

Comparison of non-invasive tear break-up time and tear meniscus height in healthy eyes and keratoconus using Oculus Keratograph 5M

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Received: 2016-11-21 Accepted: 2017-06-22

使用 Oculus Keratograph 5M 测量圆锥角膜患者 NITBUT 和 TMH

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摘要

目的:比较圆锥角膜患者和正常人使用 Oculus Keratograph 5M(K5M)测得的非侵入性泪膜破裂时间(NITBUT)和泪河高度(TMh),以确定这两项指标和圆锥角膜的关系。

方法:50例100眼圆锥角膜患者和50例100眼正常人参加了这项研究。圆锥角膜组患者的年龄为15~60(平均28.33±8.60)y。对照组为18~60(平均26.25±1.11)岁。使用K5M测量所有受试者的NITBUT和TMH。

结果:两组之间的NITBUT和TMH平均值差异无统计学意义($P=0.58,0.69$)。两参数的相关性研究显示,两组中NITBUT和TMH之间均无相关性(圆锥角膜组: $r=0.053, P=0.721$;对照组: $r=-0.0501, P=0.7098$)。

结论:研究显示圆锥角膜对泪液质量和数量都没有显著的临床影响。

关键词:圆锥角膜;非侵入性泪膜破裂时间;泪河高度;Oculus Keratography 5M

引用:Safarzadeh M, Azizzadeh P, Akbarshahi P, Heidari L. 使用 Oculus Keratograph 5M 测量圆锥角膜患者 NITBUT 和 TMH. 国际眼科杂志 2017;17(9):1625-1630

Abstract

• **AIM:** To compare the non-invasive tear break-up time (NITBUT) and tear meniscus height (TMH) measurements in keratoconus patients and normal subjects, and to determine the relationship between these measurements with keratoconus disease by the Oculus Keratograph 5M (K5M).

• **METHODS:** Fifty keratoconus patients (100 eyes) and 50 healthy subjects (100 eyes) participated in the study. The age range in keratoconus group was 15-60 (mean ± standard deviation = 28.33 ± 8.60) y, and in control group was 18-60 (26.25 ± 1.11) y. The measurements of NITBUT and TMH were performed using the K5M.

• **RESULTS:** The mean value of NITBUT between the keratoconus group and the control group showed no statistically significant difference ($P=0.58$). Also, the mean of TMH between two groups was not significantly different ($P=0.69$). The results of correlation coefficient between the variables of the study demonstrated that there was no significant relationship between the NITBUT and TMH measurements with the two groups (keratoconus group: $r=0.053, P=0.721$; control group; $r=-0.0501, P=0.7098$).

• **CONCLUSION:** Our study shows that the presence of keratoconus has no clinically significant impact on the quality and quantity of tear film.

• **KEYWORDS:** keratoconus; non-invasive tear break-up time; tear meniscus height; Oculus Keratography 5M

DOI:10.3980/j.issn.1672-5123.2017.9.05

Citation: Safarzadeh M, Azizzadeh P, Akbarshahi P, Heidari L. Comparison of non-invasive tear break-up time and tear meniscus height in healthy eyes and keratoconus using Oculus Keratograph 5M. *Guoji Yanke Zazhi(Int Eye Sci)* 2017;17(9):1625-1630

