4.03%	0.65 (0.42 to 0.82)	<0.001
RoC	1.07 mm	1.51 mm	7.19%	0.42 (0.16 to 0.67)	<0.001
PD	1.98 mm	2.8 mm	21.19%	−0.04 (−0.26 to 0.23)	0.67
A1T	0.25 ms	0.35 ms	2.08%	0.69 (0.47 to 0.85)	<0.001
A1L	0.65 mm	0.92 mm	9.64%	−0.07 (−0.26 to 0.23)	0.67
A1V	0.06 $msec^{-1}$	0.09 $msec^{-1}$	3.33%	−0.03 (−0.23 to 0.27)	0.56
A2T	0.55 ms	0.77 ms	1.94%	0.51 (0.25 to 0.74)	<0.001
A2L	0.93 mm	1.32 mm	25.37%	−0.05 (−0.25 to 0.26)	0.62
A2V	0.09 $msec^{-1}$	0.13 $msec^{-1}$	13.42%	0.23 (−0.03 to 0.52)	0.04
T HC	0.75	1.06	2.27%	0.09 (−0.14 to 0.40)	0.23

baseline characteristics for these two groups are shown in Table 5.

A comparison of mean values for each CST parameter in the two groups is provided in Table 6. The mean DA was significantly greater in the KC group compared with healthy eyes ($P < 0.001$; Fig. 2). The mean time at A1T was significantly shorter in KC than in healthy eyes ($P < 0.001$).

There was no significant difference in the mean length of applanation 1 (the distance over which the air pulse was applied; $P = 0.64$), and mean length of applanation 2 ($P = 0.4$) between the two groups.

Table 4. The Mean, SD, and Range of Values for Each Parameter Measured by the Corvis ST in Healthy Eyes ($n = 103$)

Corvis ST Parameter	Mean ± SD	Range
IOP, mm Hg	14.6 ± 2.7	10.0–25.5
Corneal thickness, μm	546 ± 36	467–633
DA, mm	1.07 ± 0.10	0.84–1.31
PD, mm	4.75 ± 1.00	2.07–7.04
RoC, mm	7.51 ± 0.84	5.47–9.55
A1L, mm	1.79 ± 0.30	0.93–2.44
A1V, m/s	0.15 ± 0.03	0.06–0.24
A1T, ms	7.21 ± 0.33	6.07–8.59
A2L, mm	1.93 ± 0.47	1.09–2.83
A2V, m/s	−0.33 ± 0.06	21.38–2.57
A2T, ms	22.57 ± 0.46	21.36–23.57
T HC, ms	16.97 ± 0.45	15.71–18.82

Multiple Linear Regression

In view of the significant differences in mean DA and A1T between KC and age-matched controls, multiple linear regression analyses were performed to determine if age, sex, ethnicity, IOP, CCT, or Kmax are predictors of these parameters.

Table 5. Baseline Characteristics in Keratoconus Group ($n = 45$) and Age-Matched Healthy Eyes ($n = 46$) Assessed by the Corvis ST

Baseline Characteristic	Healthy ($n = 46$)	KC ($n = 45$)	*P* Value
Age Mean ± SD, y	26.7 ± 9.8	25.3 ± 12.6	
Range	13–59	9–59	
Males	52.2% (24)	62.2% (28)	0.45†
Females	47.8% (22)	37.8% (17)	
Ethnicity			
European	50.0% (23)	22.2% (10)	0.01†
Asian	13.0% (6)	2.2% (1)	0.12†
Pacific Peoples	6.5% (3)	46.75 (21)	<0.01†
Maori	4.3% (2)	20% (9)	0.05†
Other	28.3% (13)	8.9% (4)	0.04†
CCT Mean ± SD, μm	548 ± 38	432 ± 80	0.0001*
Kmax Mean ± SD, ΔD	43.4 ± 1.4	52.2 ± 7.7	0.0001*
IOP Mean ± SD, mm Hg	15.0 ± 3.1	9.4 ± 3.19	0.0001*

* Independent *t*-test.
† χ^2 test.