INTRODUCTION

Keratoconus is a non-inflammatory disorder of the cornea, that is characterized by progressive conical ectasia with a protrusion of the thinned stroma area^[1-2]. The typical manifestations are inferior corneal thinning and protrusion with increasing myopia and irregular astigmatism that leads to mild or marked visual impairment^[1-4]. The etiology of the disease is unknown, but there are evidences of genetic inheritance and possible linkage with systemic disease that may be associated with the disease^[4-6]. The incidence and prevalence of keratoconus are 50–230 and 54.4 per 100000 in the general population, respectively^[1,4]. In mild cases of keratoconus or in the early stages of this condition, spectacle lenses or the use of soft contact lenses may be indicated. As the severity of keratoconus increases, due to the progression of the cone along with alterations of the corneal tissue and escalation of astigmatism, rigid gas permeable contact lenses may be an effective method^[1-2,4]. In the keratoconus, all layers except the endothelium have been shown to have histopathological structural changes^[2-3,5]. Dry eye symptoms, such as lowering in tear secretion and tear film break up time has been reported in keratoconus patients^[5,7-9]. The pathophysiology and mechanisms behind the development of keratoconus are not entirely understood, however, oxidative stress is thought to be one of the contributing factors in the pathogenesis of this corneal ectasia^[3]. The human tear film is a very thin layer of fluid that being composed of three components that work together: a complex mucus component, a watery portion (Aqueous) and a complex oil outer layer (Lipid)^[5,10]. Normal tear volume is critically important for the maintenance of ocular surface physiology and ocular comfort^[10-11]. The volume of aqueous tears contained within the upper and lower tear meniscus is approximately 75–90 percent of the total volume of the aqueous component^[12]. Therefore a reasonable assessment of the tear volume can be made by observing the height and width of this tear meniscus. An increased tear meniscus height (TMH) indicates poor tear drainage due to an obstructed punctum or an excessive aqueous layer giving a watery tear film. On the other hand a reduced TMH suggests a reduced tear volume^[12-13]. The TMH test is classified as follows: good >0.2mm, normal=0.2mm, poor <0.2mm^[13]. Non-invasive tear break-up time (NITBUT) involves the use of a grid pattern, Purkinje image I, or keratometer mires projected onto the corneal surface^[14]. In the NITBUT test, the time period between the last complete blink and the first perturbation of a grid projected onto the surface of the cornea in the normal subjects is >10s^[15-16]. Recent advances in new technologies have enabled us to non-invasively evaluate the quantity and quality of the tear film. The Oculus Keratograph 5M (manufactured by Oculus Optikgeraete GmbH, Wetzlar, Germany) is an advanced corneal topographer with a built-in real keratometer and a

color camera optimized for external imaging. Tear film volume (tear meniscus height) and the tear film stability (non-invasive tear break-up time) can easily be assessed with both white and infrared light by the K5M^[17-18]. To our knowledge, the qualitative and quantitative parameters of tear film in keratoconus patients have not been extensively investigated. Therefore, the results of this study can increase our understanding of the alterations of tear film in keratoconus patients, and also improve the management of the disease.

METHODS

Study Population This was a comparative, prospective, and single-masked study. Two hundred eyes of 100 patients were included in the study, and classified into two groups: healthy eyes ($n = 100$) and keratoconus eyes ($n = 100$). Keratoconus group was consisted of 30 females and 20 males in the age range of 15–60 (mean \pm standard deviation = 28.33 \pm 8.60)y. Healthy eyes as a control group was made up of predominantly females in the age range of 18–60 (mean \pm standard deviation = 26.25 \pm 1.11) y. Keratoconus was confirmed with the Scheimpflug topographical analysis and biomicroscopy examination by corneal specialists. The keratoconus stages were identified using the Amsler-Krumeich classification (Table 1)^[19]. Research team was blinded to the results of the eye examination conducted before the diagnosis of keratoconus. Patients with any active ocular surface disease (e.g. significant dry eye symptoms or keratitis), corneal opacities, pellucid marginal corneal degeneration, corneal astigmatism greater of 2.00 diopters (D) (except in the keratoconus group), glaucoma, using medications that could affect tear production, and a history of any type of ocular surgery were excluded. Keratoconus patients in stage IV of the disease, according to the Amsler-Krumeich classification were excluded from the study. All data from the two groups in the contact lens clinic at the private hospital (Bahman Hospital, Tehran, Iran) were obtained. The measurements were taken between 9:00 AM to 3:00 PM in a dimly lit room, where the temperature (20–25°C) and humidity (30–40%) were controlled. No eye drops were applied before the measurements. All participants signed an informed consent form in accordance with the tenets of the Declaration of Helsinki.

Non-Invasive Tear Break-Up Time The non-invasive tear break-up time measurements were performed using the Oculus Keratograph 5M (K5M) on both eyes of keratoconus patients and control subjects. The K5M measures the NITBUT by detecting localized breaks in the tear film using infrared waves. After two blinks to reconstitute the tear film, the participants were asked to refrain from blinking and fixate on the central light source. A video recording of the ocular surface begins with real-time detection and localization of breaks in the tear film. During assessment, 22 rings are projected onto the cornea, with more than 1,000 measurement

points per ring, resulting in 22,000 analyzed data points per frame. Points of break – up appear on a grid mapping the corneal surface. The video recording lasts up to a maximum of 25s, or until the patient's next blink. Two readings are provided at the end of every assessment: NITBUT–First, the time taken for the first appearance of a break in the tear film, is the parameter of interest in this study. The second reading produced by K5M, called the NITBUT–Average, that is the average of the time taken to break – up in all the regions monitored over the duration of the 25s.