Table 6. Comparison of Each Corvis ST Parameter in 46 Age-Matched Healthy Eyes and 45 Keratoconic Eyes

Parameter	Healthy, $n = 46$, Mean ± SD	KCs, $n = 45$, Mean ± SD	Mean Difference	*P* Value
DA, mm	1.05 ± 0.11	1.37 ± 0.21	−0.32	<0.001
PD, mm	4.82 ± 0.99	4.55 ± 1.36	0.27	0.28
RoC, mm	7.51 ± 0.9	4.79 ± 1.19	2.72	<0.001
A1L, mm	1.81 ± 0.31	1.78 ± 0.28	0.03	0.64
A1T, ms	7.24 ± 0.38	6.68 ± 0.30	0.55	<0.001
A1V, $msec^{-1}$	0.15 ± 0.03	0.17 ± 0.05	−0.02	0.01
A2L, mm	1.85 ± 0.46	1.73 ± 0.5	0.12	0.4
A2T, ms	22.5 ± 0.5	22.95 ± 0.46	−0.44	<0.001
A2V, $msec^{-1}$	−0.33 ± 0.07	−0.51 ± 0.16	0.18	<0.001
T HC, ms	16.93 ± 0.42	16.89 ± 0.6	0.04	0.75

Deformation amplitude correlated significantly with lower IOP (KC: $P = 0.03$, healthy: $P = 0.001$) and greater Kmax in the KC group ($P = 0.01$). When IOP was excluded from the analysis, greater DA correlated significantly with thinner CCT in both groups (KC: $P = 0.03$, healthy: $P = 0.02$), Age, sex, and ethnicity were not statistically significant predictors of DA in either group (KC: $P = 0.84$, $P = 0.31$, $P = 0.55$, respectively; healthy: $P = 0.71$, $P = 0.90$, $P = 0.72$, respectively).

Applanation-1 time correlated significantly with higher IOP in both KC and healthy eyes ($P < 0.001$), and CCT in healthy eyes ($P = 0.02$). When IOP was excluded from the analysis, CCT was a significant predictor of A1T in KC ($P = 0.003$). Table 7 summarizes the significant predictive factors for each parameter that is different in KC.

In a further analysis, KC eyes with the greatest CCT values in this study sample were selected ($n = 17$, CCT $\geq$ 460 µm) and all had moderate KC (Kmax of 45–52 D). A subset of healthy eyes ($n = 18$) were selected as pachymetry-matched controls for comparison to the keratoconic subgroup. There were no differences in mean IOP or mean CCT between the two groups ($P = 0.05$, $P = 0.27$, respectively). There was a statistically significant difference in the mean DA ($P = 0.001$) and mean Kmax (<0.001) between the two groups (Table 8). Thus, mean DA was greater in KC than healthy eyes where CCT and IOP were matched. A receiver-operating characteristic analysis was performed (Fig. 3). The area under the curve was 0.77 ± 0.08 (95% CI 0.61–0.93, $P = 0.006$), however, there was no ideal cut-

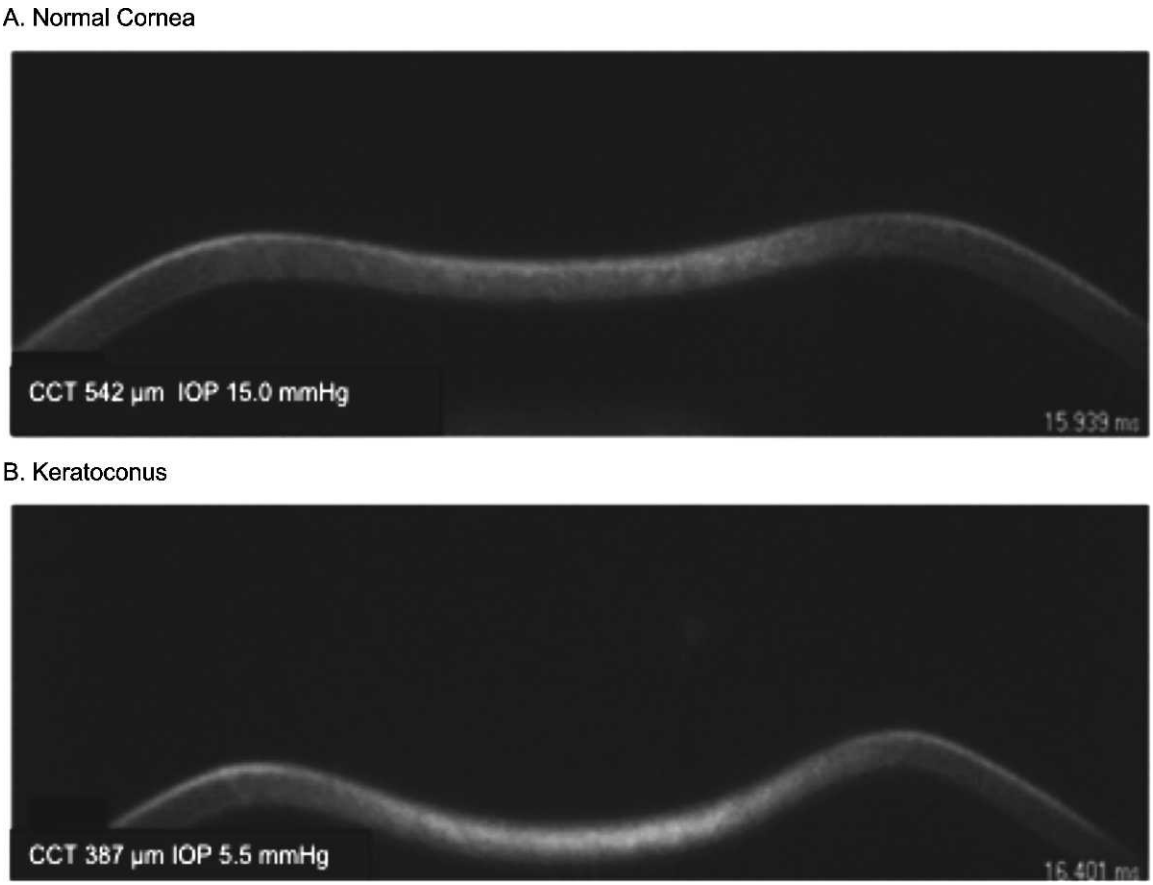


Figure 2. Corneal deformation of keratoconus compared with normal on the Corvis ST. (**A**) Healthy cornea, (**B**) KC. CCT, central corneal thicknes; IOP, intra-ocular pressure.

Table 7. Multiple Regression Analysis: Significant Predictors of Corvis ST Parameters in Healthy and Keratoconic Eyes

	Coefficient	95% CI	*P* Value
DA			
CCT KC,* μm	−0.87	−1.63 to −0.11	0.03
CCT normals,* μm	−1.06	−1.94 to −0.18	0.02
IOP KC, mm Hg	−20.4	−38.9 to −1.9	0.03
IOP normals, mm Hg	−18.7	−29.1 to −8.3	0.001
Sim Kmax KC (ΔD)	13.4	5.6 to 21.2	0.001
RoC			
CCT normals,* μm	8.82	1.04 to 16.6	0.03
A1T			
CCT KC,* μm	1.99	0.7 to 3.28	0.003
CCT normals, μm	2.19	0.38 to 4.00	0.02
IOP KC, mm Hg	0.10	0.09 to 0.10	<0.001
IOP normals, mm Hg	0.09	0.07 to 0.11	<0.001
Sim Kmax KC (ΔD)	49.19	4.07 to 94.31	0.03
A1V			
Sim Kmax KC (ΔD)	2.3	0.34 to 4.26	<0.001
IOP normals, mm Hg	−3.58	−5.85 to −1.31	0.003
A1L KC, mm	0.10	0.08 to 0.14	<0.001
A1L normals, mm	0.06	0.04 to 0.08	<0.001
A2T			
IOP KC, mm Hg,	−0.12	−0.18 to −0.07	<0.001
IOP normals, mm Hg	−0.12	−0.16 to −0.07	0.001
A2V			
CCT normals, μm	0.79	0.25 to 1.34	0.005
Sim Kmax KC (ΔD)	−11.28	−19.14 to −3.42	0.006
IOP normals, mm Hg	0.008	0.002 to 0.015	0.02

DA: R^2 KC group: 0.75. R^2 healthy group: 0.41; RoC: R^2 KC group: 0.49, R^2 healthy group: 0.14; A1T: R^2 KC group: 0.98, R^2 healthy group: 0.14; A1V: R^2 KC group: 0.68, R^2 healthy group: 0.63; A2T: R^2 KC group: 0.58, R^2 healthy group: 0.16; A2V: R^2 KC group: 0.51, R^2 healthy group: 0.50. Sim Kmax, maximum simulated keratometry value.