Tear Meniscus Height (TMH) The values of TMH were obtained on both eyes of subjects in the two groups using the Oculus Keratograph 5M. Fixation targets in the K5M were an orange ring with a black center. The subjects were asked to look directly into the center of the orange ring. Then, the TMH setting was selected, and the tear meniscus height could be measured. In the K5M for measuring TMH there is not the projection of LED lights. Instead, an infrared camera is used in order to taking photograph of TMH. Due to the position of the instrument, in order to focus the lower lid margin, the subjects were asked to fixate on the top half of the orange circle, as part of the target that was out of sight. Once the tear meniscus could be imaged clearly in the center of the screen, images were obtained by manually pressing the capture button on the K5M. Both eyes of subjects were examined three times at 5–min intervals in order to calculate an average of the TMH values. After the completion of the photographs using the K5M, each of photographs were magnified twice in its original size in order to better view the TMH. The value of TMH was in millimeters and was converted to microns. In an attempt to ensure the consistency of the measurements, the same optometrist was responsible for the measurements of scans.

Statistical Analyses Check the normality of the data was performed on the measurement results of NITBUT and TMH in the keratoconus and control groups. The Kolmogorov–Smirnov and the Shapiro – Wilk tests for normality of data were selected. In cases that the data were not normally distributed from non – parametric tests were used. Differences between groups (keratoconus and control) were compared using the Mann – Whitney *u* – test. Also, the correlation between NITBUT and TMH in the two groups was determined using nonparametric Spearman analysis. The data were analyzed using SPSS 19. Statistical significance was assessed at 0.05 probability levels, and $P < 0.05$ was considered significant.

RESULTS

Fifty keratoconus patients (100 eyes) and 50 control subjects (100 eyes) were involved in the study. The mean age of keratoconus patients and control subjects was 28.33 ± 8.60 y and 26.25 ± 1.11 y, respectively. According to the values obtained from the normality tests (Table 2), only the TMH for the control group was normally distributed ($P = 0.17$).

Table 1 Amsler–Krumreich classification for grading keratoconus

Stages	Classification methods
Stage I	Eccentric steepening Myopia and/or astigmatism < 5.00 D Mean central K readings < 48.00 D Vogt striae, no corneal opacities
Stage II	Myopia and/or astigmatism from 5.00 to 8.00D Mean central K readings < 53.00 D Absence of scarring Minimum corneal thickness $\geq 400 \mu\text{m}$
Stage III	Myopia and/or astigmatism from 8.00 to 12.00D Mean central K readings > 53.00 D Absence of scarring Minimum corneal thickness from 200 to $400 \mu\text{m}$
Stage IV	Refraction not measurable Mean central K readings > 55.00 D Central corneal scarring Minimum corneal thickness $< 200 \mu\text{m}$

K: Keratometry; D: Diopters.

Table 2 Normality tests of NITBUT and TMH for keratoconus and control groups

Parameters	Kolmogorov–Smirnov		Shapiro–Wilk	
	Statistic	<i>P</i>	Statistic	<i>P</i>
NITBUT				
Keratoconus	0.176	0.014	0.703	0.001
Control	0.119	0.024	0.824	0.002
TMH				
Keratoconus	0.148	0.002	0.840	0.001
Control	0.101	0.17 ^a	0.850	0.031

^a: A normal distribution; NITBUT: Non – invasive tear break – up time; TMH: Tear meniscus height.

Table 3 Demographic of the NITBUT and TMH measurements in keratoconus and control groups

Descriptive statistics	NITBUT (s)		TMH (μm)	
	Keratoconus	Control	Keratoconus	Control
Mean \pm standard deviation	8.9 ± 4.5	9.2 ± 3.5	263.18 ± 93.97	246.82 ± 41.52
Range	0.1–21.9	0.1–14.5	134–570	168–376
Median	4.7	5.9	234	238
<i>Z</i> ^a	–0.466		–0.283	
<i>P</i>	0.58		0.69	

NITBUT: Non – invasive tear break – up time; TMH: Tear Meniscus Height; ^a Mann–Whitney *u* – test.