* Multiple regression analysis performed excluding IOP.

off value (e.g., cutoff value of 1.18 mm for DA provides 82.4% sensitivity but only 61.1% specificity).

Discussion

With the increased focus on the biomechanics of the cornea in recent years,[4–6] the development of new instrumentation that might aid in the diagnosis of ocular disease and the clinical assessment of corneal biomechanical properties is potentially of great interest, but only if the parameters measured are repeatable, accurate, and reflect underlying disease processes. This study has shown that CST exhibits high repeatability in measuring the IOP and CCT, and fair repeatability in DA, and A1T. These factors were shown to be repeatable at different times of day and at the same time of day on different days, thus enabling the use of these parameters in clinical practice.

These results are consistent with those of Hon et al.,[21] which showed high repeatability in IOP, CCT, DA, and A1T when measured at two different times of day. Hong et al.[22] demonstrated that IOP was highly repeatable on three measurements 2 minutes apart. Leung et al.[23] also showed that DA was highly repeatable on two consecutive measurements, whereas, Reznicek et al.[24] demonstrated high repeatability of CCT and IOP (ICC 0.942 and 0.937, respectively) over five measurements on 17 patients. Nemeth et al.[25] reported

Table 8. Comparison of Deformation Amplitude on the Corvis ST in Keratoconic Corneas With CCT Greater Than 460 μm and Central Thickness–Matched Healthy Corneas

	Healthy (n = 18)	KC (n = 17)	*P* Value
Baseline characteristics			
Age, mean ± SD	40 ± 14	29 ± 15	
Sex			
Male	44% (8)	65% (10)	0.23*
Female	56% (10)	35% (6)	
Kmax, ΔD	43.8 ± 1.1	50.3 ± 2.9	<0.001†
CST parameters, mean ± SD			
CCT, μm	505 ± 24	495 ± 27	0.27†
IOP, mm Hg	12.6 ± 1.7	11.5 ± 1.8	0.05†
DA, mm	1.13 ± 0.09	1.25 ± 0.08	0.001†

* χ^2 test.

† Independent *t*-test.

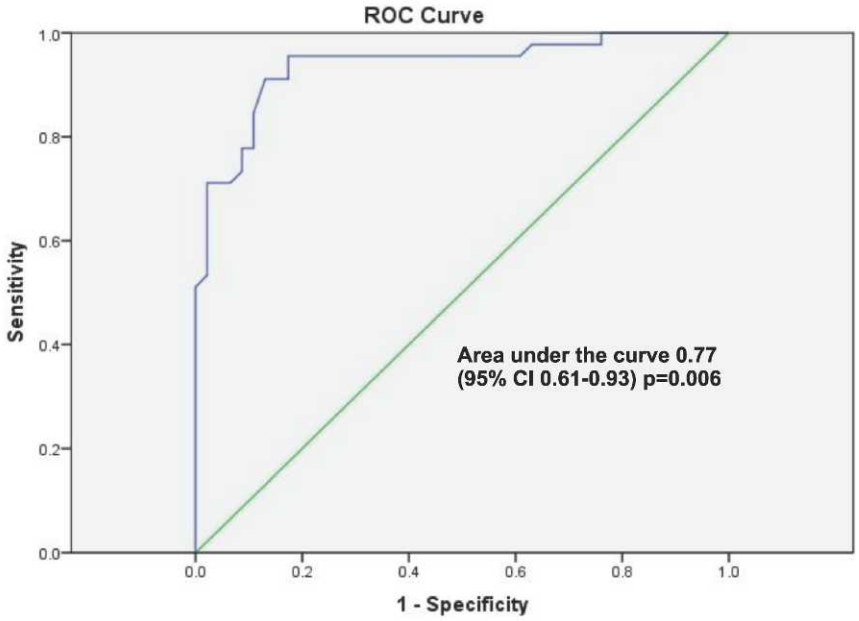
that only IOP and CCT were repeatable on three consecutive measurements, where an ICC value of 0.9 or greater was considered "highly repeatable." Their results were comparable to ours, in that DA and A1T were repeatable by the criteria used in the current study (ICC 0.76 and 0.78, CV 0.04 and 0.02, respectively).