Demographic data of the tear film tests in the two groups presented in Table 3. The mean of NITBUT in keratoconus patients was 8.9 ± 4.5 s and in control subjects was 9.2 ± 3.5 s. Statistical analysis using Mann – Whitney *u* – test showed no significant difference for the NITBUT between the two groups ($P = 0.58$) (Table 3). The mean of TMH for the keratoconus and control groups was $263.18 \pm 93.97 \mu\text{m}$ and $246.82 \pm 41.52 \mu\text{m}$, respectively, that found no statistically significant difference between them ($P = 0.69$) (Table 3). To visualize the distribution of data between the two groups, the boxes and whisker plots were used (Figure 1 and 2). As shown in

Figure 1, the minimum values of NITBUT for both the two groups were obtained 0.1s, but the maximum value for the keratoconus group was obtained 21.9s, that 7.4s were more than the control group. Also, the median of NITBUT (gray line in the center each of the boxes) for the keratoconus and control groups was 4.7s and 5.9s, respectively, that there were no statistically significant differences between them ($P>0.05$). According to the boxes and whisker plots shown in Figure 2, the difference between the median value of TMH in the two groups was demonstrated $4\mu\text{m}$. In addition, the difference between the maximum and minimum values of TMH in the two groups was obtained 194 and $34\mu\text{m}$, respectively, that was not statistically significant ($P>0.05$). The values obtained from Spearman's correlation demonstrated that there was no significant correlation between the NITBUT and TMH measurements with the keratoconus ($r=0.053$, $P=0.721$) and control eyes ($r=-0.0501$, $P=0.7098$) (Table 4).

DISCUSSION

The etiology of keratoconus has been investigated frequently, but is still largely unknown. Description the qualitative and quantitative changes of tear film may improve the management of keratoconus disease. Given the intrinsic invasiveness of conventional tear break-up time (TBUT) and its effects on the reliability of the measurements, several the non-invasive tear break-up time (NITBUT) tests have been developed. Their advantage is that they do not alter the ocular surface, which means a reduction in irritation and reflex tearing^[7]. In a study of Tian *et al*^[18], found that the repeatability and reproducibility of the tear meniscus height (TMH) and NITBUT measurements using the K5M in patients with dry eye disease (DED) is good. In a study of Best *et al*^[17], found that Keratograph NITBUT measurements are significantly lower than observation using the Tearscope.

In the current study, the mean of NITBUT for the keratoconus and control groups was $8.9 \pm 4.5\text{s}$ and $9.2 \pm 3.5\text{s}$, respectively, and there was no statistically significant difference in the two groups ($P=0.58$). In previous studies it were stated that the values of NITBUT in keratoconus group reduced than healthy subjects, may be due to steepening of the cornea, change in the quality of the mucin secretion by the diseased corneal epithelium, reducing the number of goblet cell, or change in the conjunctiva non-goblet epithelial cell^[8,20-21]. Dogru *et al*^[8], reported that the values of TBUT and NITBUT are lower in keratoconus patients compared to the normal subjects, but they found no aqueous-deficient dry eye among keratoconus patients.

In the current study, the value of NITBUT in healthy eyes with the use of Oculus Keratograph 5M was obtained less than 10s, because the Keratograph measures the first time of the tear break-up anywhere on the cornea regardless of how small or transient the area of break-up. Therefore, the K5M software detects very early tear film changes, recording significantly lower NITBUT values than conventional subjective

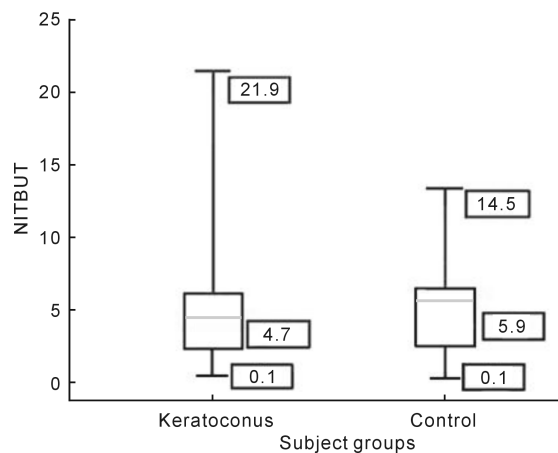


Figure 1 Box and whisker plot of non-invasive tear break-up time for the keratoconus and control groups in seconds.