In the current study, DA did not significantly correlate with age in the healthy group (n = 103), consistent with results reported by Nemeth et al.[25] and Valbon et al.[26] Leung et al.[23] found age was a significant predictor of DA in a cohort of healthy, glaucoma suspect, and glaucomatous eyes, which are not directly comparable with our cohort of healthy eyes. Furthermore, age was not a significant predictor of DA in multivariate analysis in both the healthy and KC groups in our study. While there is evidence of age-related ultra-structural corneal and ocular changes,[27–29] these may not necessarily translate to measurable biomechanical differences with age on the CST. We note that a large study of 4184 eyes using the ORA showed a significant relationship of its parameters (corneal hysteresis [CH], and corneal resistance factor [CRF]) with age, however IOP, corneal thickness, and keratometry readings were not included in the multivariate analysis.[30] Furthermore CH and CRF measurement on the ORA relies on different principles than DA on the CST so the association of age with CH and CRF is not directly comparable with DA.

The cornea has been observed to behave in a viscoelastic manner.[1] There is an initial fast deformation in response to a force, followed by a further time-dependent deformation. Energy is dissipated in this process, resulting in a slower rebound.[31] It has long been conjectured that KC corneas are more distensible than healthy corneas. Edmund[32] used positional IOP measurements, corneal thickness, and topographic information to demonstrate that Young's modulus of the cornea in the steady state (or time-dependent part of the viscoelastic response) is decreased in keratoconus. Ex vivo studies using extensiometry techniques have also provided supporting evidence of increased elasticity in KC compared with healthy corneas.[15,16]

Researchers have used the ORA device to investigate and attempt to define the biomechanical properties of the cornea in KC.[33–36] As noted, the ORA provides CRF and CH measurements, the latter being the pressure difference between the two applanation events, postulated to be equivalent to energy dissipation, or 'dampening', as part of the viscoelastic response.[33–36] Although studies have demonstrated statistically significant differences in CH and CRF in KC or subclinical (forme fruste) KC compared with healthy corneas, CH and CRF have been shown to be poorly

A. Receiver-operating characteristic analysis in the identification of keratoconus by Deformation Amplitude on the Corvis ST.



B. Coordinates of the curve showing cut-off values of Deformation Amplitude with sensitivity and 1-specificity

Positive DA if ≥	Sensitivity	1 - Specificity
1.0950	0.889	0.550
1.1050	0.889	0.500
1.1150	0.889	0.400
1.1350	0.889	0.350
1.1600	0.889	0.300
1.1800	0.778	0.300
1.1950	0.722	0.250
1.2050	0.611	0.250
1.2150	0.611	0.200

FIGURE 3. (**A**) Receiver-operating characteristic analysis in the identification of KC by DA on the CST. (**B**) Coordinates of the curve showing cutoff values of DA with sensitivity and 1-specificity.

discriminative between the two states either due to confounding by a lower average CCT in the KC samples or due to the lack of a highly-sensitive and highly-specific cutoff value from an ROC analysis.[6,7,37,38] Furthermore, it has also been suggested that CH and CRF are significantly measurement-dependent, and therefore do not reflect objective biomechanical properties of the cornea.[3]

The current study is the first to extensively analyze corneal biomechanical parameters measured using CST in KC. In this study, the comparison between KC and age-matched healthy corneas demonstrated statistically significant differences in mean DA and A1T. Additionally, IOP was a significant predictor of deformation amplitude and A1T in KC, which is consistent with previous reports of IOP-affecting corneal deformation.[31,39] It is well established that IOP measurements are affected by CCT,[40] and thus when IOP is excluded from the analysis the effect of CCT was significant on DA and A1T. Interestingly, in subsequent versions of CST software a new parameter named the cornea-compensated IOP was added. This parameter was not examined in this study that used software version V2.x, the IOP given by the instrument in this version of the software was not cornea-compensated.