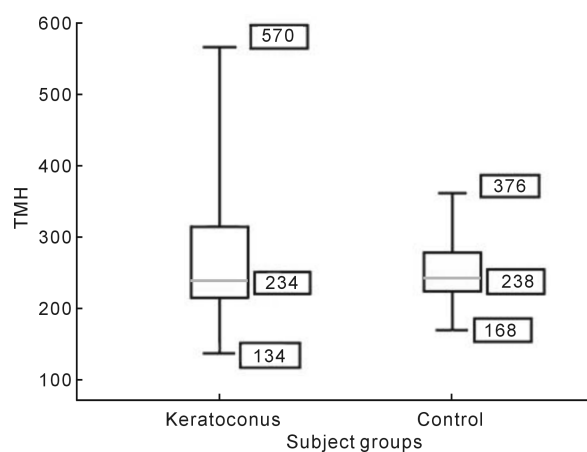


Figure 2 Box and whisker plot of tear meniscus height for the keratoconus and control groups in micrometers.

Table 4 Spearman's rank correlation between NITBUT and TMH in groups

Spearman's rank correlation	Keratoconus	Control
Correlation coefficient (r)	0.053	-0.0501
P	0.721	0.7098

NITBUT; Non-invasive tear break-up time; TMH; Tear meniscus height.

assessment. Adjustments to instrumentation software have the potential to enhance the value of Keratograph objective measures in clinical practice.

The TMH is indicative of tear volume and may be used as a diagnostic criterion for dry eye disease^[8]. In previous studies found that the TMH values obtained using optical coherence tomography are significantly lower in patients with dry eyes than healthy eyes^[9,22-23]. In Lei *et al*'s^[18] study, the mean of TMH was $0.22 \pm 0.07\text{mm}$ for DED group and $0.27 \pm 0.12\text{mm}$ for healthy group. Correspondingly, Hong *et al*^[24], had compared the value of TMH in dry eye patients and healthy subjects using Keratograph 4. They reported that the value of TMH is lower for dry eyes than control eyes (0.269 ± 0.011 versus $0.379 \pm 0.015\text{mm}$, respectively).

In another study of Sarac *et al*^[5], the tear meniscus height

was measured using optical coherence tomography (average of TMH in keratoconus eyes: $250.77 \pm 66.47 \mu\text{m}$ and control eyes: $233.00 \pm 76.99 \mu\text{m}$), that despite higher TMH in keratoconus group, found no significant difference between the keratoconus and control groups. They thought that the severity of keratoconus, type and location of conus are the reasons for higher TMH in keratoconus eyes than healthy subjects.

Koh *et al*^[25], reported that the TMH values using K5M are 0.14 ± 0.03 and $0.20 \pm 0.05 \text{mm}$ in patients with DED and healthy subjects, respectively. In a study of Nguyen *et al*^[26], the correlation between the clinical parameters of dry eye disease and the TMH values was compared. They concluded that the TMH measurements have significant correlation with the results of Schirmer's test, but they found no correlation between the NITBUT measurements with dry eye disease. In a study of Golding *et al*^[27], the TMH and NITBUT measurements in dry eye patients were evaluated, and found that there is a positive linear correlation between them in dry eye group ($P < 0.05$).

In our study, we concluded that there is no statistically significant correlation between the NITBUT and TMH measurements with keratoconus ($r = 0.053$, $P = 0.721$). Also, according to the negative correlation presented between the non-invasive tear break up time and tear meniscus height measurements in control group ($r = -0.0501$), demonstrated that there is no significant relationship between them ($P = 0.7098$).

Also, in the current study, the mean of TMH for the keratoconus group was a little greater than control group (keratoconus group: 263.18 ± 93.97 and control group: 246.82 ± 41.52). It has been thought that higher TMH in the keratoconus patients than normal subjects may be due to effect compensatory tear film production, the discrepancy in the severity distribution of the keratoconus between subjects, and the type of software used in Oculus Keratograph. For this reason, we have investigated the correlations between TMH in each of the two groups of keratoconus and control.

Finally, based on the results obtained from Spearman's rank correlation (Table 4) demonstrated that keratoconus has not significant effect on the TMH measurement in the keratoconus eyes compared with healthy eyes. Some studies had expressed that age can be an important risk factor for DED, and shown that the TMH values vary with age^[23,28]. In accordance with our study, Lei *et al*^[18] showed that with age, values of TMH do not change, so it is not the reason for the differences observed in the TMH values between groups.

To our knowledge, this is the first study for comparing the same time both the NITBUT and TMH measurements to determine the correlation between the two tests scores in keratoconus eyes and healthy eye using the Oculus Keratograph 5M.

The results of qualitative and quantitative evaluation of the

tear film using the Oculus Keratograph 5M showed that the keratoconus has no significant effect on the non-invasive tear break-up time and tear meniscus height measurements.

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