To discern whether there was a genuine difference in deformation amplitude between healthy and KC corneas, we also assessed two subgroups matched for CCT and IOP. This analysis showed that deformation amplitude is statistically greater in the KC group than the healthy group (1.25 ± 0.08 μm and 1.13 ± 0.09 μm, respectively, $P = 0.001$) even when controlling for IOP and CCT variables ($P = 0.05$ and $P = 0.27$, respectively). A receiver operator characteristic curve did not give a highly sensitive and specific cutoff diagnostic value for DA when controlled for IOP and CCT variables, however, this subgroup analysis consisted of modest patient numbers, thus larger studies are required to confirm and further refine these observations.

Alterations in corneal microstructure in KC may explain the difference observed in deformation amplitude in this study. It has been postulated that corneal deformation in response to a uniaxial stress perpendicular to the plane of collagen lamellae, such as the uniaxial air pulse from the CST, consists of compression of the extracellular matrix and sliding of the

collagen lamellae.[31] There is evidence corneal volume loss,[41,42] and collagen structural compromise in KC, namely a distortion of their orthogonal arrangement and distortion of the orientation of single collagen fibrils, as well as a reduction in the number of lamellae.[43,44] These factors may facilitate increased sliding between lamellae perhaps resulting in a greater corneal deformation to a given force.

Furthermore, there is evidence of extracellular matrix degradation such as a reduction corneal volume, alteration of proteoglycan content,[45–47] and reduction in keratocyte density, which have a role in structural support and maintenance of the corneal stroma.[12-14,48] These factors may result in a decreased resistance to deformation and greater mean deformation amplitude compared with healthy corneas, as observed in the current study.

The relationship of DA with CCT and Kmax predicted by linear regression would conform to the hypothesis of lamellar slippage. Furthermore, given that the R^2 value is 0.75 for the combination of IOP, CCT, and Kmax as predictors of DA, 25% of the magnitude of DA is unaccounted for. There may be other undefined anatomical factors contributing to this, however, it may be hypothesized that where CCT, IOP, and Kmax are corrected for, DA may be proportional to, or a surrogate indicator of the corneal elastic modulus. To date, there is no evidence in the published literature suggesting that any factors other than those we have identified are significantly related to deformation amplitude on the CST. A study by Smedowski et al.[49] examined DA in a subgroup of 15 eyes with KC and found it to be significantly related to IOP. Axial length has been examined by Nemeth et al.[25] and Leung et al.,[26] and was found to not have a significant relationship with DA in healthy eyes, the latter study also found spherical error was not significant. With regards to the ORA, axial length has been shown to be a significant predictor of CH and CRF.[30] We did not examine axial length in our patients thus further studies are needed to investigate whether it is predictive of DA on the CST between healthy and keratoconic patients specifically.

Notably the time to applanation 1 is shorter in KC than healthy corneas. The A1T in KC was accounted for by IOP, CCT, Kmax (R^2 value 0.98), properties, which are readily measurable. Consequently, while A1T can be used as an indicator of KC, it is difficult to determine whether the significant differences in A1T provide any additional information in the identification of KC beyond its high correlation with corneal thickness, IOP, and Kmax.

Theoretically, the time to second applanation (A2T) may be an indicator of the total viscoelasticity of the cornea as it marks the end of corneal deformation and represents the time-dependent element of the viscoelastic response. This parameter was not repeatable on successive measurement or at different times of day. While this may be explained by diurnal variation in IOP or corneal hydration, further conclusions about the use of this parameter cannot be formulated at present.

In conclusion, the CST is a new instrument, which visualizes corneal deformation using ultra–high speed Scheimpflug technology and enables calculation of biomechanical responses. Intraocular pressure and CCT are highly repeatable; while DA and A1T are fairly repeatable as single measurements and highly repeatable when the average of three consecutive measurements is taken. Deformation amplitude is significantly greater in KC than healthy eyes, even when controlled for IOP, and this cannot be fully accounted for by the current factors used to describe KC including CCT and Kmax. Deformation amplitude may be indicative of the corneal elastic modulus in established KC, and thus could be a useful adjunct in the clinical assessment and monitoring of KC as well as assessment of treatments such as corneal collagen cross-linking. At present, DA measured by the CST is not sufficiently discriminative to differentiate healthy and keratoconic corneas in individual eyes. Further studies are needed to confirm these observations and to delineate the use of the CST in differentiating mild KC from healthy corneas.

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